

**MORPHO-MOLECULAR CHARACTERISATION OF  
INTERGENERIC HYBRIDS OF *Ascocentrum***

*by*

**KATARE RENUKA SHAMRAO**

**(2014-22-106)**



**DEPARTMENT OF FLORICULTURE AND LANDSCAPING  
COLLEGE OF HORTICULTURE  
VELLANIKKARA, THRISSUR – 680 656  
KERALA, INDIA  
2019**

**MORPHO-MOLECULAR CHARACTERISATION OF  
INTERGENERIC HYBRIDS OF *Ascozentrum***

*by*  
**KATARE RENUKA SHAMRAO**  
**(2014-22-106)**

**THESIS**  
**Submitted in partial fulfilment of the**  
**requirements for the degree of**

**Doctor of Philosophy in Horticulture**

Faculty of Agriculture  
**Kerala Agricultural University**



**DEPARTMENT OF FLORICULTURE AND LANDSCAPING**  
**COLLEGE OF HORTICULTURE**  
**VELLANIKKARA, THRISSUR – 680 656**  
**KERALA, INDIA**  
**2019**



Affectionately .Dedicated to  
Beloved Mother



## **DECLARATION**

I, hereby declare that the thesis entitled “**Morpho-molecular characterisation of intergeneric hybrids of *Ascocentrum***” is a bonafide record of research work done by me during the course of research and the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other university or society.

Vellanikkara,

Date: 02/11/2019

**Katare Renuka Shamrao**

(2014-22-106)

Dr. A. Sobhana  
Professor and Head,  
Fruit crops Research Station,  
Kerala Agricultural University,  
Vellanikkara, Thrissur, Kerala

Date: 2/11/2019

### **CERTIFICATE**

Certified that this thesis entitled “**Morpho-molecular characterisation of intergeneric hybrids of *Ascocentrum***” is a record of research work done independently by **Ms. Katare Renuka Shamrao (2014-22-106)**, under my guidance and supervision and that it has not previously formed the basis for the award of any degree, diploma, associateship or fellowship to her.

**Dr. A. Sobhana**  
Chairperson,  
Advisory Committee

## **CERTIFICATE**

We, the undersigned members of the advisory committee of **Ms. Katare Renuka Shamrao (2014-22-106)** a candidate for the degree of **Doctor of Philosophy in Horticulture**, with major field in **Pomology and Floriculture**, agree that the thesis entitled “**Morpho-molecular characterisation of intergeneric hybrids of *Ascocentrum***” may be submitted by **Ms. Katare Renuka Shamrao (2014-22-106)**, in partial fulfilment of the requirement for the degree.

**Dr. A. Sobhana**

(Chairperson, Advisory Committee)

Professor and Head,

Fruit crops Research Station,

Kerala Agricultural University, Vellanikkara

**Dr. U. Sreelatha**

(Member, Advisory Committee)

Professor and Head,

Department of Floriculture and Landscaping,

College of Horticulture, Vellanikkara

**Dr. Mini Sankar**

(Member, Advisory Committee)

Assistant professor,

AICRP on Floriculture Improvement,

Department of Floriculture & Landscaping,

College of Horticulture, Vellanikkara

**Dr. P. S. Abida**

(Member, Advisory Committee)

Professor and Head,

Division of Crop Improvement,

RARS, KAU, Pattambi, Palakkad

**Dr. S. Krishnan**

(Member, Advisory Committee)

Professor and Head, (Retd.)

Department of Agricultural Statistics,

College of Horticulture, Vellanikkara

**Dr. S. Praneetha**

(External Examiner)

Professor, (Hort.)

Horticultural Research Station,

Yercaud, 636 602, Salem (District), T.N.

## *ACKNOWLEDGEMENT*



*“Om Namō Bhagwate Varsudevaya” I bow to the lotus feet of Lord Vasudeva whose grace had endowed me the inner strength and confidence and blessed me at each step during my long sojourn at Kerala. I am at that golden moment of my life where my heart is overwhelmed with gratitude and I wish if these words could convey my subtle feelings.*

*I am ineffable in expressing my deepest sense of gratitude to my supervisor **Dr. A. Sobhana**, Professor and Head, Fruit crops Research Station, KAU, Vellanikkara and Chairperson of my advisory committee for her for their excellent supervision, consistent inspiration, and moral support throughout the course of my research work,*

*I feel immense pleasure to express my deep sense of gratitude to the members of my Advisory Committee **Dr. U. Sreelatha**, **Dr. P. S. Abida**, **Dr. Mini Sankar** and **Dr. S. Krishnan** for their inspiring guidance, keen interest and timely support throughout the period of investigation.*

*It is my proud privilege to place my sincere thanks to **Dr. C. George Thomas**, Former Associate Dean, College of Horticulture, KAU, Vellanikkara for being my guiding light, for having belief in me, timely support and encouragement.*

*I would like to express my heartfelt gratitude to **Dr. Jiji Joseph** and **Dr. Rose Mary Francies** for the crucial support and the facilities provided in the molecular laboratory for the course of my research work and wholehearted co-operation.*

*I wish to express my profound sense of gratitude to Librarian **Dr. A. T. Francis** and office staff of the institute **Smt. Shakila Begum** for constant encouragement and timely*

support. I also owe my warm thanks to all the non-teaching and labour staff of institute *Lucy matron, Sindhu matron, Rajani Chechi and Najeeba Chechi* for love and care.

Today, here is smile on my lips and the light of hope in my eyes, because my fortunate has changed for better. Behind this picture of propriety lies the hard work, bondless love and firmless sacrifice of my loving Mother *Sulochana*, father *Shamrao*, elder sister *Padmaja* and twin brother *Sushant* who supported me throughout my education, keeping in all comforts without which this work could not have seen the light of this day. I am also lucky enough to have such loving, caring and helpful relatives and family friends *Ganga, Manik, Yamuna, Nanda, Shifa, Varsha, Sangita, Mira, Suvarna, Nivrutti, Vishnu, Rushi, Gau, Rutu, Sush, Shub, Mayu, Harshu, Kittu, Kishu, and Shri*.

Ultimately the bond of all companionship is conversation. I owe my deep thanks to my best ones with whom I share my feelings, experiences and my deepest thoughts with *Priya, Ashu, Rina, Kshitija, Mukul, Sudhir, Sumit, Akki, Shanky and Suraj*. I owe my special thanks to my closest one *Ajinkya* not only for his support in difficulties but also for kind co-operation and technical guidance throughout the thesis completion and timely helped during my academic attainments. It is not so our friend's help that help us, as the confidence of their help. I also owe my warm thanks to my special ones *Divya, Midhun, Mohit, Radhu, Sri, Annjoe, Jeevan, Vipul, Deepak, Jessabelle and Nayra*.

I would like to express my profound gratitude to Ministry of Science and Technology, Government of India for providing me the **DST INSPIRE Fellowship** which was an enormous financial help throughout the course of my degree programme. At the end, I owe my un-expressible gratitude to the Almighty and **all those who have been forgotten due to the shortcomings**.

**Place:** *Vellanikkara*

**Date:** *02/11/2019*

*Katara Renuka Shamrao*



## CONTENTS

<b>Chapter</b>	<b>Title</b>	<b>Page No</b>
<b>1</b>	<b>INTRODUCTION</b>	1-4
<b>2</b>	<b>REVIEW OF LITERATURE</b>	5-29
<b>3</b>	<b>MATERIALS AND METHODS</b>	30-59
<b>4</b>	<b>RESULTS</b>	60-142
<b>5</b>	<b>DISCUSSION</b>	143-174
<b>6</b>	<b>SUMMARY</b>	175-182
<b>7</b>	<b>REFERENCES</b>	i-xxiii
<b>8</b>	<b>APPENDICES</b>	I-LV
<b>9</b>	<b>ABSTRACT</b>	LVI-LVIII

## LIST OF TABLES

Sl. No.	Title	Page No.
1	Classification of hybrids of <i>Ascocentrum</i> used for the study	32
2	List of intergeneric hybrids/varieties used for the study	33
3	List of parent hybrids selected for compatibility studies	44
4	Cross combinations using selected two male parents	47
5	Intergeneric hybrids of <i>Ascocentrum</i> selected for molecular characterization	49
6	List of SSR primers (with their forward and reverse sequences) used for the study	50
7	List of ISSR primers used for the study	51
8	Quantitative plant, shoot and root characters of intergeneric hybrids of <i>Ascocentrum</i> during March 2018	61
9	Quantitative morphological leaf characters of intergeneric hybrids of <i>Ascocentrum</i> during March 2017 and 2018	67
10	Floral characters of <i>Ascocentrum</i> hybrids/varieties	75-76
11	Members of different clusters in <i>Ascocentrum</i> hybrids	78
12	Inter cluster distance between 12 clusters in <i>Ascocentrum</i> hybrids/varieties	79
13	Mean values of floral characters for clusters in <i>Ascocentrum</i> hybrids/varieties	79
14	Phenological leaf and floral characters of <i>Ascocentrum</i> hybrids/varieties	81
15	Post-harvest floral characters of <i>Ascocentrum</i> hybrids/varieties	84-85
16	Qualitative shoot characters of <i>Ascocentrum</i> hybrids/varieties	87
17	Qualitative leaf characters of <i>Ascocentrum</i> hybrids/varieties	89-90
18	Qualitative root characters of <i>Ascocentrum</i> hybrids/ varieties	91
19	Qualitative flowering/spike characters of <i>Ascocentrum</i> hybrids/ varieties	93-94

<b>Sl. No.</b>	<b>Title</b>	<b>Page No.</b>
20	Qualitative floret characters of <i>Ascozentrum</i> hybrids/ varieties	97-98
21	Qualitative petal characters of <i>Ascozentrum</i> hybrids/varieties	100-101
22	Qualitative petal characters of <i>Ascozentrum</i> hybrids/varieties	102-103
23	Qualitative lip characters of <i>Ascozentrum</i> hybrids/varieties	105-106
24	Qualitative lip characters of <i>Ascozentrum</i> hybrids/varieties	108-109
25	Qualitative column and spur characters of <i>Ascozentrum</i> hybrids/varieties	111
26	Visual scoring for the spikes of <i>Ascozentrum</i> hybrids/varieties	113
27	Plant quality rating of <i>Ascozentrum</i> hybrids/varieties	114
28	Incidence of pests and diseases throughout study period	116
29	Anthesis time and stigma receptivity period in the parents	118
30	Pollen studies in eleven selected hybrids/varieties	120
31	Self-compatibility and post pollination changes in the selected parents	123-124
32	Cross compatibility and post pollination changes by using <i>Ascda</i> . Sirichi Fragrance as the male parent	128-129
33	Cross compatibility and post pollination changes by using <i>Mok</i> . Sayan × <i>Ascda</i> . Doung Porn as the male parent	130-131
34	Relationship matrix of compatibility studies	133
35	Analysis of DNA determined by Nanodrop spectrophotometer in different varieties	134
36	Particulars of polymorphic SSR markers	136
37	Pair wise similarity between varieties based on SSR data	137
38	Clustering based on SSR scoring	138
39	Particulars of polymorphic ISSR markers	140
40	Pair wise similarity between varieties based on ISSR data	141
41	Clustering based on ISSR scoring	142

## LIST OF PLATES

Sl. No.	Title	Between Page No.
1	Selected intergeneric hybrids in vegetative phase for field evaluation: (A) Selected hanging orchids in vegetative phase, (B) Prostrate nature of growth in selected orchids	32-33
2	Flowering phase of intergeneric hybrids of <i>Ascocentrum</i> : (A) Selected hanging orchids in flowering phase, (B) Selected prostrate orchids in flowering phase	73-74
3	Intergeneric hybrids of <i>Ascocenda</i>	73-74
4	Intergeneric hybrids of <i>Vascostylis</i>	73-74
5	Intergeneric hybrids of <i>Mokara</i>	73-74
6	Intergeneric hybrids of <i>Mokara</i>	73-74
7	Intergeneric hybrids of <i>Kagawara</i>	73-74
8	Floral parts of a typical monopodial orchid ( <i>Kag. Youthong Beauty</i> )	74-75
9	Micro graphs of pollen count using haemocytometer and viability under low and high power microscope: (A) Pollen count- <i>Ascda. Kultana</i> × <i>V. Bitz's Heartthrob</i> and <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i> , (B) Pollen count- <i>Ascda. Sirichi Fragrance</i> , (C) Viable pollens and sterile pollens, (D) Pollen viability of <i>Ascda. Sirichi Fragrance</i> under 10x and 40x lenses, (E) Pollen viability of <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i> under 10x and 40x lenses	119-120

<b>Sl. No.</b>	<b>Title</b>	<b>Between Page No.</b>
10	Pollen viability and germination in selected male parents under low and high power microscope: (A) Pollen viability of <i>Ascda</i> . Sirichi Fragrance under 10x and 40x lenses and germination under 10x lens, (B) Pollen viability of <i>Mok</i> . Sayan × <i>Ascda</i> . DOUNG PORN under 10x and 40x lenses and germination under 10x lens	119-120
11	Enlargement of pedicel after pollination in <i>Ascocentrum</i> hybrids: (A) <i>Vasco</i> . Aroonsri Beauty, (B) <i>Kag</i> . Youthong Beauty, (C) <i>Ascda</i> . Udomochai, (D) <i>Mok</i> . Chao Praya Sunset Yellow Spot, (E) <i>Vasco</i> . Pine Rivers Fuchsia Delight, (F) <i>Vasco</i> . Pine River Blue	120-121
12	Post pollination changes in different hybrids/varieties: (A) Flower fall, (B) Pods of <i>Vasco</i> . Pine River Pink turned pinkish, (C) Pods turned yellow and shrivelled in <i>Ascda</i> . Sirichi Fragrance, (D) Swelling of pod in <i>Mok</i> . Sayan × <i>Ascda</i> . DOUNG PORN, (E) Developed pods shrivelled later in <i>Vasco</i> . Aroonsri Beauty, (F) <i>Mok</i> . Chao Praya Sunset Yellow Spot, (G) <i>Ascda</i> . Kultana × <i>V</i> . Bitz's Heartthrob	125-126
13	Successfully matured pods in different hybrids: (A) <i>Vasco</i> . Pine River Pink, (B) <i>Mok</i> . Chao Praya Sunset Yellow Spot, (C) <i>Vasco</i> . Pine River Blue, (D) <i>Ascda</i> . Sirichi Fragrance	126-127
14	Different stages of germination to seedling formation: (A) Harvested pod, (B) Seed germination, (C) Protocorms showing declined growth, (D) Leaf and root initiation, (E) Multiplication stage, (F) Fully grown seedlings	127-128
15	Agarose gel electrophoresis of isolated DNA with selected 20 intergeneric hybrids	134-135

<b>Sl. No.</b>	<b>Title</b>	<b>Between Page No.</b>
16	Amplification pattern generated by DQ494847 and DQ501383 with 20 hybrids of <i>Ascocentrum</i>	135-136
17	Amplification pattern generated by DQ501384 and DQ501387 with 20 hybrids of <i>Ascocentrum</i>	135-136
18	Amplification pattern generated by FJ539050 and FJ539054 with 20 hybrids of <i>Ascocentrum</i>	135-136
19	Amplification pattern generated by FJ539057 and FJ539061 with 20 hybrids of <i>Ascocentrum</i>	135-136
20	Amplification pattern generated by JN375713 and JN375718 with 20 hybrids of <i>Ascocentrum</i>	135-136
21	Amplification pattern generated by ISSR 901 and ISSR 17899A with 20 hybrids of <i>Ascocentrum</i>	139-140
22	Amplification pattern generated by ACTG(4) and AW3 with 20 hybrids of <i>Ascocentrum</i>	139-140
23	Amplification pattern generated by (CT)10G and DAT with 20 hybrids of <i>Ascocentrum</i>	139-140
24	Amplification pattern generated by (GACAC)4 and GOOFY with 20 hybrids of <i>Ascocentrum</i>	139-140
25	Amplification pattern generated by MANNY and MAO with 20 hybrids of <i>Ascocentrum</i>	139-140
26	Amplification pattern generated by OMAR and UBC 807 with 20 hybrids of <i>Ascocentrum</i>	139-140
27	Amplification pattern generated by UBC 808 and UBC 809 with 20 hybrids of <i>Ascocentrum</i>	139-140
28	Amplification pattern generated by UBC 810 and UBC 814 with 20 hybrids of <i>Ascocentrum</i>	139-140
29	Amplification pattern generated by UBC 841 and UBC 858 with 20 hybrids of <i>Ascocentrum</i>	139-140
30	Amplification pattern generated by ISSR 7 with 20 hybrids of <i>Ascocentrum</i>	139-140

## LIST OF FIGURES

Sl. No.	Title	Between pages
1	Plant height of <i>Ascocentrum</i> hybrids/varieties during the period 2016-18	60-61
2	Plant spread of <i>Ascocentrum</i> hybrids/varieties during the period 2016-18	62-63
3	Shoot girth of <i>Ascocentrum</i> hybrids/varieties during the period 2016-18	63-64
4	Shoot diameter <i>Ascocentrum</i> hybrids/varieties during the period 2016-18	64-65
5	Internodal length of <i>Ascocentrum</i> hybrids/varieties during the period 2016-18	65-66
6	Number of leaves of <i>Ascocentrum</i> hybrids/varieties during the period 2016-18	66-67
7	Leaf length of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17	68-69
8	Leaf length of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18	68-69
9	Leaf breadth of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17	68-69
10	Leaf breadth of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18	68-69
11	Leaf area of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17	69-70
12	Leaf area of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18	69-70
13	Number of roots of <i>Ascocentrum</i> hybrids/varieties during the period 2016-18	70-71
14	Root girth of <i>Ascocentrum</i> hybrids/varieties during the period 2016-18	72-73
15	Dendrogram showing clustering of 30 intergeneric hybrids of <i>Ascocentrum</i>	78-79

<b>Sl. No.</b>	<b>Title</b>	<b>Between pages</b>
16	Dendogram showing clustering of 20 hybrids/varieties based on SSR profile	138-139
17	Dendogram showing clustering of 20 hybrids/varieties based on ISSR profile	139-140
18	Number spikes produced per plant per year (yield of spikes) of intergeneric hybrids of <i>Ascocentrum</i>	147-148
19	Spike, peduncle and rachis length of intergeneric hybrids of <i>Ascocentrum</i>	148-149
20	Number of florets per spike of intergeneric hybrids of <i>Ascocentrum</i>	149-150
21	Internodal length of spikes of intergeneric hybrids of <i>Ascocentrum</i>	149-150
22	Fresh weight of spikes and physiological loss in weight of <i>Ascocentrum</i> hybrids	153-154
23	Post harvest floral characters of intergeneric hybrids of <i>Ascocentrum</i>	154-155
24	Water uptake of intergeneric hybrids of <i>Ascocentrum</i> in vase	155-156



## LIST OF APPENDICES

Sl. No.	Title	Page No.
I	Plant height of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17, cm	I-II
II	Plant height of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18, cm	III-IV
III	Plant spread of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17, cm	V-V1
IV	Plant spread of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18, cm	VII-VIII
V	Shoot girth of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17, cm	IX-X
VI	Shoot girth of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18, cm	XI-XII
VII	Shoot diameter <i>Ascocentrum</i> hybrids/varieties during the period 2016-17, cm	XIII-XIV
VIII	Shoot diameter <i>Ascocentrum</i> hybrids/varieties during the period 2017-18, cm	XV-XVI
IX	Internodal length of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17, cm	XVII-XVIII
X	Internodal length of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18, cm	XIX-XX
XI	Number of leaves of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17	XXI-XXII
XII	Number of leaves of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18	XXIII-XXIV
XIII	Leaf length of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17, cm	XXV-XXVI
XIV	Leaf length of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18, cm	XXVII-XXVIII

<b>Sl. No.</b>	<b>Title</b>	<b>Page No.</b>
XV	Leaf breadth of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17, cm	XXIX-XXX
XVI	Leaf breadth of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18, cm	XXXI-XXXII
XVII	Leaf area of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17, cm <sup>2</sup>	XXXIII-XXXIV
XVIII	Leaf area of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18, cm <sup>2</sup>	XXXV-XXXVI
XIX	Number of roots of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17	XXXVII-XXXVIII
XX	Number of roots of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18	XXXIX-XL
XXI	Length of roots of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17, cm	XLI-XLII
XXII	Length of roots of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18, cm	XLIII-XLIV
XXIII	Girth of roots of <i>Ascocentrum</i> hybrids/varieties during the period 2016-17, cm	XLV-XLVI
XXIV	Girth of roots of <i>Ascocentrum</i> hybrids/varieties during the period 2017-18, cm	XLVII-XLVIII
XXV	Description of leaf characters and floral morphological characters	XLIX-LIII
XXVI	Monthly distribution of weather parameters during the experiment April 2016 - October 2019	LIV-LV
XXVII	List of laboratory equipment used for the study	LVI

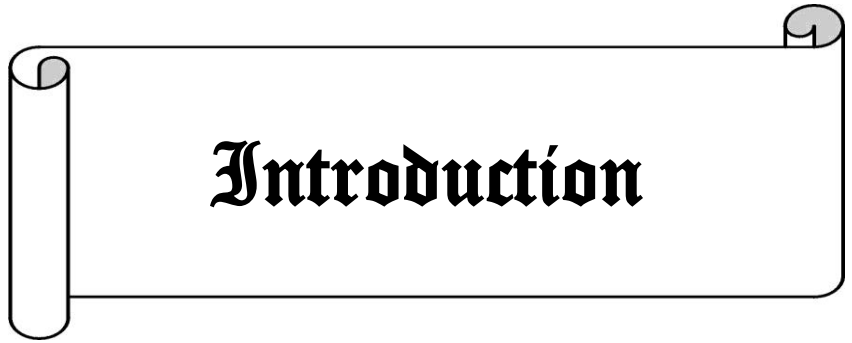
## ABBREVIATIONS

-	Substraction, dash and to
<	Less than
>	More than
\$	Dollar
%	Percentage
&	And (in addition)
/	For each and oblique (or)
:	Ratio
@	At the rate of
'16	2016
'17	2017
'18	2018
'19	2019
+	Addition
=	Equal to
×	Crossed with
µg	Microgram
µl	Microliter
♀	Female parent
♂	Male parent
°	Degree
°C	Degree celsius
10x lens	Low power microscope
40x lens	High power microscope
A.	<i>Arachnis</i>
AFLP	Amplified fragment length polymorphism
AgNO <sub>3</sub>	Silver nitrate
am.	Ante meridiem (/before noon)

Apr.	April
Ar.	<i>Aranda</i>
Ascda.	<i>Ascocenda</i>
Asctm.	<i>Ascocentrum</i>
AT	Annealing temperature
Aug.	August
bp	Base pair
C	Cross compatibility
C.	<i>Cattleya /Cymbidium</i>
C.D.	Critical difference
Cal.	<i>Calanthe</i>
cm	Centimeter (s)
cm <sup>2</sup>	Centimeter square
cm <sup>3</sup>	Centimeter cube
CTAB	Cetyl trimethyl ammonium bromide
Cx	Cross incompatibility
D.	<i>Dendrobium</i>
DDH <sub>2</sub> O	Double distilled water
Dec.	December
DNA	Deoxyribonucleic acid
dNTP	Deoxyribonucleotide triphosphate
EC	Electrical conductivity
EDTA	Ethylene diamine tetra acetic acid
<i>et al.</i>	<i>Et alia</i> (and others or and coworkers)
<i>etc.</i>	<i>Et cetera</i> (and the rest)
F	Forward sequence
Feb.	February
Fig.	Figure
FYM	Farm yard manure
g	gram (s)

hrs.	Hours
hyb.	Hybrid (s)
<i>i.e.</i>	<i>Id est.</i> (that is)
ISSR	Inter simple sequence repeat
Jan.	January
Jul.	July
Jun.	June
K <sub>2</sub> O	Potassium oxide
<i>Kag.</i>	<i>Kagawara</i>
Kg	Kilogram (s)
kmph	Kilometer (s)
L	Ladder
lit.	Litter (s)
M	Molar
Mar.	March
Max.	Maximum
mg	Milligram (s)
MgCl <sub>2</sub>	Magnesium chloride
Min.	Minimum/minute
ml	Milliliter (s)
mm	Millimeter (s)
mM	Millimolar
mm <sup>2</sup>	Millimeter square
<i>Mok.</i>	<i>Mokara</i>
N	Nitrogen
NaCl	Sodium chloride
No.	Numbers
Nov.	November
NS	Non-significant
Oct.	October

P <sub>2</sub> O <sub>5</sub>	Phosphorus oxide
PCR	Polymerase chain reaction
pH	Potential of hydrogen ion
PIC	Polymorphic Information Index
PLW	Physiological Loss in Weight
pm.	Post meridiem (after noon)
ppm	Parts per million
PVP	Polyvinyl pyrrolidone
R	Reverse sequence
RAPD	Restriction fragment length polymorphism
RNA	Ribonucleic acid
rpm	Revolution per minute
S	Self compatibility
SE (m)	Mean standard error
Sept.	September
Sl.	Serial number
sp.	Species
Sq.	Square
SSR	Simple sequence repeat
Sx	Self incompatibility
TAE	Tris acetate EDTA
UPGMA	Unweighted pair group method with arithmetic mean
UV	Ultra-violet
V	Volts
V.	<i>Vanda</i>
Var.	Variety
<i>Vasco.</i>	<i>Vascostylis</i>
<i>viz.,</i>	<i>Vi delicet</i> (namely)
wt.	Weight
yr.	Year



# Introduction



## 1. INTRODUCTION

Orchidaceae is one of the largest and most diverse families, accounting for more than seven per cent of flowering plant species of the world. This family is the dominant group of flowering plants and known to have about 28,000 species, distributed in about 763 genera (WCSP, 2019; Christenhusz and Byng, 2016). According to Pijl and Dodson (1966), enormous diversity is found in their mode of growth, seed production, habitat, and are also unique with their versatility in the formation of their flowers, flower colour, size, shape and longer life span of flowers. These all together make it very interesting to study about orchids. Everything about the orchids is unique and conceivably this is what makes them so mesmerising (Brain and Wilma, 1979).

Orchids are classified according to the way they live and survive, *i.e.*, epiphytes, lithophytes and terrestrials. On the basis of growth habit, they can be classified as monopodial and sympodial. The monopodial orchids grow from a single bud, leaves are added from the apex each year, the stem grows vertical and longer and the stem can reach several metres in length, as in *Ascocentrum*, *Vanda*, *Arachnis*, *etc.* On the other hand, sympodials have a front (the newest growth) and a back (the oldest growth) (Nash and Frownie, 2008). The plant produces a series of neighbouring shoots, grow laterally rather than vertically as in *Dendrobium*, *Cattleya*, *etc.*

Orchids have a very wide-range of distribution. They are observed in almost every part of the world except in Antarctica. Most of the orchid cultivation is native of tropical areas and found with the great diversity in humid tropical forests of India, Australia, China, Philippines, Malaysia, Mexico, Myanmar, New Guinea, Sri Lanka, South and Central America, Thailand (Arditti, 1992). In India, the Western Ghats harbours a vast diversity of orchids. The Indian states *viz.*, Andamans, Himachal Pradesh, Karnataka, Kerala, Maharashtra, Sikkim, Tamil Nadu, West Bengal, Hills of Uttar Pradesh and North-Eastern states are suitable for commercial cultivation of



orchids (Singh, 1991). About 1,600 species of orchids are reported in India which constitutes almost 10 per cent of the world orchid flora (Singh, 1990). Indian orchids known to have medicinal traits and also floricultural traits which reached majority of the famous botanical gardens of the world (Abraham and Vatsala, 1981).

Attractiveness, diversity in forms, shape and colour, high productivity, a right season of flowering and easy packing and transportation made this flower important in the world floriculture trade. In a billion dollar international industry, orchids are priced for their spectacular beauty and longevity. The export/import trade in world orchid's market, cut flowers and pot plants exceeds \$150 million dollars. Out of these, above 80 per cent are cut orchids, and the 20 per cent are comprised of pot plants.

Orchids contribute an important part in Indian export of cut flowers which have enhanced with the recent increase in world floriculture trade, they have become the second most popular plants for cut flowers as well as pot plants with an annual growth rate of 10-20 per cent (Sudeep *et al.*, 2018). It has also gathered momentum in Kerala and North Eastern states during the early nineties. This shows that there is a huge scope for increasing India's, commercial cultivation of orchids.

Despite the vast diversity, orchids which are commercially important as cut flowers, belong to relatively a few genera viz., *Arachnis*, *Vanda*, *Phalaenopsis*, *Renanthera*, *Cymbidium*, *Cattleya*, *Dendrobium*, a few others and their hybrids like *Aranda*, *Aranthera*, *Ascocenda*, *Mokara* and *Vascostylis*.

Monopodials have recently gained popularity due to the large number of varieties and hybrids involving intergeneric ones which show a wide range of variability in floral characters. Among the monopodial orchids, *Phalaenopsis*, *Aranda*, *Aranthera*, *Mokara* and *Vanda* are most popular in the world market. *Vanda* have become extremely popular for outdoors or greenhouse cultivation in the warmer regions of the world. It is very crucial to assess wide-ranging variation of characters present in the monopodials.

*Ascocentrum* is a small-flowered vandaceous orchid which has erect inflorescence with long lasting flowers and which is not commercially exploited so far. *Vanda* has more popularity through the bigeneric hybrid *Ascocenda*, which has bright orange flowers, more number of florets and longevity which are contributed by *Ascocentrum*. This was subsequently used for the production of multi generic vandaceous hybrids viz.; *Mokara*, *Kagawara*, *Vascostylis*, etc. and many other bigeneric and multi generic hybrids have been evolved in *Asctm.* alliance. The species and hybrids are visually differentiated on the basis of colour and size of flowers.

The morphological diversity of *Ascocentrum* at intraspecific, intergeneric and varietal levels necessitated for studies at molecular level as well. The morphological and molecular characterisations are the important possible ways to study diversity. The potential of *Ascocentrum* has not been systematically exploited so far in the country. Therefore it is an essential to evaluate the influence *Ascocentrum* in their intergeneric hybrids for commercial exploitation.

Among the molecular markers used for assessment of genetic diversity, SSR markers were found to have widespread application because of their high reproducibility, codominant nature and extensive genome coverage (Agarwal *et al.*, 2008; Xu and Crouch, 2008) and easy detection by polymerase chain reaction (Castillo *et al.*, 2010). Inter-simple sequence repeats are regions in the genome flanked by microsatellite sequences. The PCR amplification of these regions using single primer yields multiple amplification products that can be used as dominant multilocus marker system for the study of genetics of various cultivated and non-cultivated orchids. The ISSR markers are easy to use, low-cost and methodologically less demanding, which makes it an ideal genetic marker for diversity studies (Ng and Tan, 2015).

Several morpho-anatomical characters have been used widely in taxonomic and phylogenetic studies as they provide valuable information on classification. The

importance of some of the modern taxonomic parameters is being increasingly realized but sparingly used for characterization.

Breeding in Orchidaceae family is unique because hybridization is conceivable not only between species but also between related genera. At this juncture, it is worth studying the possibility of self and cross compatibilities among different genera and hybrids. Establishment of an indigenous orchid industry depends on rapid multiplication and production of superior hybrids which are free flowering, and this can be made possible only by the development of novel breedings and bio techniques.

Molecular characterization is done to distinguish between hybrids/varieties so that they can be used for the future breeding programme. Hence, the present study was proposed to characterize the selected intergeneric hybrids of *Ascocentrum* at the morphological and molecular levels for maintaining their identity, knowing the genetic relation and for exploitation in future breeding programmes.

Keeping these points in view, the present study entitled 'Morpho-molecular characterization of intergeneric hybrids of *Ascocentrum*' was undertaken with the following objectives.

- To assess the performance of intergeneric hybrids of *Ascocentrum*.
- To characterise them based on morphological and molecular parameters for commercial exploitation.
- Pollen studies to find out the best male parents for cross compatibility studies.
- Compatibility studies in selected parents for exploring possibilities of self and cross compatibility, which can be further used in future breeding programme.
- Screening of more SSR and ISSR primers for diversity analysis and unique banding pattern.



# Review of literature

## 2. REVIEW OF LITERATURE

Flamboyant, intriguing and bewitchingly beautiful orchids have evolved to become the largest and most diverse family of flowering plants in the world. The Orchidaceae family was reported to have about 28,000 currently accepted species, distributed in 763 genera (WCSP, 2019; Christenhusz and Byng, 2016). In the billion-dollar international industry, orchids are prized for their spectacular beauty and longevity. Orchid flowers are known for their high magnitude of diversity in form, colour, size, shape, growth habit, leaf and root morphology, flower size, blooming period, attractiveness, longer life span and response to the environment. The main reason for such variation and perplexing range of floral structures is their high cross pollination nature and better evolutionary response of germplasm to macro and microclimatic conditions (Bose *et al.*, 1980). The manifold and perplexing range of floral structures arouse the highest admiration and attention among the flower growers (Bose *et al.*, 1999).

Vandaceous orchid comes under special group of tropical orchids, characterised by monopodial growth habit. They are coming under sub-tribe Sarcanthinae which is classified into some 86 genera consisting of 1000 species. Over the past few decades, thousands of vandaceous and *Ascocentrum* orchids were produced through interspecific and intergeneric hybridization. Some of these cultivars are commercially grown for cut flower production in countries like Malaysia and Singapore. *Aranda* is a bigeneric hybrid and *Mokara* is a trigeneric *Ascocentrum* hybrid (Chen *et al.*, 1998).

Intermediate types of *Ascocentrum* are extremely popular in the warmer region of the world because of their large dimension and unusual shaped attractive flowers of long-lasting nature. More than ten species of vandas were identified from India, China, The Himalayas, Sri Lanka, Philippines and most part of South East Asia. They are commercially exploited in Thailand, Singapore, Malaysia and Hawaii (Mukherjee, 1983; De and Debnath, 2011; Behra *et al.*, 2013).

Attractiveness, diversity in shape, size, forms, and colour, high productivity, right season of blooming and easy packing and transportation made this flower important in the world floriculture trade. The world export/import trade of orchid cut flowers and plants increased tremendously. Among these, about 80 per cent are cut orchids, and the rest pot plants. Orchids are also known to have medicinal properties. In China, it is used as an important component in herbal medicine to treat several diseases, from centuries (Bulpitt *et al.*, 2007). For this reason, it is very crucial to assess wide ranging variation of characters present in orchids as well as their hybrids, and the morphological and molecular characterisation is the possible way to achieve this goal. The works done on morphological and molecular characterisation of different orchids are comprehensively reviewed in this chapter.

## 2.1 DIVERSITY IN THE ORCHID FAMILY

Orchids are classified according to the way they live and survive, whether they are supported by trees or rocks or grow in the ground, *i.e.*, epiphytes, lithophytes and terrestrials. The mosaic of geo-climatic conditions occurring in India had resulted in a great range of habitats for the rich diversity of orchid flora (Arora, 1983). The North Eastern states has been recognised as the mega diversity area for orchids (Kumaria and Tandon, 2010). In these areas, epiphytic orchids grow up to an elevation of 2000 m from mean sea level (De *et al.*, 2014a). Amin *et al.* (2004) reported the performance of six indigenous monopodial orchids of Bangladesh.

While considering these fascinating plants, there exists an endless scope for genetic improvement and industrial development. Over the past few decades, thousands of orchids were produced through interspecific and intergeneric hybridization and nearly two lakh registered hybrids are available in the India and around world. *Ascocentrum*, *Cattelya*, *Cymbidium*, *Dendrobium*, *Neofenetia*, *Oncidium*, *Phalenopsis*, *Paphiopedilum*, *Rhynchostylis* and *Vanda* are top in the list. In the production of several excellent hybrids quality attributes were

contributed by the parent plants which were originated in India. *Vanda* and *Ascocenda* and their combinations were used to produce new hybrids (Grove, 1995). Among those, some cultivars have become particularly important for cut flower production in countries like Singapore.

Growth requirements varies among different varieties. Many species of *Arachnis*, *Renanthera*, *Ascocentrum* and *Vanda* require full sun light for their free flowering and any shading delays the flowering process. Many of the intergeneric hybrids such as *Aranda* and *Aranthera* required the same conditions for flowering whereas *Phalenopsis*, *Dendrobium* etc., required shading for flower production (Soon, 1980). According to Brian and Wilma (2014) *Vanda coerulea* was grown on oak trees where it received full sun for flowering.

### **2.1.1 Distribution of orchids in India and Kerala**

The history of orchids in India goes back to the Vedic period. The ancient Sanskrit literature holds the references to orchid like 'Nighantus' and 'Amarakosha'. As per the Botanical Survey of India, 1256 species belonging to 155 genera and 388 species of orchids are prevalent in India (Singh, 2019). The species are distributed throughout the country and can be found in North Eastern India (800 species), North Western Himalayas (200 species), Western Ghats (200 species) and a few other regions have about 100 species (Rajeevan *et al.*, 2002). About 13 species of orchids known to have medicinal traits and used as a traditional medicine in Kerala.

India is considered as the primary as well as a secondary centre of orchid biodiversity and the largest diversity of orchids found in Northeastern Himalayas, Western Ghats, and Andaman and Nicobar Islands. Small flowered orchids occurs in the Western Ghats, whereas, epiphytic orchids are common in the North Eastern states (De *et al.*, 2014a). Certain Indian orchids like *Vanda coerulea* and species of *Cymbidium*, *Dendrobium* and *Paphiopedilum* are recognised as the monarchs in the orchid flora (Behera *et al.*, 2013). The diverse agro-climatic conditions of India make it suitable to grow all types of orchids.

There is tremendous scope for different species of orchids in India, many of them are important parent plants and contributed in the production of certain beautiful hybrids. According to Rajeevan (1995), prevalence of salubrious climatic conditions greatly favoured cultivation of a vast majority of orchids in Kerala. North-East India and the Western Ghats, due to their agro-climatic diversity with high humidity and rainfall, form the richest orchid belt in the country, and around 750-800 species occur in this region. Maximum diversity of orchids is found in Arunachal Pradesh followed by Sikkim (Chowdhery, 2001). The Niyamgiri hills of Orissa is also popular to be known as an important orchid rich region, next only to Similipal and Rehana (Dash *et al.*, 2008). Forty orchid species belonging to 16 genera were evaluated by Kumar *et al.* (2012) for vegetative and flowering characters in the sub-tropical mid-hills of Meghalaya. They reported that the *Calanthe masuca*, *Cymbidium giganteum*, *Dendrobium nobile*, *Phaius tankervilleae*, *Renanthera imshootiana*, *Thunia marshalliana* and *Vanda coerulea* were the promising species for cut flowers.

### **2.1.2 Orchids in Kerala**

The Western Ghats is the natural home of about 300 species of orchids due to its suitable climatic environment (Jain, 1986). Kerala is considered as one of the very few places in the world, where worldly and well-furnished infrastructure is not mandatory for orchid cultivation. Orchids can grow well in open because of the good rainfall, high humidity and favourable temperature, which gave immense possibility for the development of orchid industry in the region (Rajeevan, 1995).

In the early nineties, the commercial aspect of orchids has been realized and since then the commercial cultivation of orchids gained momentum in Kerala. The popularity of monopodial orchids was multiplied by many folds because of the availability of the large number of hybrids and varieties including intergeneric, and the wide range of variability in floral characters (Rajeevan *et al.*, 2002). *Ascocentrum*, *Vanda*, *Arachnis*, *Rhynchostylis*, *Renanthera*, *etc.* are the monopodial genera that flourish in Kerala. Intergeneric monopodial hybrids *viz.*,



*Aranda*, *Mokara* and *Ascocenda* also performed well in Kerala (Rajeevan, 2003). George (2019) reported the favourable response of growth regulator and micro nutrients on *Ascocenda* in humid tropics of Kerala.

### **2.1.3 Description of certain genera of Orchidaceae family**

#### **2.1.3.1 *Ascocentrum***

*Ascocentrum* is a genus belonging to Orchidaceae family. There are about 19 species reported under this genus, out of which 13 are accepted (Anon, 2019a). *Ascocentrum* is extremely popular in the warmer region of the world because of the large dimension and unusual shaped attractive flowers with long lasting nature. These are monopodial epiphytes which grow on deciduous trees. It has short, simple to bifurcate stem with a compact, upright, conical to racemose inflorescence, consisting of small brightly coloured flowers. The flowers have prominent spur and strap-like lip. Their bright colours varied among orange, yellow, red or pink (Kaveriamma, 2007; Kaveriamma *et al.*, 2010).

#### **2.1.3.2 *Vanda***

The name vanda is originated from Sanskrit language (*वन्दाका*) (Wikipedia, 2019), and represent the species which are used in the treatment of ear infection rheumatic pain, and nervous system ailments (Hossain, 2011; Khan *et al.*, 2019). In the study conducted at a Botanical Research Institute in India, scientists reported the role of *V. tessellata* as a potent aphrodisiac and fertility booste (Kumar *et al.*, 2005). They also reported that the species of vanda had a history of use by the native population for its anti-inflammatory properties. The Indian vandas are also known for its antiproliferative effects against certain types of cancers, including choriocarcinoma (cancer of germ cells), lung cancer and stomach cancer (Ho and Chen, 2003).

There are about 184 names included in The Plant List of vandaceous species (Anon, 2019b), out of which 62 are accepted names and the remaining 122 are synonyms. *Vanda* species are dispersed across New Guinea, East and Southeast

Asia, Asia, Himalaya, northern Australia with some of the species outspreading into Queensland and some of the islands of the western Pacific (WCSP, 2019 and Khan *et al.*, 2019).

In the last three decades, the species of *Vanda* have gained extreme popularity and they are now amongst the leading orchids cultivated outdoors and in greenhouse in the warm regions of the world. They are attractive because of large dimension and unusual shaped flowers. The flowers also last longer and in some species the flower may remain fresh for a period of three months. The genus is commonly cultivated and highly valued for its fragrant, long-lasting, showy and deeply colourful flowers for indoor display (Dressler, 1981).

These orchids are mostly epiphytic but sometimes also found to be terrestrial. The members of this genus have monopodial growth habit with highly diverse leaves strap shaped, terete and semi terete shaped, *etc.* (Sebastian, 2015). *Vanda* orchids usually have flat, broad, ovoid leaves, whereas some have cylindrical, fleshy leaves too. The inflorescence bears a few to many flattened flowers which grow laterally. The roots are long and hung freely thus they indicated the need to cling on support (Bose *et al.*, 1999). Most of the members of this genus bear yellow brown coloured flowers with brown markings, but a few also observed to have white, green, orange, red, and burgundy shades of flower (Dressler, 1981).

### **2.1.3.3 *Arachnis***

The genus *Arachnis*, popularly known as spider orchid, comprises of 20 species native to India, China, Indonesia, Southeast Asia, New Guinea, Philippines and the Solomon Islands (WCSP, 2019). This monopodial genus is characterised by long stems, long and thin dark green alternately arranged leaves, long and branched inflorescence bearing attractive and strikingly coloured flowers giving a spidery image. The flowers are medium to large in size, showy and fragrant, typically in shades of yellow with red-brown markings. This genus is represented

by five species in India viz., *A. cathcartii*, *A. clarkei*, *A. labrosa* *A. senapatiana* and *A. labrosa* var. *Zhaoi* (Jakha *et al.*, 2015).

#### **2.1.3.4 *Rhynchostylis***

*Rhynchostylis* is a genus belonging to the orchid family, closely allied to the genus *Vanda*. The genus is native to the Indian Subcontinent, China, Indochina, Malaysia, Indonesia and the Philippines (WCSP, 2019). Currently, this genus is known to include 49 species, out of these, only three are accepted (Anon, 2019c). The flowers are borne in dense racemes and are very well known for their intense, spicy fragrance. The plants have leathery leaves and thus contribute to drought resistance. The genus can grow in a warm, moist and shaded tropical regions and can flourish in cultivation if given consistent warmth, uniform moisture and bright but indirect light. The hobbyist who wants to grow the *Rhynchostylis* needs a warm and humid tropical weather with gentle air movement.

Despite being in the *Vanda* family they are very different from vandas and also grow slower than vandas, their roots are brittle and any direct sun can harm them. The species of this genus were known to possess valuable medicinal properties (Lawler, 1984). Several species also have been found to contain alkaloids (Pridgeon *et al.*, 2001).

#### **2.1.3.5 *Renanthera***

*Renanthera* is a genus of scrambling style terrestrial and epiphytic orchid. The genus is usually found in the Himalayas, China, Southeast Asia, New Guinea and Melanesia (WCSP, 2019). *Renanthera* are narrow endemics, found only in a small area with much specialised habitat. Species of this genus produces branched inflorescence containing numerous flowers ranging in colour from yellow and orange to red and possess large lateral sepals (Rice, 2008). The genus requires an intermediate to hot climate with good air movement and bright light. Their scrambling style of growth indicate that they are best grown for hanging pots after excellent drainage.

#### **2.1.4 Description of intergeneric hybrids of *Ascocentrum***

Four important intergeneric hybrids of *Ascocentrum* viz., *Ascocenda*, *Vascostylis*, *Mokara* and *Kagawara* are described below.

##### **2.1.4.1 *Ascocenda***

*Ascocenda* is a bigeneric hybrid with intermediate climbing habit. These hybrids/varieties have different genes of *Ascocentrum* and *Vanda*, *Ascocenda* are compact monopodial plants that can easily grow indoors and bear the jewel like flowers of the *Vanda*, whereas *Mokara*, *Vascostylis* and *Kagawara* have flowering pattern unlike their parents. Studies regarding growth and flowering behavior of different *Ascocenda* and vandaceous orchids are also reported by Kaveriamma (2007); Kaveriamma *et al.* (2010); Sebastian (2015); Deepa (2017) and George (2019).

##### **2.1.4.2 *Vascostylis***

*Vascostylis* is a trigeneric hybrid of *Ascocentrum*, *Vanda* and *Rhyncostylis* and these hybrids have outrageously same growth habit and intermediate climbing growth pattern, while the flowering pattern is quite similar to *Rhyncostylis*. Studies regarding growth and flowering behavior of different *Vascostylis* orchids are also reported by Kaveriamma (2007) and Deepa (2017).

##### **2.1.4.3 *Mokara***

*Mokara* is a trigeneric hybrid cross between *Ascocentrum* × *Arachnis* × *Vanda*. Some of the *Mokara* hybrids offer the very best of their parents. They are having intermediate growth pattern like vandas. Since *Arachnis* is one of the parents, most of *Mokara* hybrids also retain the classic climbing orchid growth pattern with tall stems reaching two or more meters. They require stakes or posts as supports or else they can be supported by horizontal wires stretched between occasional upright supports. Studies regarding such tall climbing hybrids were also reported by Kaveriamma (2007) and Kaveriamma *et al.* (2008). *Mokara* hybrids were also popular among the consumers as *Aranda* hybrids (Chen *et al.*, 2000).

#### 2.1.4.4 *Kagawara*

*Kagawara* is a trigenic hybrid of *Ascocentrum* × *Vanda* × *Renanthera*. Some of them have similar characteristics of vandaceous orchids (Sebastian, 2015) and the *Kagawara* plants offer the best of both of their parents, and their flowering pattern is quite similar to their parents. These plants also exhibit intermediate climbing growth pattern like classic climbing *Mokara* varieties.

## 2.2 MORPHOLOGICAL VARIABILITY

The primary grouping of orchids on the basis of their morphological features was made by Ernst Pflizer in the late 19<sup>th</sup> century. As per the growth habits, orchids are classified as monopodials, sympodials and pseudomonopodials. Akshata (2018) reported the morphological variability in wild orchids. Wide-ranging variation existed in orchids which makes them as most highly ecologically adapted flowering plants (Mehra and Vij, 1974). The extraordinary nature of the Orchidaceae traits reflected in its huge amount of diversity coupled with peculiar pollination contrivances and wide natural hybridization; believed to be an active state of “evolutionary flux” (Chatterji, 1986).

### 2.2.1 Plant characters

Plant characters of the two groups, monopodial and sympodial, are considered to be important. Monopodial grows with single non-branching stem and are mostly climbers. Leaves, roots and inflorescences are produced from the nodes, along the entire length of the stem. The roots absorb moisture and nutrients from the air. *Vanda*, *Arachnis*, *Aerides*, *Ascocentrum*, *Rhynchostylis* and *Phalenopsis* belong to this group.

A most of the orchids come under sympodials. Sympodial orchids have rhizomes or modified bulbs which grow horizontally, producing new growths. The new growth originating from the base of a sympodial orchid is called a 'lead', which indicates the direction of growth of the plant. The flower spikes may be either lateral or terminal. Orchids of this group produce pseudobulbs with swollen stems

which store water and food. Some examples of this group are *Cattleya*, *Dendrobium*, *Oncidium*, etc.

In general, the monopodial ('single footed', by meaning) has a vertical growth, whereas the sympodial ('united feet') has a horizontal growth (Rajeevan *et al.*, 2002). The stem of both monopodial and sympodial is usually thick green and hardier which, store food and water. Pseudomonopodials are intermediate in between monopodial and sympodial orchids.

### **2.2.1.1 Leaves**

Orchid leaves are plicate strap-shaped, terete or conduplicate with sheathed bases and of different thickness, grow in all possible dimensions (Bose *et al.*, 1999). The leaves of vandaceous orchids are simple, arising from the main stem and green in colour. Terete vanda has cylindrical leaves and semi-terete vanda is a hybrid between the terete type and strap leaved type. However, other shapes are also possible owing to hybridisation (Soon, 1980). When terete leaved orchids are crossed with strap leaves orchids, the offspring observed to have intermediate leaves characterized by long and rather succulent, but with a deep channel in the upper surface, these leaves are known as semi-terete. Likewise, when flat leaved parents were used in crossing, it diluted the terete effect further, resulting in broadly channelled, succulent leaves (Elliot, 1994). Leaf tips are serrated and these enable the plant to dispose of any excess moisture taken up through the roots.

Mostly leaves are oriented in two ranks, alternating on opposite sides of the stem and rarely are opposite or whorled. In a few cases, by compression of the internodes, there are two or more leaves arising at the same level. Most orchid leaves are typical to the monocot leaves, with many parallel veins with inconspicuous cross connections between them. In many cases, the basal portion of the leaf forms a sheath around the stem (Dressler, 1993). Leaves were usually green but occasionally had silvery or golden veins which gave beautiful ornamentation as in *Anoectochilus* (Bose *et al.*, 1999).

### **2.2.1.2 Roots**

The aerial roots of orchids are unique in the plant kingdom. They are thick and mostly white. Roots consisted of a thick inner core, with an absorbent outer covering made up of layers of dead cells, help for the absorption of moisture and minerals, called velamen and progress behind the green growing tips (Khasim and Rao, 1986). However, the main function of velamen is to protect underlying tissues. Depending on habit, different orchids have different kinds of roots. However, in common, the roots of most orchids are cylindrical, often threadlike, branched and frequently elongated. Roots are extremely vulnerable and can be easily broken when it is outside the pots. They provide the plant with essential water and nutrients from the atmosphere.

## **2.2.2 Floral characters**

### **2.2.2.1 Inflorescence**

The flowers may develop singly or in a group as inflorescence. The inflorescence is solitary or spike, racemes or panicle (*Oncidium*). Development of inflorescence in *Vanda* and *Arachnis* usually requires a time of two months. During early period of growth, floral bud differentiation proceeds very slowly but becomes much faster when the growth of the inflorescence stalk ends. In monopodial orchids, inflorescences arose from axillary buds at nodes some distance from the shoot apex (Hew *et al.*, 1996), whereas in *Phalaenopsis*, the inflorescences arose on alternate sides between the leaves (Chen and Jeffrey, 2009). In case of ever blooming orchids like *Arachnis*, *Vanda* and *Aranda*, the new inflorescence initiates as soon as all the old blooms have dropped off.

The orchid inflorescence is usually raceme or indeterminate. The flowers in the inflorescence of *Vanda* and *Arachnis* normally open acropetally at one day interval (Goh, 1977). The flowers are spirally arranged on the rachis, but the bracts and flowers are distichous in some groups. The flowers are whorled in a few cases, as in *Chamaeangis* and some *Oberonia* species. The flowers were axillary on the rachis and usually opened from the base upward (Dressler, 1993). The bract is

usually inconspicuous, but maybe large or coloured, as in *Cyrtopodium* or *Lockharti* (Dressler, 1993).

#### 2.2.2.2 Flower

The flowers of orchids are perfect (containing both female and male reproductive structures) rarely unisexual, every so often showy, bracteate, epigynous, trimerous, mostly resupinate *i.e.* twisted to 180° or upside down. Usually, orchid flower is zygomorphic (bilaterally symmetric), though few exceptions like *Mormodes*, *Ludisia* and *Macodes* are also noticed.

The details regarding the complex structure of the orchid flower were first reported by Brown (1833) and later by Darwin (1862). The orchid flower is made up of three sepals in the outer whorl and three petals in the inner whorl. The medial petal is typically modified and called the labellum or lip forming a platform for pollinators. The reproductive organs (stamens and pistil) in the centre have adapted to become a cylindrical structure called the column.

The labellum is the most prominent of all perianth parts with attractive colours. The lip is trilobed, with the central lobe much more prominent than the side lobes. The lip is attached to the base of the column, the mode of the attachment may be rigid or loose (Abraham and Vatsala, 1981a). The intricate detail on the lip formed the minute decoration and rather mysterious shape that attracted the pollinating insects (Rittershausan and Rittershausan, 1999).

Lot of variabilities exist in orchids with respect to flowers, particularly in shape, size and colour. The largest orchid flower observed was of *Sobralia macrantha* (15 to 30 cm across), and the tiniest one was that of the *Bulbopyllum minutissimum* (Bose *et al.*, 1999). Some of the orchids have an appearance like ladies' slipper (*Cypripedium*). *Ophyrus apifera* appears like a bee whereas, *Coeloglossum viride* looks like a minute frog. *Brassia* spikes is like a small collection of colourful spiders.



Six tepals are present in two whorls. Outer three tepals (representing calyx) green in colour and inner three tepals coloured (representing corolla). Labellum or lip (broad, shoe-like spurred, tubular, strap-shaped or butterfly-shaped) variously branched and contributing most to the beauty of the flower. The labellum is actually posterior and lies on the anterior side of the flower, due to twisting of the inferior ovary through 180° or by the bending back of pedicel over the apex of the stem, at the time of anthesis, flower resupination occurs.

#### **2.2.2.3 Androecium**

Three stamens are present in male part of the flower. The stamens are united with the pistil to form a column. The pollens form compact and waxy masses called pollinia, which was considered as an important character for orchid taxonomy (Dressler, 1981). Two or four pollinia are observed in a flower. A connection between ovary and stamen is made by the beak like sterile stigma which almost occupies the centre of the column. In vanda, the pollinia occur as two notched pollinia, whereas, four pollinia are present in pairs, in *Arachnis*, *Phalaenopsis*, *Aerides*, *Renanthera* and *Angraecum*. In monopodial orchids, only the odd stamen of the outer whorl, placed opposite the labellum, is fertile. Occasionally staminodes are also present.

#### **2.2.2.4 Gynoecium**

The gynoecium is tricarpeal, syncarpous, unilocular, shows parietal placentation, rarely trilocular with axile placentation. Three stigmas are present, of which two lateral are often fertile, the third stigma is sterile forming a small beaked outgrowth. It had inferior ovary which consist of numerous minute ovules (Abraham and Vatsala, 1981; Mukherjee, 1990). In vanda, the ovary has ridges and was twisted due to a process called resupination (Bose and Bhattacharjee, 1980).

For a successful hybridisation, the selection of parents with good ornamental traits especially in floral characters is very important. Studies are to be undertaken to understand the diversity in both sympodial and monopodial orchid genotypes. Amin *et al.* (2004) observed significant differences on both quantitative

and qualitative characters among varieties of indigenous monopodial orchid genotypes belonging to genera *Rhynchostylis* and *Vanda*. Thomas and Rani (2008) evaluated 15 monopodial orchids belonging to the genera *Aranda*, *Aranthera*, *Kagawara*, *Mokara*, *Renanthera* and *Vanda* and found a wide range of variability among orchids, enabling for selection of suitable parents in hybridisation.

#### **2.2.2.5 Colour and fragrance**

Chlorophyll (green), carotene (yellow and orange), anthocyanidins (red, blue, purple) and flavones (pale yellow) are the four pigments which are the potential genetic colour palette. Unique colour combinations or shades are created by several pigments which are present within the plant and is predetermined at the cellular level. Vandaceous orchids vary in colour from red, yellow, pink, green, brown, violet, purple and its different shades. *Vanda coerulea* is one of the few orchids which can produce varieties with blue coloured flowers. *V. dearie* is one of the sources of yellow colour.

Colour and scent are the major pollinator attractants in flowers, and their production may be linked by shared biosynthetic pathways. Delle-Vedove *et al.* (2017) conducted a study to find out the relation between colour and scent, and reported that in varieties of *Calanthe sylvatica*, which displayed three colours, two scents it was proved that colour was not always a good indicator of odour and that colour-scent associations may be complex, depending on pollination ecology of the populations concerned.

Raguso (2008) stated that fragrance was a highly complex component of floral phenotype for its dynamic patterns of emission and chemical composition. Floral volatiles have antimicrobial properties which could be used by the plants to protect their vital floral reproductive parts from potential predators. In almost all major civilizations, people have used flowers with vibrant colours and scents to enhance their beauty. According to Baudino *et al.* (2007) in most of the plants, economic importance relied on petals which were found to be the main site of natural fragrances and flavour.

Tatsuzawa *et al.* (2004) isolated eight major acylated anthocyanins from vanda hybrid cultivars and more than 11 anthocyanins were observed in these hybrids. Alkaline floral pH determined the blue colour in many orchid flowers (Griesbach, 2005). Yokoi (1975) and Arditti (1992), reported the occurrence of delphinidin and cyanidins as anthocyanidins in the flowers of *Vanda coerulea*.

Frowine (2005) reported that more than 400 orchids emitted fragrance. Only two per cent of fragrant orchids were studied for their fragrance. Fragrant studies of orchids were not well established as in other flowers such as rose, petunia, *etc.*

Thomas and Rani (2008) examined the performance of 15 monopodial orchid genotypes of different genera (*Aranthera*, *Aranda*, *Kagawara*, *Renanthera*, *Mokara* and *Vanda*). These genotypes were commercially popular for quantitative floral characters *viz.*, number of inflorescence produced per year, length of inflorescence and length and width of flowers; and qualitative floral characters *viz.*, flowering nature, mode of display of flowers and vase life. In their study they observed noticeable variation in most of the characters studied and they further concluded that some of these genotypes can be used for breeding novel orchids.

### **2.2.3 Floral biology**

#### **2.2.3.1 Anthesis**

The mode of flowering behaviour has a direct relation to pollination biology (Croat, 1980). Sobhana, (2000) evaluated the anthesis time of ten different varieties of *Dendrobium* and reported that generally flowers opened during the day and the time of anthesis varied from variety to variety. Anthesis time for all the varieties was 7:30 am to 12.30 pm except Banyat Pink which opened during 11:00 am - 2:30 pm. Also reported that a day was required for the complete opening of the flower. Selection of a specific colour of shade net for the greenhouse or nursery also proved beneficial for the anthesis time. Leite *et al.* (2008) observed, red

coloured shade net preponed anthesis time and also increased the number of flowers as compared to blue and black shades of shad nets.

### **2.2.3.2 Stigma receptivity**

Clader and Slater (1985) studied the stigma receptivity in *Dendrobium speciosum* and reported that the pollination did not affect stigma receptivity but shortened the vase life of flowers. However, stigmas could remain receptive up to six days after pollination.

Heslop-Harrison (2000) reported that the stigma of the studied orchids could retain receptivity for a long time (up to 60 days). Further reported that the cuticular layer covering the stigma might protect and prolong the life of the stigmatic cells. Calder and Slater (1985) observed that the orchid stigma without a cuticular layer, as in *Dendrobium*, could also remain receptive over long periods.

The wet stigma of orchids is different in morphology from that of other plants. They have a specialised detached secretory cell (eleutherocytes) in a mucilage matrix. This stigma may be an evolutionary development resulting from the special needs of both pollination and nutritional support of a large number of developing pollen tubes (Clader and Slater, 1985). As per the report of Sobhana (2000), the *Dendrobium* variety New Pink was observed to have retained stigma receptivity up to 10 days after anthesis. Stpiczyńska (2003) studied the stigma receptivity of *Platanthera chlorantha* orchid and found receptivity in buds one day before anthesis, however, because of lack of fluid on their surfaces, pollinia did not adhere to them. Stigmatic receptivity lasted for 15 days.

### **2.2.3.3 Pollen studies**

Hyde and Williams (1944) coined the term palynology for the study of pollen and spore science. Niimoto and Sagawa (1961) stated that the orchids were unusual with respect to their placental proliferation and ovule formation which occurred only after anthesis and required pollination to stimulate development.

Successful hybridisation programme in orchids depends upon numerous factors viz., pollen production, viability, germination, dissemination and fertility.

#### **2.2.3.3.1 Pollen production**

Haemocytometer is an instrument used for visual counting of the number of cells in a blood sample or other fluid. It is specialised slide that has a counting chamber and with a known volume of sample liquid added to it, the number of mixed cells can be visualised and counted under a microscope. This instrument also can be used for counting the pollen grains for assessing the pollen production. Oberle and Goertzen (1952) used haemocytometer for the first time to determine pollen production in the study to reveal the remarkable inter varietal difference in some fruit species. Later on, several researchers have used this technique for the determination of pollen production.

Godini (1981) used haemocytometer for counting pollen grains of some almond cultivars. Prasad *et al.* (1999) investigated the pollen production of groundnut by using haemocytometer and established positive linear relationships between fruit set as well as pollen production and pollen viability. Sobhana (2000) studied the pollen production of 10 varieties of *Dendrobium* by crushing and staining the pollinia of 3-4 days old opened flower and observed under Neubauer haemocytometer. Lehnebach and Riveros (2003) used haemocytometric method to assess pollen production in order to study pollination biology of the Chilean endemic orchid *Chloraea lamellata*. Kumar *et al.* (2015) and Hicks *et al.* (2016) also used the same technique to study pollen production in *Canarium strictum* and urban flowers, respectively.

#### **2.5.2.3.2 Pollen fertility and germination**

Sobhana (2000) standardised the media for pollen germination by using varying concentrations of sucrose and agar. Highest level of germination was recorded in 2 per cent sucrose + 1 per cent agar (80.33 %) and 2 per cent sucrose + 2 per cent agar (78.00 %). Whereas, the lowest percentage was found in 1 per cent sucrose + 1 per cent agar (26.67 %). Addition of 75 mg l<sup>-1</sup> did not enhance

germination percentage significantly except in 1 per cent sucrose + 1 per cent agar which led to the enhancement percentage from 26.67 to 41.67.

According to Sobhana (2000) maximum pollen fertility was recorded in Emma White (86.09 %) and Pink Tips (82.25 %), whereas minimum fertility in *Dendrobium chrysanthemum*. maximum pollen germination was recorded in *Dendrobium* variety Emma White (80.63 %) and lowest in *D. pierardii*.

In *Phalaenopsis aphrodite*, the pollen germination was studied in 10 per cent sucrose in the presence of stigmatic tissue (Chen and Fang, 2016) and revealed that the pollen tube entered the ovary three days after pollination. Whereas, the pollen tube entered the matured embryo sac after 60-65 days of pollination.

#### **2.2.4 Longevity of floret**

According to De *et al.* (2014b) opening of florets in a vase, changes in the fresh weight, diameter and length of florets, diameter or length of stem or pedicel, senescence pattern, colour of petal, total longevity and foliage burning were the attributes which determined the vase life or longevity of cut flowers.

AgNO<sub>3</sub> (10-30 ppm) and HQS (50-100 ppm) extended vase life and bud opening of cut flowers, especially in tropical orchids like *Dendrobium* (De *et al.*, 2014b) and also reported that the highest per cent of fully opened buds (75 %) and maximum vase life (45 days) in *Cymbidium*, with the chemical combination of sugar 4 per cent + salicylic acid 200 ppm (De *et al.*, 2014b). Emasculation accelerated colouration of labellum and senescence, whereas aminoxy acetic acid retarded these processes and with a higher concentration of Amino Oxyacetic Acid and ethylene, the colouration of the labellum can be delayed (Harkema and Struijlaart, 1989). Tatsuzawa *et al.* (2004) analyzed and enlisted the flower colour and pigments in *Disa* hybrids. Pollination of flowers led to the acceleration of senescence and showed symptoms like discolouration, wilting, anthocyanin production and abscission.

### 2.3 COMPATIBILITY STUDIES IN MONOPODIAL ORCHIDS

Orchidaceae has attracted plant breeders due to the unrestricted combination of genomes possible within the family. As a result, more than one lakh orchid hybrids have been registered over the past century.

The monopodial orchids fetch low market value even though they have longer vase life. An important reason for this is the cultivation of old obsolete varieties and lack of novelty. It is essential to take up monopodial orchid breeding earnestly to produce novel and adapted quality hybrids.

An in depth understanding of the compatibility relationships of the concerned genera is crucial for successful hybrid development in orchids. Many of the time self incompatibility and cross sterility were commonly observed among the cultivated orchid hybrids. This can be attributed to one of the two causes, hybrid sterility or polyploidy (Lenz and Wimber, 1959).

Chen *et al.* (2000) reported that out of 520 hybridizations conducted with the aim of developing white Taisuco *Phalaenopsis*, only 46.2 per cent cross combinations produced viable seeds.

Melendez-Ackerman and Ackerman (2001) reported self compatibility in *Lisiera cordata*, as all self pollinations produced fruits. Cross pollinations differed significantly from the self, registering a higher number of seeds per capsule and a higher percentage of fertilized ovules.

Rani (2002) performed a total of 190 self and cross combinations in dendrobium to determine cross compatibility, out of which 84 combinations including seven selves produced harvestable green capsules, the relative success being 44.21 per cent. Progeny from 67 hybrid combinations were established successfully in the greenhouse.

## 2.4 ARTIFICIAL POLLINATION AND HYBRIDIZATION

Artificial hybridization in orchids was started much later than in other angiosperm families. The main hurdles were the complexity of their flower structure and the consequent lack of understanding of the method of pollination.

John Dominy cross pollinated two *Calanthe* sp., *Cal. masuca* × *Cal. Furcata* in 1852 and the resultant plants flowered in 1856 and thus became the first man assisted orchid hybrid *Cal. dominyi* (Dressler, 1981). At the same time Dominy also cross pollinated *Cattleya guttata* with *C. loddigesii* and got the successful hybrids (*C. hybrida*) (Abraham and Vatsala, 1981a).

In hybridization, selection of good and healthy parent plant and flower by visual observation accounts to a great extent. Very young plants or seedlings, as well as plants with unhealthy looking canes blooming for the first time, should not be selected as mother plant (Bose and Bhattacharjee, 1980).

Warren (1981) described different pollination mechanisms in orchids which varied widely depending on the floral morphology of the genera concerned.

Rhodehamel (1994) observed that cross pollination in monopodial orchids was effected by the deposition of pollinia from one flower into the stigmatic cup of another flower in another plant. Adherence of pollen to stigma was effected by the wetness of pollen or stigma.

## 2.5 ASSESSMENT OF PARENTAL POLYMORPHISM THROUGH SSR

Successful breeding requires profound information on the diversity available within the species. This information helps the breeder to decide appropriate and diverse parents as per the objectives of the breeding programmes. The parents which are phenotypically dissimilar need not to be always dissimilar genotypically as the phenotype is a product of genotype and micro/macro environment. Moreover, morphological and physiological traits of different members of same species have a high level of genetic variation associated with a



population of different geographic origin (Libby *et al.*, 1969). These facts make an assessment of parental polymorphism in available genotypes of great importance. It is also presumed that the varieties developed through crossing will give a more frequency of transgressive segregants if the parents used for crossing were, to some extent, genetically diverse (Simioniuc *et al.*, 2002). Knowledge of genetic diversity among the genotypes also helps in determining core collection for plant biodiversity conservation. The flowering season of orchids and its duration were both determined genetically (Goh and Arditti, 1985).

### **2.5.1 Molecular DNA based markers**

The development and use of molecular markers for the detection and exploitation of DNA level polymorphism is a single most momentous development in the field of molecular genetics. Molecular markers (DNA markers) are developed to overcome the limitations of morphological markers which tend to express differently as the environment around plants changes. However, it does not mean that any of the biochemical or molecular techniques or both have replaced morphological markers. Molecular markers own great potential for its use in the breeding programmes. These markers are distinguishable DNA sequences, found at specific loci and transmitted by the standard laws of inheritance from one generation to the next (Semagn *et al.*, 2006). Apart from this, DNA markers are stable in different environments and plant developmental stages. The polymorphism in these markers provide the ability to discriminate between individuals, thereby helps in the careful selection of parents for a breeding programme.

A large number of PCR based DNA markers *viz.*, Random Amplified Polymorphic DNA (RAPD), Amplified Fragment Length Polymorphism (AFLP), or Simple Sequence Repeats (SSR), Inter-Simple Sequence Repeats (ISSR), *etc.* provide an opportunity for fine-scale genetic characterisations. Nevertheless, they also generate a large amount of data in a short period of time (Powell *et al.*, 1996; Hokanson *et al.*, 1998). Therefore, these DNA markers are often used for

assessment of genetic diversity and relationships, DNA fingerprinting, genome mapping, in the conservation of genetic resources, studies of phylogeny and evolutionary biology, gene tagging, selection of targeted traits, *etc.* (Tautz, 1989; Williams *et al.*, 1990; Reddy *et al.*, 2002). Among *Vigna* genotypes, genetic diversity and intraspecific or interspecific relationships have been worked out based on several DNA markers *viz.*, RAPD, SSR, AFLP, ISSR, *etc.* by several researchers (Fatokun *et al.*, 1993; Kaga *et al.*, 1996; Ajibade *et al.*, 2000; Li *et al.*, 2001; Souframanien and Gopalakrishna, 2004).

Chen *et al.* (1999) investigated the application of the PCR based fingerprinting technique, amplified fragment length polymorphism (AFLP), in orchids. The optimal AFLP patterns have been determined using primer combinations of EcoRI '4 and MseI '3 selective nucleotides. The same reproducible AFLP patterns were demonstrated in genomic DNAs isolated both from (1) different orchid tissues, e.g. leaves and flowers; and (2) orchid flowers collected at different times. Genomic variations among different cultivars of vandaceous orchid hybrids were successfully determined by AFLP analysis. More than 10 per cent of the AFLP bands were polymorphic DNA when siblings, derived from the same original crosses (two cultivars of *Aranda* Christine, five cultivars of *Mokara* Willie How), were used. Only 0.3–0.7 per cent of the AFLP patterns were shown to be polymorphic when different cultivars, originating from somatic mutations during meristem culture for massive propagation, were used (two cultivars of *Ar.* Christine, four cultivars of *Mok.* Chark Kuan).

### 2.5.2 Simple Sequence Repeats (SSR)

The term microsatellite was coined by Litt and Luty (1989). They are also called Simple Sequence Repeats (SSR) which are species-specific and belong to the repetitive DNA family. The SSRs are short tandem repeats consisting of 1-6 bp long monomer sequence that is repeated number of times (Joshi *et al.*, 2000). These sections of DNA contain repeating mono, di, tri, tetra or pentanucleotide units (Powell *et al.*, 1996). Dinucleotides are generally abundant in genomes. The

SSR markers are PCR based and genetically co-dominant in nature. They are robust, reproducible, hypervariable, abundant, with good genome coverage and uniformly dispersed in the plant genome, and have a significance in plant genetics and breeding (Powell *et al.*, 1996; Gupta and Prasad, 2009; Sharma *et al.*, 2015).

Microsatellite markers have applications in genetic mapping, functional diversity and comparative mapping (Jonah *et al.*, 2011). They have been successfully adopted to analyse the genetic diversity in a variety of different plant species (McCouch *et al.*, 1997; He *et al.*, 2003; Frary *et al.*, 2005; Sarikamis *et al.*, 2010). The SSRs have been widely used in major crops. The first attempt to map microsatellites in plants was in rice using (GGC)<sub>n</sub> by Zhao and Kochert (1993) followed by mapping of (GA)<sub>n</sub> and (GT)<sub>n</sub> by Tanksley *et al.* (1992) and (GA/AG)<sub>n</sub>, (ATC)<sub>10</sub> and (ATT)<sub>14</sub> by Panaud *et al.* (1995). Several researchers have used the SSR marker system to assess genetic diversity in barley (Saghai Maroof *et al.*, 1994; Holton *et al.*, 2002), wheat (Gupta and Varshney, 2000), rice (Chakravarthi and Naravaneni, 2006), sugarcane (Sharma *et al.*, 2014), Brazilian barley (Ferreira *et al.*, 2016), Chinese jujube (Fu *et al.*, 2016), some accessions of African plum in Cameroon (Tchinda *et al.*, 2016), *etc.*

### **2.5.3 Inter-Simple Sequence Repeats (ISSR)**

In the early 1990s, several research groups (Meyer *et al.*, 1993; Gupta *et al.*, 1994; Wu *et al.*, 1994 and Zietkiewicz *et al.*, 1994) independently developed the technique of Inter-Simple Sequence Repeats (ISSR) markers. ISSR is region in the genome flanked by microsatellite sequence. During single primer PCR amplification, it targets variation in the DNA between two identical, oppositely oriented microsatellite loci present at an amplifiable distance. ISSR primer is the repetitive (di, tri, tetra or pentanucleotide) sequence complementary to microsatellite regions, either unanchored (Meyer *et al.*, 1993 and Gupta *et al.*, 1994) or more usually anchored at 3' or 5' end with one to four degenerate bases extended into the flanking sequences (Zietkiewicz *et al.*, 1994) can be used.

The efficacy of ISSR marker technique for molecular characterization, identification and assessment of genetic diversity and relationship of *Cucurbita* genotypes was reported by investigators around the globe (Heikal *et al.*, 2008; Xanthopoulou *et al.*, 2015). With the advantage of using arbitrarily designed primers (Joshi *et al.*, 2000), this multilocus, dominant marker with 86 per cent to 94 per cent reproducibility, have been used for cultivar identification in carrot (Briard *et al.*, 2000), groundnut (Raina *et al.*, 2001), rice (Dharmaraj *et al.*, 2018), wheat (Tungalag *et al.*, 2018), genetic purity testing in cotton (Dongre *et al.*, 2011), to evaluate crop genetic diversity, fingerprinting, *etc.* in crops like soybean, cashew, radish, broccoli, *etc.*

#### **2.5.4 Assessment of diversity in orchids through molecular markers**

Huang *et al.* (2010) used 13 expressed sequence tag (EST)-derived simple sequence repeat (SSR) to analyse 103 cultivars of six species of Chinese orchids. They reported a total of 168 polymorphic bands with 13 SSR primer pairs generated which clearly revealed the difference between cultivars inter- or intra-species of Chinese orchid.

Fatimah and Sukma (2011) have taken up the research with the objective to develop sequence-based microsatellite (eSSR) markers, wherein, Seventeen primers were designed and thirteen primer pairs could amplify the DNA giving the expected PCR product with polymorphism. A total of 51 alleles and polymorphism information content (PIC) values at 0.674, were detected at the 16 SSR loci. They also reported the presence of noticeable variation in the *Phalaenopsis* orchids.

Lu *et al.* (2011) used inter-simple sequence repeat (ISSR) markers to assess the genetic diversity of 151 *Cymbidium sinense* cultivars collected from China and Japan, and observed moderate level of diversity in *C. sinense*.

Cai *et al.* (2012) used 12 new microsatellite markers to estimate genetic diversity and population structure of *Dendrobium loddigesii*. They detected a total of 98 scoreable alleles with the expected heterozygosity of each SSR locus varied

from 0.454 to 0.857. Polymorphism information content (PIC) of each SSR locus ranged from 0.358 to 0.838 with an average of 0.637, they reported a high level of genetic diversity in the population of *D. loddigesii*.

Chen *et al.* (2014) investigated the genetic pattern in eight SSR loci within eight *Gastrodia elata* populations from central China and detected a high level of genetic diversity in the medicinal orchid population.

Kang *et al.* (2015) used 320 *Dendrobium* SSR markers to test their transferability and polymorphism in different orchid genera. Total 109 SSR showed high transferability (an average of 24.71 %) and polymorphism (an average of 85.91 %) across 44 species of 15 orchid genera.

Ueno *et al.* (2015) used 13 ISSR primers to assess the genetic diversity of 152 individuals of *Oeceoclades maculata*. Their study revealed low intra-population genetic diversity and are remarkably divergent among the population of *O. maculate*.

Tian *et al.* (2018) used ISSR and SCoT markers to investigate genetic diversity in 17 populations of terrestrial orchid *Cypripedium japonicum* in East Asia. They found low levels of genetic diversity at the species level with a considerably high degree of genetic divergence.

Several such studies were carried out by the researchers around the globe to access the genetic diversity among different orchids. Parab, 2008; Xia *et al.*, 2008; Phuekvilai *et al.*, 2009; Lopez Roberts *et al.*, 2012; Moraes *et al.*, 2014; Peyachoknagul *et al.*, 2014a and Dharmarathna *et al.*, 2018 reported the SSR and ISSR marker system to assess genetic diversity among different orchid species and genera.



# Materials and methods

### 3. MATERIAL AND METHODS

The present study entitled “Morpho-molecular characterisation of intergeneric hybrids of *Ascocentrum*” was carried out at the Department of Floriculture and Landscaping, College of Horticulture, Kerala Agricultural University, Thrissur during 2016-2019. The research programme was designed with the objective to assess the performance of intergeneric hybrids of *Ascocentrum*, to characterize them based on morphological and molecular analysis for commercial exploitation and for use in future breeding programmes.

The materials used and the methodologies adopted for the investigation are dealt in this chapter.

#### 1) LOCATION

Vellanikkara is situated at the latitude of 10°31' N and longitude of 76°13' E. The experimental area was located at 22.25 m above mean sea level.

#### 2) CLIMATE

**The study comprised the following experiments**

#### **I. Experiment 1: Morphological characterisation/field evaluation**

##### **A. Quantitative morphological characters**

- Plant characters
- Floral characters

##### **B. Post-harvest characters**

##### **C. Phenological characters**

- Plant characters
- Floral characters

##### **D. Qualitative morphological characters**

- Plant characters
- Floral characters

#### **E. Visual evaluation**

- Spike for use as cut flower
- Plant for use in indoor display

#### **II. Experiment 2: Compatibility studies**

- Pollen studies
- Self-compatibility studies
- Cross compatibility studies
- Post pollination changes

#### **III. Experiment 3: Molecular characterisation**

- Isolation of genomic DNA
- Screening of SSR marker
- Screening of ISSR marker

### **3.1 MORPHOLOGICAL CHARACTERISATION**

#### **3.1.1 Materials**

The morphological characterisation was done in the selected healthy plants of intergeneric hybrids *viz.*, *Ascocenda*, *Vascostylis*, *Mokara*, and *Kagawara*, in which one of the parent is *Ascocentrum*. Two to three years old, fifteen full grown plants, each of 30 hybrids/varieties, grown in the orchidarium were used to evaluate their morphological characters for two years. Classification of intergeneric hybrids of *Ascocentrum* and their names included in the study are given in Table 1 and Table 2, respectively.

#### **3.1.2 Shade**

Thirty intergeneric hybrids of *Ascocentrum* were grown in even span open ventilated poly house/rain shelter of size 21 m × 6 m, having 200 micron UV film covering. The shade was provided with 25 per cent shade net as per the requirement for the best growth and flowering.



**Table 1. Classification of hybrids of *Ascocentrum* used for the study**

Sl. No.	Intergeneric hybrid	Parent	No. of hybrids included in the study	Special characters
1.	<i>Ascocenda</i>	<i>Ascocentrum.</i> × <i>Vanda</i>	6	Bigeneric, intermediate climbing
2.	<i>Vascostylis</i>	<i>Ascocentrum.</i> × <i>Vanda</i> × <i>Rhyncostylis</i>	5	Trigeneric, intermediate climbing
3	<i>Mokara</i>	<i>Ascocentrum.</i> × <i>Arachnis</i> × <i>Vanda</i>	15	Trigeneric, intermediate and tall climbing
4.	<i>Kagawara</i>	<i>Ascocentrum.</i> × <i>Vanda</i> × <i>Renanthera</i>	4	Trigeneric, intermediate climbing

### 3.1.3 Media

Intermediate climbing orchids in the category of *Vascostylis*, *Ascocenda*, *Mokara* and *Kagawara* were grown in perforated plastic pots hung from the roof, filled with media of broken tiles, coconut husk and brick pieces or without any media (Plate 1). Tall varieties were grown in trenches with broken tiles, coconut husk and brick pieces.

### 3.1.4 Cultural practices

#### 3.1.4.1 Staking

The tall climbing orchids were supported by a horizontal wire stretched between intermittent upright supports (Plate 1). Poles and stakes were provided for pot plants as support. Since the plants required good aeration around the roots, slits were provided in hanging pots to ensure better aeration and free downward growth of the roots.

#### 3.1.4.2 Irrigation

Plants were watered twice a day to provide enough moisture ensuring good drainage and aeration around the roots.



(A) Selected hanging orchids in vegetative phase



(B) Prostrate nature of growth in selected orchids

**Plate 1. Selected intergeneric hybrids in vegetative phase for field evaluation**

**Table 2. List of intergeneric hybrids/varieties used for the study**

Sl. No.	Variety No.	Hybrid/variety name
1	V <sub>1</sub>	<i>Ascocenda</i> Udomochai
2	V <sub>2</sub>	<i>Ascocenda</i> Kraillerk White × <i>Vanda</i> Sanderiana
3	V <sub>3</sub>	<i>Ascocenda</i> Kultana × <i>Vanda</i> Bitzs Heartthrob
4	V <sub>4</sub>	<i>Ascocenda</i> Yip Sum Wah × <i>Vanda</i> Josephine Van Brero
5	V <sub>5</sub>	<i>Ascocenda</i> Suksamran Sunlight
6	V <sub>6</sub>	<i>Ascocenda</i> Sirichi Fragrance
7	V <sub>7</sub>	<i>Vascostylis</i> Pine River Blue
8	V <sub>8</sub>	<i>Vascostylis</i> Pine River Pink
9	V <sub>9</sub>	<i>Vascostylis</i> Aroonsri Beauty
10	V <sub>10</sub>	<i>Vascostylis</i> Pine Rivers Fuchsia Delight
11	V <sub>11</sub>	<i>Vascostylis</i> Blue Bay White
12	V <sub>12</sub>	<i>Mokara</i> Walter Oumae Pink
13	V <sub>13</sub>	<i>Mokara</i> Calypso × <i>Vanda</i> Doctor Anek
14	V <sub>14</sub>	<i>Mokara</i> Rassmatozz
15	V <sub>15</sub>	<i>Mokara</i> Khaw Phiak Suan × <i>Ascocenda</i> Bicentennial Yellow Spot
16	V <sub>16</sub>	<i>Mokara</i> Khaw Phiak Suan × <i>Ascocenda</i> Jiraprapa
17	V <sub>17</sub>	<i>Mokara</i> Sayan × <i>Ascocenda</i> Bangkuntein Gold
18	V <sub>18</sub>	<i>Mokara</i> Calypso Pink
19	V <sub>19</sub>	<i>Mokara</i> Calypso Jumbo
20	V <sub>20</sub>	<i>Mokara</i> Chao Praya Sunset Yellow Spot
21	V <sub>21</sub>	<i>Mokara</i> Chao Praya Sunset Orange
22	V <sub>22</sub>	<i>Mokara</i> Sunspot
23	V <sub>23</sub>	<i>Mokara</i> Omayaiy Yellow
24	V <sub>24</sub>	<i>Mokara</i> Omayaiy Orange
25	V <sub>25</sub>	<i>Mokara</i> Sayan × <i>Ascocenda</i> Doung Porn
26	V <sub>26</sub>	<i>Mokara</i> Chark Kuan Pink
27	V <sub>27</sub>	<i>Kagawara</i> Youthong Beauty
28	V <sub>28</sub>	<i>Kagawara</i> Christie Low
29	V <sub>29</sub>	<i>Kagawara</i> Boon Ruby
30	V <sub>30</sub>	<i>Kagawara</i> Samrong

### 3.1.4.3 Manuring

Monopodial orchids grown in trenches were given cow dung slurry once in a month. One kg fresh cow dung mixed in 5 liters of water was applied for one square meter. Two to three applications were given in a year. Foliar spray of N:P<sub>2</sub>O<sub>5</sub>:K<sub>2</sub>O in the ratio of 3:1:1 was applied during the vegetative phase and 1:2:2

was applied during the flowering phase. The dosage was 2-3 g/lit. of water which was applied twice a week (KAU, 2016).

#### ***3.1.4.4 Plant protection***

Need based application of plant protection chemicals was carried out to control pests and diseases.

#### ***3.1.4.5 Design of the experiment***

Two to three years old, full grown, flower bearing plants of 30 hybrids/varieties, were arranged in Completely Randomised Design (CRD) with three replications in each variety and 5 plants in each replication.

#### **3.1.5 Observations**

Detailed observations were made to assess the performance of intergeneric hybrids of *Ascozentrum* and to classify them based on morphological characterisation.

##### ***3.1.5.1 Quantitative morphological characters***

At the beginning of the experiment, during the month of April, the plants were tagged and the following observations were recorded on the growth (at monthly interval), flowering and post harvest characters.

##### ***3.1.5.1.1 Plant characters***

#### **1. Plant height**

The height of the plant was measured from the base to the growing apex at monthly intervals and expressed in centimetres.

#### **2. Plant spread**

The plant spread was measured at monthly intervals and expressed in centimetres.

## **A. Shoot characters**

### **1. Shoot girth**

The girth of the shoot was measured at 10.0 cm above the base and expressed in centimetres.

### **2. Diameter of shoot**

The diameter of the shoot was measured with Vernier calliper and expressed in centimetres.

### **3. Internodal length**

The internodal length was measured at 20.0 cm below the growing tip of the shoot and expressed in centimetres.

## **B. Leaf characters**

### **1. Number of leaves**

The total number of leaves present on the plant was counted and recorded at monthly intervals.

### **2. Leaf length**

Length of the leaf was measured from base to the tip at the monthly intervals and expressed in centimetres.

### **3. Leaf breadth**

The width of the fully expanded leaf was measured and recorded at monthly intervals and expressed in centimetres.

### **4. Leaf area**

Dot method (Bleasdale, 1973) was used to measure the standard leaf area. Regression equation was used for calculating leaf area. The constant was calculated using statistical package of nonlinear regression method (Sankar *et al*, 2010), the same was expressed in a square centimetres.

$$\text{Predicted leaf area} = \text{length} \times 1.2959 + \text{breadth} \times 0.3214 + 17.2047.$$

### ***C. Root characters***

#### **1. Number of roots**

The number of roots produced by the plant was counted and recorded.

#### **2. Length of roots**

Length of the roots was measured from the base to the tip and expressed in centimetres.

#### **3. Girth of roots**

The girth of the root at five centimetres from the base was measured and expressed in centimetres.

### ***3.1.5.1.2 Floral characters***

#### ***A. Spike characters***

##### **1. Number of spikes produced per year**

The number of spikes produced on each plant was noted and number of spikes per plant per year in each variety/hybrid was calculated.

##### **2. Length of spike**

The total length of the spike from the base of the spike to tip in each plant was recorded in centimetres.

##### **3. Length of rachis**

The length of the flowering area (rachis) per spike in each plant was recorded in centimetres.

##### **4. Length of the flower stalk**

The length of flower stalk (whole spike/peduncle length) in each plant was recorded in centimetres.

##### **5. The girth of a spike at the base**

The circumference of the spike at 5.0 cm from the point of attachment to the stem was recorded as the spike girth and expressed in centimetres.

## ***B. Floret characters***

### **1. Number of florets per spike**

The number of florets per spike in each plant was recorded.

### **2. Internodal length**

The length between the nodes of the base and top florets was recorded and expressed in centimetres.

### **3. Pedicel length (Individual flower stalk length)**

Stalk length of individual floret was recorded and expressed in centimetres.

### **4. Length of floret**

Length of individual floret was recorded (vertically) and expressed in centimetres.

### **5. Breadth of floret**

The breadth of individual floret was recorded (across) and expressed in centimetres.

### **6. Length of labellum**

Length of the labellum (lip) was observed and expressed in centimetres.

### **7. Width of labellum**

Width of the labellum (lip) was observed and expressed in centimetres.

## ***3.1.5.2 Phenological characters***

### ***3.1.5.2.1 Plant characters***

#### **1. Interval of leaf production**

The interval between the productions of two successive leaves was observed and expressed in days.

## **2. Leaf longevity (Days)**

Time taken from leaf emergence to its complete turning of pale green and stopped growing at a point was recorded as leaf longevity in days.

### **3.1.5.2.2 *Floral characters***

#### **1. Days from spike emergence to the opening of the first floret**

Time was taken for the opening of the first floret after spike emergence was recorded. Flower buds were labelled and observed at hourly intervals and opening of the first floret was recorded in days.

#### **2. Days from spike emergence to harvest**

Time taken for the opening of 75 per cent of the flowers after spike emergence was recorded in days.

#### **3. Days from spike emergence to complete opening of florets**

Time taken for complete (100 %) opening of all the florets on the spike was recorded in days.

#### **4. The longevity of spike on the plant**

Longevity was measured from the day the spike becomes suitable for use as a cut flower to wilting of one floret and expressed in days.

#### **5. Life of individual floret on the spike**

Life of individual floret on the spike was measured for six florets per spike from the day the floret opened to the day it wilted and the mean value expressed in days.

### **3.1.5.3 *Post-harvest characters***

Post-harvest studies were conducted in the thirty hybrids selected for morphological studies. Spikes were harvested when 50 to 75 per cent flowers were opened. The harvested spikes were kept in tap water for vase studies and following characters were observed during the vase study.



### **1. Fresh weight of spike (g)**

Fresh weight of the cut spike was taken immediately after harvest and recorded in grams.

### **2. Days for wilting of the first floret**

Days taken for wilting of the first floret from the day of harvest was noted.

### **3. Life span of floret**

Life of individual floret in the vase was measured for four florets per spike from the day of the harvest till the day it wilted in a vase and mean values were expressed in days.

### **4. Spike longevity**

Time was taken from the harvest of the spike till it remained fresh without wilting, shrivelling or drooping and expressed in days.

### **5. Water uptake**

The quantity of vase solution remaining at the end of the experiment was recorded and by finding the difference between the initial and final volumes of the vase solution, total uptake was worked out and expressed in millilitres.

### **6. Physiological loss in weight (PLW)**

The loss in weight of the spike in vase was recorded by deducting the weight at the end of the experiment from the initial fresh weight of the spike and expressed in grams.

#### ***3.1.5.4 Qualitative morphological characters***

Qualitative characters were recorded in the selected thirty intergeneric hybrids/varieties by observing and measuring plant and floral parts. Description for qualitative characteristics used for grouping hybrids/varieties (PPV & FRA 2012, PPV & FRA 2012a and De *et al.*, 2018) is given in Appendix XXV.

### **3.1.5.4.1 Plant characters**

#### **A. Shoot characters**

- 1. Nature of growth-** hanging, prostrate
- 2. Nature of shoot-** medium sized (slightly sturdy), sturdy or slender
- 3. Shoot colour-** brown, dark brown or greenish brown
- 4. Branching of the shoot-** present, absent

#### **B. Leaf characters**

- 1. Shape-** terete, semi-terete, strap, linear, quarter terete, deeply channelled
- 2. Texture-** smooth, verrucose, rigid, leathery, glabrous, pubescent, fleshy
- 3. Margin-** entire, wavy, serrate, coriaceous
- 4. Apex-** acute, obtuse, emarginated (bilobed or forked), truncate
- 5. Leaf colour-** green, dark green, dark green with reddish-purple underneath
- 6. Pigmentation-** colour changes during maturity
- 7. Orientation-** straight, arching, deflexed, horizontal
- 8. Nature of sheath-** membranous, thick, soft, nerved or not
- 9. Colour of sheath-** green, dark green
- 10. Leaf base-** sheathed, keeled, sessile

#### **C. Root characters**

- 1. Root origin -** basal, along the stem
- 2. Branching of roots -** present, absent
- 3. Colour of roots -** grey, light green, white
- 4. Nature of roots -** cylindrical, thread like, shrivelled, creeping, robust or flattened

### 3.1.5.4.2 *Floral characters*

#### *A. Spike characters*

1. **Blooming period** - spike emergence in each hybrid was observed throughout the year and recorded as the blooming period for that particular hybrid.
2. **Spikes per plant at a time** - single, double, triple or multiple
3. **Spike orientation**- erect, horizontal, drooping
4. **Nature of inflorescence**- dense, lax
5. **Spike colour** - green, brown
6. **Colour of inflorescence** - predominant colour was observed and recorded
7. **The orientation of florets on a spike**- facing in one direction, facing in two directions, facing in all directions

#### *B. Floret characters*

1. **Colour of florets** - recorded as per Royal Horticultural Society colour chart
2. **Pigmentation**- present, absent
3. **Fragrance**- present, absent

#### *C. Petal characters*

1. **Petal shape**- linear, oblong, ovate, elliptic, lanceolate, obovate, orbicular
2. **Petal curvature**- incurved with deflexed apex, incurved with straight apex, straight, deflexed, deflexed with an incurved apex, incurved
3. **Petal apex**- acute, obtuse, truncate, bilobed
4. **Petal margin** - entire, erose, undulate, deeply undulate
5. **Petal base colour**-single, double, triple or multiple
6. **Petal margin colour**-single, double, triple or multiple
7. **Petal apex colour** - single, double, triple or multiple

- 8. Petal colour pattern-** uniform, spotted, blotched, streaked/striped, tessellated

***D. Lebellum or lip characters***

- 1. Lip shape at the apical lobe region-** ovate, lanceolate, orbicular
- 2. Nature of apical lip-** straight, deflexed with straight apex, deflexed with the incurved apex
- 3. Lip shape at the lateral-lobe region-** obtriangle, oblanceolate, suborbicular, semicircular, oblong
- 4. Nature of lip-** incurved, strongly incurved
- 5. Lip apex-** acute, obtuse, bilobed
- 6. Lip surface-** glabrous, pubescent, leathery, rigid
- 7. Lip base colour-** single, double, triple or multiple
- 8. Lip margin colour-** single, double, triple or multiple
- 9. Lip apex colour-** single, double, triple or multiple
- 10. Lip colour pattern-** uniform, spotted, blotch, streaked/ striped, tessellated

***E. Column characters***

- 1. Column colour-** single, double, triple or multiple
- 2. Column colour pattern-** uniform, shaded, spotted, blotched, streaked/ striped
- 3. Column length-** short (< 0.5 cm), medium (0.5 to 1.0 cm), long (> 1.0 cm)

***F. Spur characters***

- 1. Spur length -** short (< 0.5 cm), medium (0.5 to 1.0 cm), long (> 1.0 cm)
- 2. Spur type-** saccate, conical, cylindric, tubular

***3.1.5.5 Visual evaluation***

A 9 point hedonic scale was used for this purpose and scoring was done for spike and plant quality.

### **9 point hedonic scale:**

9: Like extremely	6: Like slightly	3: Dislike moderately
8: Like very much	5: Neither like nor dislike	2: Dislike very much
7: Like moderately	4: Dislike slightly	1: Dislike extremely

#### **3.1.5.5.1 Spike for use as a cut flower**

The spikes of the *Ascozentrum* hybrids were visually scored, for use as a cut flower and for the indoor display. The general acceptability was observed. Scoring was done based on floret colour & pigmentation, texture, shape and pattern, size and orientation of floret, spike longevity in vase, compactness and visual appeal.

#### **3.1.5.5.2 Plant for indoor display**

Plant quality rating was done based on growth and fullness, spread and orientation of leaves, spike colour and pigmentation, size and orientation of spike, spike longevity on the plant, visual appeal and general appearance during the growth period.

#### **3.1.5.6 Incidence of pests and diseases**

Incidence of pests and diseases was also observed during the period of investigation and recorded.

### **3.2 COMPATIBILITY STUDIES**

Eleven different hybrids/varieties were selected (Table 3) for compatibility studies. Observations were recorded on anthesis, stigma receptivity, pollen production, pollen viability (fertility) and pollen germination.

#### **3.2.1. Anthesis**

Mature bud in each of the hybrid selected for the study was tagged at full bud stage for observing the time of flower opening. The flower buds were observed at hourly intervals to record fully opened flowers which were used for further pollen study.

### 3.2.2. Stigma receptivity

The flowers of the selected hybrids were pollinated from the day of anthesis in order to study the stigma receptivity period. Pollination was done in alternate days 1<sup>st</sup>, 2<sup>nd</sup>, 4<sup>th</sup>, 6<sup>th</sup>, 8<sup>th</sup>, 10<sup>th</sup>, 12<sup>th</sup> and 14<sup>th</sup> day of anthesis.

**Table 3. List of parent hybrids selected for compatibility studies**

Sr. No.	Variety No.	Hybrid/variety name.
1	V <sub>1</sub>	<i>Ascda</i> . Udomochai
2	V <sub>2</sub>	<i>Ascda</i> . Kultana × <i>V</i> . Bitzs Heartthrob
3	V <sub>3</sub>	<i>Ascda</i> . Sirichi Fragrance
4	V <sub>4</sub>	<i>Vasco</i> . Pine River Blue
5	V <sub>5</sub>	<i>Vasco</i> . Pine River Pink
6	V <sub>6</sub>	<i>Vasco</i> . Aroonsri Beauty
7	V <sub>7</sub>	<i>Vasco</i> . Pine Rivers Fuchsia Delight
8	V <sub>8</sub>	<i>Mok</i> . Calypso Pink
9	V <sub>9</sub>	<i>Mok</i> . Chao Praya Sunset Yellow Spot
10	V <sub>10</sub>	<i>Mok</i> . Sayan × <i>Ascda</i> . Doung Porn
11	V <sub>11</sub>	<i>Kag</i> . Youthong Beauty

### 3.2.3. Pollen Studies

Eleven hybrids/varieties were used for estimation of pollen production per pollinium, pollen fertility and pollen germination.

#### 3.2.3.1. Pollen production

The number of pollen grains per pollinium was estimated with haemocytometer. Matured pollinia were gathered from fully opened flowers. Pollen counts were taken as suggested by Rao and Khader (1962).

In a glass vial containing the pollina, 1.25 ml of water containing 1.0 per cent extran was added and the contents were shaken thoroughly. To this, 1.25 ml of glycerine was added. The pollinia were crushed with the edge of the glass rod in order to disperse all the pollen grains properly. Ten microliter of the 2.5 ml of suspension drawn in a fine pipette was transferred to each of two counting chambers of a haemocytometer and observed under the phase contrast microscope. Pollen grains in each of the four corner square in both the counting chambers were counted under low power of a microscope and the mean number in eight corner square was calculated for each hybrid and three such estimates were made. The counting chambers were 0.1 mm in depth and 0.1 mm<sup>3</sup> volume for each chamber.

The number of pollen per pollinium was calculated using the following formula.

If, N = Average number of pollen grains counted per corner square

X = Number of pollen per pollinia

N : X = 0.1 : 2500

0.1 X = 2500 N

X = 25000 N

### **3.2.3.2. Pollen viability (fertility)**

The fertility of pollen grains was estimated by acetocarmine staining technique. Pollen grains were dispersed in a drop of acetocarmine glycerin medium on a clean microscopic slide for 10 to 30 minutes for proper staining and examined under low and high power of Motic image analyser (BA210). Pollen fertility was estimated by counting fertile and sterile pollen grains separately. Pollen grain which stained well, looked plumpy and well-shaped were considered as fertile and those unstained, small or shrivelled as sterile or non viable (Zirkle, 1937). The observations were made on five different microscopic fields of one slide. Such estimate was made in three different slides. The mean percentage of viable pollen grain then calculated.was

### **3.2.3.3. Pollen germination**

*In vitro* germination of pollen grain was studied in artificial media with the selected eleven hybrids.

The medium was fixed following previous studies for pollen germination (2 % sucrose and 1 % agar). Sucrose-agar medium in the above concentration was then prepared in combination with 75 ppm boric acid and hanging drop technique was performed. This was kept for incubation of 12 to 14 hrs in the moist climate. The mean percentage of pollen germination was then calculated. The study was examined under motic image analyser (BA210) and the experiment was replicated thrice.

### **3.2.4. Self-compatibility**

Selfing was conducted in the eleven selected hybrids (Table 3). Self-compatibility was assessed in the selected hybrids/varieties and post pollination changes recorded accordingly.

### **3.2.5. Cross compatibility**

Two hybrids were selected on the basis of pollen fertility/germinations and used as male parents for crossing, *viz.*, *Ascda*. Sirichi Fragrance and *Mok*. Sayan × *Ascda*. Doung Porn. These male parents were used for crossing with selected hybrids/varieties. The crossing attempted in order to study the cross compatibilities between parents are presented in Table 4.

### **3.2.6. Post pollination changes**

#### **3.2.6.1. Flower fall**

The pollinated flowers were carefully observed and setting of pods or flower fall was recorded.

#### **3.2.6.2. Enlargement of pedicel**

Enlargement of pedicel was observed and recorded.



**Table 4. Cross combinations using selected two male parents**

Sl. No.	Crosses made	
	<i>Ascda. Sirichi Fragrance</i> (♂)	<i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i> (♂)
1	<i>Ascda. Udomochai</i> × <i>Ascda. Sirichi Fragrance</i>	<i>Ascda. Udomochai</i> × <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i>
2	<i>Ascda. Kultana</i> × <i>V. Bitzs Heartthrob</i> × <i>Ascda. Sirichi Fragrance</i>	<i>Ascda. Kultana</i> × <i>V. Bitzs Heartthrob</i> × <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i>
3	<i>Vasco. Pine River Blue</i> × <i>Ascda. Sirichi Fragrance</i>	<i>Ascda. Sirichi Fragrance</i> × <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i>
4	<i>Vasco. Pine River Pink</i> × <i>Ascda. Sirichi Fragrance</i>	<i>Vasco. Pine River Blue</i> × <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i>
5	<i>Vasco. Aroonsri Beauty</i> × <i>Ascda. Sirichi Fragrance</i>	<i>Vasco. Pine River Pink</i> × <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i>
6	<i>Vasco. Pine Rivers Fuchsia Delight</i> × <i>Ascda. Sirichi Fragrance</i>	<i>Vasco. Aroonsri Beauty</i> × <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i>
7	<i>Mok. Calypso Pink</i> × <i>Ascda. Sirichi Fragrance</i>	<i>Vasco. Pine Rivers Fuchsia Delight</i> × <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i>
8	<i>Mok. Chao Praya Sunset Yellow Spot</i> × <i>Ascda. Sirichi Fragrance</i>	<i>Mok. Calypso Pink</i> × <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i>
9	<i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i> × <i>Ascda. Sirichi Fragrance</i>	<i>Mok. Chao Praya Sunset Yellow Spot</i> × <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i>
10	<i>Kag. Youthong Beauty</i> × <i>Ascda. Sirichi Fragrance</i>	<i>Kag. Youthong Beauty</i> × <i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i>

### 3.2.6.3. Pod development

Development of pod was observed and recorded.

### 3.2.6.4. Percentage of pod set

Percentage of pod set per spike per hybrid/variety was recorded.

### 3.2.6.5. Days taken for maturity

Number of days was recorded from pollination to the full maturity of pods.

### 3.2.6.6. Seed germination (%)

Seeds were extracted from the matured pod and were cultured under *in vitro* condition and the germination was recorded and expressed in per cent.

### 3.2.6.7. Days for planting out

A number of days from inoculation to deflasking was recorded as days for planting out.

### 3.3. MOLECULAR CHARACTERISATION

Molecular characterisation was done in twenty selected hybrids belonging to different intergeneric groups of *Ascocentrum*. Total genomic DNA was isolated from orchid leaves using procedure suggested by Lim *et al.* (1997). The list of twenty selected hybrids is given in Table 5.

The genomic DNA was amplified with 50 primers (ISSR and SSR) so as to amplify ISSR and SSR regions. The amplicons were separated on an agarose gel and minimum 10 primers were selected based on good amplification. The selected primers were further used for molecular assays. The DNA samples of 20 selected orchid hybrids was subjected to SSR and ISSR assays. The potential of SSR and ISSR primers was used for classification of hybrids. The list of SSR and ISSR primers used for the study is given (Table 6 and Table 7).

The products were gel electrophoresed to observe possible polymorphism. The gel profiles of all accessions were scored for presence/absence of band for further analysis. The extent of variability/similarity among the varieties was summarised in a matrix using MVSP-A (multivariate statistical package 3.22) software. The morphological data were compared with molecular data for grouping the varieties.

#### 3.3.1 Laboratory chemicals

The AR (analytical reagents) grade chemicals (extra pure) from Sisco Research Laboratories (SRL) were used in this study. The constituents for PCR reaction mixture *viz.*, Taq buffer (A and B), MgCl<sub>2</sub>, dNTPs, Taq DNA polymerase, *etc.* used in this study were procured from Genei Pvt. Ltd., Bangalore. The SSR (Simple Sequence Repeats) primers synthesised by Sigma Aldrich Chemicals Pvt. Ltd., Bangalore were used. Details about equipments and the plastic wares used are given in Appendix XXVII.

**Table 5. Intergeneric hybrids of *Ascozentrum* selected for molecular characterization**

<b>Sr. No.</b>	<b>Var. No.</b>	<b>Hybrid/variety name.</b>
1	V <sub>1</sub>	<i>Kag.</i> Christie Low
2	V <sub>2</sub>	<i>Mok.</i> Omayaiy Yellow
3	V <sub>3</sub>	<i>Mok.</i> Sunspot
4	V <sub>4</sub>	<i>Vasco.</i> Pine River Pink
5	V <sub>5</sub>	<i>Ascda.</i> Sirichi Fragrance
6	V <sub>6</sub>	<i>Ascda.</i> Udomochai
7	V <sub>7</sub>	<i>Vasco.</i> Blue Bay White
8	V <sub>8</sub>	<i>Vasco.</i> Pine Rivers Fuchsia Delight
9	V <sub>9</sub>	<i>Ascda.</i> Kultana × <i>V.</i> Bitzs Heartthrob
10	V <sub>10</sub>	<i>Kag.</i> Samrong
11	V <sub>11</sub>	<i>Mok.</i> Sayan × <i>Ascda.</i> Bangkuntein Gold
12	V <sub>12</sub>	<i>Mok.</i> Khaw Phiak Suan × <i>Ascda.</i> Jiraprapa
13	V <sub>13</sub>	<i>Ascda.</i> Suksamran Sunlight
14	V <sub>14</sub>	<i>Ascda.</i> Yip Sum Wah × <i>V.</i> Josephine Van Brero
15	V <sub>15</sub>	<i>Vasco.</i> Aroonsri Beauty
16	V <sub>16</sub>	<i>Mok.</i> Walter Oumae Pink
17	V <sub>17</sub>	<i>Mok.</i> Rassmatozz
18	V <sub>18</sub>	<i>Mok.</i> Calypso × <i>V.</i> Doctor Anek
19	V <sub>19</sub>	<i>Mok.</i> Khaw Phiak Suan × <i>Ascda.</i> Bicentennial Yellow Spot
20	V <sub>20</sub>	<i>Mok.</i> Chao Praya Sunset Yellow Spot

**Table 6. List of SSR primers (with their forward and reverse sequences) used in the study**

Sl. No.	Primer name	Sequence	AT (°C)	Reference
1	FJ539050	F: AATGGACCTTCTTTGCATTAC R: ATTACCGTTCATTCTGGTGC	46.0	Phuekvilai <i>et al.</i> , 2009
2	FJ539051	F: AAGTCTAGCTTTTGGTTGAGG R: ATCGATGGTTTGTCTTCTAGC	55.1	Phuekvilai <i>et al.</i> , 2009
3	FJ539052	F: TCATTGATGTTGGGAGCCTAA R: CTTGCCCTCTATCTTCTCTT	50.0	Phuekvilai <i>et al.</i> , 2009
4	FJ539053	F: AGAACTAGATGACTTCAAAACG R: GAA CTCAGAAAAATTACCGCG	47.0	Phuekvilai <i>et al.</i> , 2009
5	FJ539054	F: TGG AAATGCATGTTGCCCGA ACT GAG TGA CCT TGG AAG AC	60.2	Phuekvilai <i>et al.</i> , 2009
6	FJ539055	F: CTTTGAGTAATGTCTCTCAGTG R: CCCTCACGCACTCTCTACC	45.0	Phuekvilai <i>et al.</i> , 2009
7	FJ539056	F: AGAATGAGGGAGGTATAGGG R: TGCCTTGGATGTGCGTTCCG	52.0	Phuekvilai <i>et al.</i> , 2009
8	FJ539057	F: TTCAGCGTTTCCATGTCGAAG R: AGTAAAGCCGCCATCTTGG	52.0	Phuekvilai <i>et al.</i> , 2009
9	FJ539058	F: AGAGTGAAGAGAGTGTGG R: GGACTGTAAACTTCATGAGC	50.2	Phuekvilai <i>et al.</i> , 2009
10	FJ539059	F: AGAATGCCACAATATCATCACC R: CTGTGTCTGTTTCTATTTATGTG	48.0	Peyachoknagul <i>et al.</i> , 2014
11	FJ539060	F: TCTAGACATGTTTGAGAGGTGC R: TTAATCTTCCACTCTTCCATCC	56.0	Peyachoknagul <i>et al.</i> , 2014
12	FJ539061	F: CGCCCAACGAATAGAATGTTGG R: ACTATCTTCCTTACTCTTGCCCTC	56.0	Peyachoknagul <i>et al.</i> , 2014a
13	DQ494847	F: TAGTGCCATCTAATCTAATG R: TTTTCTTGTGCTCGAAG	48.3	Xia <i>et al.</i> , 2008
14	DQ501382	F: GTGAAAGCCACCTCCATG R: GATGGATACCTCGCACTGG	48.0	Xia <i>et al.</i> , 2008
15	DQ501383	F: AGTTGCGGGTCAGTGTAAC R: TTAAGCAGGAGCCGTCACAG	50.0	Xia <i>et al.</i> , 2008
16	DQ501384	F: CAGATGGATACCTCGCACTG R: TAACTTCCCAGGTTTAC	47.0	Xia <i>et al.</i> , 2008
17	DQ501385	F: CAGAGCAGCGGACATCA R: CCGCATACATGTTACAAGTC	55.3	Xia <i>et al.</i> , 2008
18	DQ501387	F: CGCCAGCCCTGTTAGGA R: TAGACTGGTGAGGCGTCAAG	59.1	Xia <i>et al.</i> , 2008
19	JN375718	F: CCCAACATTTGCAAGTCATC R: GAGATTTGTTGCCATTAC	48.0	Lopez Roberts <i>et al.</i> , 2012
20	JN375723	F: GAGAGCTGTCTTTTCTTGA R: GCACTAAAACTCCTGTT	54.0	Lopez Roberts <i>et al.</i> , 2012
21	JN375713	F: GGATTCCCTGACAAGTTGGA R: GGTCTCTGTTCCCAAAATGA	58.9	Lopez Roberts <i>et al.</i> , 2012

**Table 7. List of ISSR primers used in the study**

Sl. No.	Primer name	Primer sequence (5'-3')	AT (°C)	Reference
1	7	CTCTCTCTCTCTCTRG	54.0	Moraes <i>et al.</i> , 2014
2	901	GTGTGTGTGTGTYR	54.0	Moraes <i>et al.</i> , 2014
3	17899A	CACACACACACAAC	48.0	Parab, 2008
4	(GACAC)4	GACACGACACGACACGACAC	52.0	Parab, 2008
5	ACTG(4)	ACTGACTGACTGACTG	52.0	Parab, 2008
6	(CT)10G	CTCTCTCTCTCTCTCTCTG	60.0	Parab, 2008
7	(CT)10A	CTCTCTCTCTCTCTCTCTA	55.0	Parab, 2008
8	(TC)10G	TCTCTCTCTCTCTCTCTCG	62.0	Parab, 2008
9	AW3	GTGTGTGTGTGTRG	54.0	Moraes <i>et al.</i> , 2014
10	DAT	GAGAGAGAGAGAGARG	54.0	Moraes <i>et al.</i> , 2014
11	GOOFY	GTGTGTGTGTGTGTYG	56.0	Moraes <i>et al.</i> , 2014
12	HB 8	GAGAGAGAGAGAGG	59.0	Parab, 2008
13	HB 9	GTGTGTGTGTGTGG	52.0	Parab, 2008
14	MANNY	CACCACCACCACRC	52.0	Moraes <i>et al.</i> , 2014
15	MAO	CTCCTCCTCCTCRC	45.0	Moraes <i>et al.</i> , 2014
16	OMAR	GAGGAGGAGGAGRC	54.3	Moraes <i>et al.</i> , 2014
17	UBC 807	AGAGAGAGAGAGAGAGT	53.0	Dharmarathna <i>et al.</i> , 2018
18	UBC 808	AGAGAGAGAGAGAGAGC	54.0	Dharmarathna <i>et al.</i> , 2018
19	UBC 809	AGAGAGAGAGAGAGAGG	53.5	Dharmarathna <i>et al.</i> , 2018
20	UBC 810	GAGAGAGAGAGAGAGAT	50.4	Dharmarathna <i>et al.</i> , 2018
21	UBC814	CTCTCTCTCTCTCTTG	52.0	Moraes <i>et al.</i> , 2014
22	UBC830	TGTGTGTGTGTGTGG	52.0	Dharmarathna <i>et al.</i> , 2018
23	UBC 841	GAGAGAGAGAGAGAGACC	53.0	Dharmarathna <i>et al.</i> , 2018
24	UBC843	CTCTCTCTCTCTCTRA	54.0	Moraes <i>et al.</i> , 2014
25	UBC844	CTCTCTCTCTCTCTRC	54.8	Moraes <i>et al.</i> , 2014
26	UBC855	ACACACACACACACACYT	53.0	Dharmarathna <i>et al.</i> , 2018
27	UBC 858	TGTGTGTGTGTGTGRT	54.0	Dharmarathna <i>et al.</i> , 2018
28	UBC873	GACAGACAGACAGACA	54.0	Dharmarathna <i>et al.</i> , 2018
29	UBC899	CACACACACACARG	55.0	Moraes <i>et al.</i> , 2014

### 3.3.2 DNA isolation

#### 3.3.2.1 Reagents used

- I. Liquid nitrogen
- II. Polyvinylpyrrolidone (PVP)
- III. Extraction buffer
  - 100 mM Tris-HCl, pH 8.0
  - 50 mM EDTA, pH 8.0
  - 500 mM NaCl
  - 100 mM mercaptoethanol
- IV. Sodium dodecyl sulphate (20 %)
- V. Potassium acetate (pH 5.2) 5 M
- VI. Isopropanol (100 %)
- VII. Tris EDTA buffer (10 mM Tris-HCl and 1 mM EDTA, pH 8.0)
- VIII. Chloroform: Isoamyl alcohol (24:1 v/v)
- IX. Sodium acetate 3 M (pH 5.2)
- X. Ethanol (70 % and 100 %)
- XI. Sterile autoclaved distilled water

#### 3.3.2.2 Procedure for extraction of genomic DNA

The DNA was isolated by following the protocol of Lim *et al.* (1997). The young leaves were collected from the varieties/ hybrids maintained in the greenhouse. The extraction of genomic DNA was done using the following protocol.

1. Tender leaves were collected and 0.1 g sample was ground to a fine powder in liquid nitrogen using pre-chilled autoclaved mortar and pestle with a pinch of polyvinylpyrrolidone (PVP).
2. Then 600  $\mu$ l extraction buffer was added followed by 40  $\mu$ l of 20 per cent sodium dodecyl sulphate.
3. Homogenised samples were transferred to autoclaved 2 ml centrifuge tube.

4. The tubes were inverted a few times to mix the contents and incubated at 65 °C in water-bath for 10 min. with gentle inversion once.
5. After incubation, the tubes were taken out and one-tenth volume of 5 M potassium acetate (pH 5.2) was added and the mixture was incubated at -20 °C for 20 min.
6. The contents then centrifuged at 10,000 rpm for 20 min. at 4 °C.
7. After centrifugation, the contents got separated into two distinct layers.
  - Aqueous topmost layer: contained DNA and RNA
  - Lower layer: salted tissue debris
8. The supernatant was decanted in 1.5 ml centrifuge tube and 0.4 cm<sup>3</sup> ( $\approx$ 400  $\mu$ l) of isopropanol was added and the tubes were incubated at -20 °C for one hour to precipitate the DNA.
9. The mixture was then centrifuged at 10000 rpm for 15 min. at 4 °C to pellet the DNA.
10. Then, the supernatant was discarded and the pellet was then resuspended in 0.2 cm<sup>3</sup> ( $\approx$ 200  $\mu$ l) Tris EDTA buffer and 1  $\mu$ l of RNase was added and the mixture was incubated at 37 °C for 30 min.
11. After this 200  $\mu$ l of chilled chloroform: isoamyl alcohol (24:1) was added, inverted to mix and emulsified followed by centrifugation at 10000 rpm for 10 min. at 4 °C.
12. After centrifugation, the contents got separated into three distinct layers.
  - Aqueous topmost layer: containing DNA
  - Interphase: contained fine particles and proteins
  - Lower layer: contained chloroform and some pigments
13. Aqueous topmost layer was removed and chloroform: isoamyl alcohol (24:1) wash was repeated.

14. The aqueous phase was removed and 0.1 volume of 3 M sodium acetate (pH 5.2) and 2.5 volumes of 100 per cent ethanol was added and the mixture was left to incubate at -20 °C for one hour.
15. The DNA was pelleted by centrifugation at 10,000 rpm for 10 min. at 4 °C.
16. The pellet was washed with 70 and 100 per cent ethanol and dried in laminar airflow.
17. The pellet was then dissolved in 50 µl sterile autoclaved DH<sub>2</sub>O by gentle tapping to dissolve pellet and then the DNA samples were stored at -20 °C.

### **3.3.3 Quality and quantity estimation of DNA using spectrophotometer**

The purity and quantity of the DNA were estimated using a Nanodrop Spectrophotometer (Jenway- Genova Nano). Since the absorption maxima for nucleic acids and proteins were at 260 and 280 nm respectively, absorbance was recorded at both the wavelengths and purity of the sample was estimated using the OD<sub>260</sub>/OD<sub>280</sub> ratio. The DNA sample was considered to be pure if the OD<sub>260</sub>/OD<sub>280</sub> value is between 1.8 and 2.0. Values below 1.8 and above 2.0 were due to contamination by protein and RNA, respectively. The concentration of DNA in the sample was estimated using the relation, 1 OD at 260 nm = 50 ng DNA/µl, hence, OD<sub>260</sub>×50 gave the quantity of DNA (ng/µl).

#### **3.3.3.1 Procedure**

1. The lid of spectrophotometer was opened followed by the sampling arm, and the pedestal was wiped with tissue paper to remove any dust particles.
2. The reading was set to zero with a blank sample (DDH<sub>2</sub>O which used to dissolve the DNA pellet).
3. Then, 1 µl of the test sample was loaded on to the pedestal and measure option was selected and the necessary readings were recorded.
4. After the measurements, the pedestal was wiped clean with 70 per cent ethanol using a soft laboratory wipe.



### **3.3.4 Agarose gel electrophoresis**

#### **3.3.4.1 Reagents used**

- I. Agarose (0.8 %)
- II. 50X TAE buffer (pH 8.0)
  - Tris buffer (1 M)
  - Glacial Acetic acid
  - 0.5 M EDTA
- III. Tracking/loading dye (6X)
- IV. Ethidium bromide (stock 10 mg /ml, working concentration 0.5 µg/ml)

#### **3.3.4.2 Procedure**

1. The gel casting tray was placed appropriately in a gel caster and the movable wall was adjusted such that the gel casting tray was closed at both ends. A comb was selected depending on the number of samples to be electrophoresed and positioned on the grooves provided on the gel casting tray.
2. The gel was prepared by adding 0.8 g of agarose in 100 ml of 1X TAE buffer in a glass conical flask. The mixture was heated in a microwave oven until all the agarose particles were completely dissolved and a clear solution was obtained.
3. Then the solution was allowed to cool down to 40 to 50 °C and an appropriate amount (1 µl per 10 ml of gel) of ethidium bromide was added and mixed well. The warm gel was then poured into the gel casting tray and left to solidify for 20 minutes at room temperature.
4. Special care was taken to avoid any air bubbles near the wells or on the gel.
5. Once the gel was solidified, a small amount of 1X TAE was poured on top of the gel and the comb was removed carefully without breaking the gel. The TAE solution was discarded and the gel along with the tray was kept inside the electrophoresis tank with the wells on the negative electrode side.

6. The electrophoresis tank was filled with 1X TAE sufficient enough to submerge the wells.
7. The samples to be electrophoresed were prepared by mixing 5  $\mu$ l of the DNA sample with 1  $\mu$ l of 6X gel loading dye. After mixing, the total volume of 6  $\mu$ l was loaded into individual wells.
8. The samples were electrophoresed at 75 volts until gel tracking dye reached two-third of the gel length.

#### **3.3.4.3 Gel documentation**

Documentation of the electrophoresed gel was done under UV with a gel documentation system (Gel Doc, Uvitech, Cambridge) using Fire Reader software.

#### **3.3.5 Preparation of reaction mixture for thermal cycling**

The reaction mixture consisted of template DNA, Taq buffer (A and B),  $MgCl_2$ , forward and reverse SSR primer/ISSR primer, dNTPs,  $DDH_2O$  and Taq DNA polymerase. The desired number of PCR cycles, time and temperature for denaturation, annealing (AT) and extension were standardised based on the primers used and the conditions were programmed and saved in the thermal cycler (model- Veriti 96 Well Thermal Cycler, made: Applied Biosystems and model- Master Gradient, made: Eppendorf).

##### **3.3.5.1 Thermal cycling**

1. PCR microcentrifuge tubes (0.2 ml) were numbered from 1 to 20.
2. Added 2  $\mu$ l of template DNA from individual hyb. was added to each tube.
3. Added 18  $\mu$ l of the master mix was added to all the tubes and was given a short spin to mix the contents.
4. Thermal cycling was carried out with 20  $\mu$ l reaction mixture. The composition of the reaction mixture for SSR is as follows:

Genomic DNA (25 ng/ $\mu$ l)	2 $\mu$ l
------------------------------	-----------

10X Taq buffer A	2.7 $\mu$ l
dNTP mix (2.5 mM of each)	1.5 $\mu$ l
Primer (10 pM)	1.5 $\mu$ l each of forward and reverse primer
Taq DNA polymerase (3 Units)	0.4 $\mu$ l
Chilled autoclaved distilled water	10.4 $\mu$ l
<b>Total</b>	<b>20 <math>\mu</math>l</b>

5. Thermal cycling was carried out with 20  $\mu$ l reaction mixture. The composition of the reaction mixture for ISSR as follows:

Genomic DNA (25 ng/ $\mu$ l)	2 $\mu$ l
10X Taq buffer A	3 $\mu$ l
MgCl <sub>2</sub> (25 mM)	0.7 $\mu$ l
dNTP mix (2.5 mM of each)	1.5 $\mu$ l
Primer (10 pM)	1.5 $\mu$ l primer
Taq DNA polymerase (3 Units)	0.4 $\mu$ l
Chilled autoclaved distilled water	10.9 $\mu$ l
<b>Total reaction volume</b>	<b>20<math>\mu</math>l</b>

6. The tubes were placed in the thermal cycler for 35 cycles of PCR. The PCR programme followed was

94 °C for 4 min.	Initial denaturation	
94 °C for 45 sec.	Denaturation	} 35 cycles
50 °C to 55 °C for 1 min.	Primer annealing	
72 °C for 2 min.	Primer extension	
72 °C for 8 min.	Final extension	
4 °C hold for infinity	Storage	

- I. Samples were held at 4 °C in the thermal cycler followed by storage at -20 °C until the contents were loaded on to the gel for electrophoresis.
- II. The PCR amplified products were electrophoresed on 2 per cent agarose gel and 1.8 per cent agarose gel for SSR and ISSR, respectively at 70 volts. A ProxiO 100 bp DNA Ladder Plus (SRL) was used. The gel profile was visualised under UV and was saved for further analysis.

### **3.3.6 Scoring of primers for all genotypes**

The gel profiles of individual SSR and ISSR primer were carefully observed and scored and this data was used for further analysis.

### **3.3.7 Molecular weight analysis**

The analysis of the molecular weight of PCR images was done by using Navigating 1D MAX software, UVITECH Cambridge.

## **3.4 STATISTICAL ANALYSIS**

### **3.4.1 Morphological data**

The data on quantitative plant characters and post-harvest characters were subjected to analysis of root square transformation using Web Agri Stat Package 2.0, an online-based tool. The data on quantitative floral characters was data subjected to one way ANOVA using OPSTAT (online-based software developed by CCS HAU, Hisar) and the cluster analysis was performed using Minitab V18.

### **3.4.2 Molecular data**

The data generated from molecular weight analysis of all polymorphic SSR and ISSR primers were compiled together to form a data sheet for cluster analysis. The SSR primers across the 20 genotypes were scored. For the presence of each band 1 code has been used, while, for its absence in another genotype, 0 code has been allotted for each primer.

Pair-wise similarity coefficient matrix was generated by Jaccard's coefficient of similarity by using MVSP-A (multivariate statistical package 3.2). The cluster analysis was performed from the distance matrix using Jaccard's similarity coefficient. Distance matrix and dendrogram were constructed based on diversity coefficient generated from pooled data by using the unweighted pair group method of arithmetic means (UPGMA), a computer programme for distance estimation.

Other parameters *i.e.* PIC (Polymorphic Information Content) was calculated using the following formulae. A PIC value of each primer was determined using PIC calculator (Jan, 2002).

$$PIC = \frac{\text{Total no. of bands} - \text{Highest allelic Frequency}}{\text{Total no. of bands}}$$

$$H_e = 1 - \sum p_i^2$$

Where  $p_i$  represents the frequency of the  $i^{\text{th}}$  allele



**Results**



## 4. RESULTS

The results of the ‘Morpho molecular characterisation of intergeneric hybrids of *Ascocentrum*’ conducted in the Department of Floriculture and Landscaping, College of Horticulture, Kerala Agricultural University, Thrissur during 2016-2019 are presented in this chapter.

### 4.1 MORPHOLOGICAL CHARACTERISATION/FIELD EVALUATION

#### 4.1.1 Quantitative morphological characters

##### 4.1.1.1 Plant characters

The data pertaining quantitative plant characters for two years (Apr. 2016-Mar. 2018) are presented in Appendix I-XXVII and also depicted in Fig. 1-15. The results are described based on the four monthly interval data, and the March 2018 values are given in Table 8 and 9 for the convenience and for easy interpretation.

##### 4.1.1.1.1 *Plant height*

Plant height was recorded at monthly intervals for two years during the period of study (April 2016 to March 2018) and are presented in Appendix I, II and Fig. 1.

High difference was observed with respect to plant height among all the thirty hybrids/varieties studied. The variety V<sub>23</sub> (*Mok. Omayaiy Yellow*) was observed to have the highest plant height during the entire period of study. During the month of April 2016, V<sub>23</sub> (*Mok. Omayaiy Yellow*) recorded maximum plant height (105.8 cm) which was on par with V<sub>25</sub> (98.8 cm) (*Mok. Sayan × Ascda. Dounng Porn*). During the month of August 2016, again V<sub>23</sub> was found significantly superior to all other varieties with respect to plant height (112.40 cm) followed by V<sub>25</sub> (103.26 cm) and V<sub>24</sub> (87.60 cm) respectively. Almost similar result was continued in V<sub>23</sub> throughout the period of study. The plant height for V<sub>23</sub> in the month of December 2016 was 117.40 cm, April 2017 (125.07 cm),

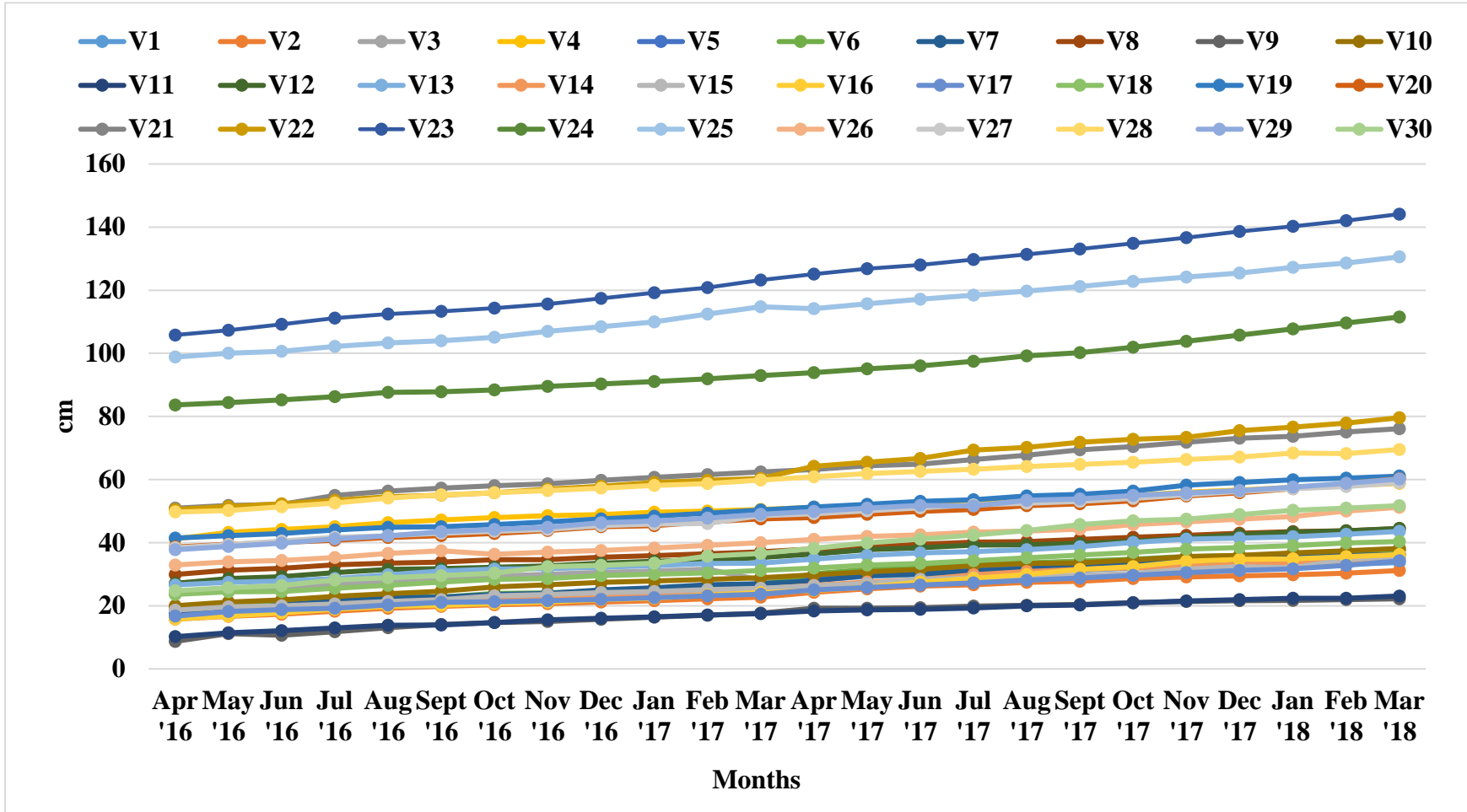


Fig. 1. Plant height of *Ascocentrum* hybrids/varieties during the period 2016-18



August 2017 (131.38 cm), December 2017 (138.59 cm) and March 2018 (144.11 cm) which was followed by V<sub>25</sub> (*Mok. Sayan* × *Ascda. Doung Porn*) [December 2016 (108.40 cm), April 2017 (114.13 cm), August 2017 (119.68 cm), December 2017 (125.45 cm) and March 2018 (130.55 cm)].

**Table 8. Quantitative plant, shoot and root characters of intergeneric hybrids of *Ascocentrum* during March 2018**

Var. No.	Plant height (cm)	Plant spread (cm)	Shoot girth (cm)	Shoot diameter (cm)	Internodal length (cm)	No. of roots	Root length (cm)	Root girth (cm)
V <sub>1</sub>	33.72	47.83	4.57	1.45	2.70	22.00	153.67	2.57
V <sub>2</sub>	31.17	42.00	5.27	1.68	3.17	17.40	166.67	2.77
V <sub>3</sub>	37.85	54.93	6.07	1.93	3.90	16.20	161.00	3.17
V <sub>4</sub>	58.78	41.93	5.30	1.69	2.60	28.80	184.67	2.73
V <sub>5</sub>	36.22	44.17	4.63	1.48	2.57	18.40	165.60	3.03
V <sub>6</sub>	37.63	39.77	5.30	1.69	2.30	22.40	194.33	2.83
V <sub>7</sub>	36.20	37.97	5.63	1.79	2.67	16.60	191.67	3.10
V <sub>8</sub>	44.41	50.23	4.83	1.54	2.43	20.40	189.80	2.77
V <sub>9</sub>	22.21	21.07	3.40	1.08	2.10	10.20	45.23	1.93
V <sub>10</sub>	38.17	49.50	5.03	1.60	2.53	14.40	149.00	2.73
V <sub>11</sub>	23.09	24.43	4.17	1.33	2.37	16.40	121.30	2.23
V <sub>12</sub>	44.51	27.87	5.13	1.63	2.43	14.60	127.00	3.33
V <sub>13</sub>	43.57	47.53	5.30	1.69	2.53	16.20	192.00	2.70
V <sub>14</sub>	34.98	52.43	5.13	1.63	2.50	12.80	193.33	3.10
V <sub>15</sub>	35.48	53.67	5.33	1.70	2.93	15.60	193.67	3.10
V <sub>16</sub>	36.21	43.00	4.77	1.52	3.17	16.00	205.30	2.87
V <sub>17</sub>	34.22	35.67	3.97	1.26	2.37	14.20	153.53	2.57
V <sub>18</sub>	40.34	47.00	4.87	1.55	2.97	12.00	196.20	3.23
V <sub>19</sub>	61.08	37.13	5.90	1.88	4.27	15.40	68.17	3.23
V <sub>20</sub>	60.30	41.23	6.07	1.93	4.40	14.40	73.07	3.93
V <sub>21</sub>	76.12	52.33	5.97	1.90	4.77	14.60	68.40	3.67
V <sub>22</sub>	79.58	58.60	6.07	1.93	4.80	14.40	101.53	3.60
V <sub>23</sub>	144.11	57.93	6.97	2.22	5.13	24.60	114.17	3.53
V <sub>24</sub>	111.50	42.60	6.73	2.14	4.97	16.80	120.00	3.67
V <sub>25</sub>	130.55	40.90	7.10	2.26	5.00	22.20	111.66	3.87
V <sub>26</sub>	51.19	39.80	6.17	1.96	4.20	20.00	57.82	3.73
V <sub>27</sub>	58.98	63.00	5.93	1.89	4.17	15.60	61.09	3.23
V <sub>28</sub>	69.52	60.53	5.70	1.82	4.77	15.80	80.27	3.33
V <sub>29</sub>	60.17	59.73	6.30	2.01	4.13	17.60	124.16	3.13
V <sub>30</sub>	51.73	42.73	4.27	1.36	3.43	19.40	140.27	2.73
<b>C.D.</b>	<b>0.729</b>	<b>0.404</b>	<b>0.139</b>	<b>0.067</b>	<b>0.124</b>	<b>0.479</b>	<b>0.973</b>	<b>0.131</b>
<b>SE(M)</b>	<b>0.26</b>	<b>0.142</b>	<b>0.049</b>	<b>0.024</b>	<b>0.044</b>	<b>0.171</b>	<b>0.343</b>	<b>0.046</b>
<b>C.V.</b>	<b>8.069</b>	<b>3.652</b>	<b>3.355</b>	<b>2.486</b>	<b>3.62</b>	<b>9.028</b>	<b>5.161</b>	<b>3.978</b>

The least plant height was observed during April 2016 in V<sub>9</sub> (*Vasco. Aroonsri Beauty*) (8.7 cm) which was on par with V<sub>11</sub> (10.2 cm), V<sub>2</sub> (15.76 cm), V<sub>16</sub> (15.8 cm), V<sub>14</sub> (16.6 cm), V<sub>17</sub> (16.7 cm), V<sub>1</sub> (17.26 cm) and V<sub>7</sub> (17.60 cm). Similarly, for August 2016 and December 2016 lowest values were recorded in V<sub>9</sub> (*Vasco. Aroonsri Beauty*) (13.00 cm) and (15.70 cm), respectively. However, it was on par with V<sub>11</sub> (*Vasco. Blue Bay White*), V<sub>2</sub> (*Ascda. Kraillerk White* × *V. Sanderiana*), V<sub>16</sub> (*Mok. Khaw Phiak Suan* × *Ascda. Jiraprapa*), V<sub>17</sub> (*Mok. Sayan* × *Ascda. Bangkuntein Gold*), V<sub>14</sub> (*Mok. Calypso* × *V. Dr. Anek*) and V<sub>15</sub> (*Mok. Rassmatozz*) during Dec. 2016. Varieties viz., V<sub>1</sub> (*Ascda. Udomochai*), V<sub>5</sub> (*Ascda. Suksamran Sunlight*) V<sub>6</sub> (*Ascda. Sirichi Fragrance*) and V<sub>7</sub> (*Vasco. Pine River Blue*) were also found to be on par with V<sub>9</sub> (*Vasco. Aroonsri Beauty*).

In the month of April 2017, minimum plant height was observed in V<sub>11</sub> (*Vasco. Blue Bay White*) (19.98 cm), which was on par with V<sub>9</sub> (20.13 cm), V<sub>2</sub> (27.37 cm), V<sub>17</sub> (28.06 cm), V<sub>15</sub> (29.23 cm), V<sub>16</sub> (29.9 cm), V<sub>14</sub> (30.86 cm), V<sub>1</sub> (19.98 cm) and V<sub>7</sub> (19.98 cm). In August 2017, minimum plant height was observed in V<sub>11</sub> (19.98 cm). But, at the end of the observation period in December 2017 and March 2018, the lowest values of plant height were again recorded in V<sub>9</sub> (21.56 cm) and 22.21 cm, respectively). At the end of the study period during Mar. 2018 (Table 8) least plant height was found V<sub>9</sub> (*Vasco Aroonsri Beauty*) which was on par with V<sub>11</sub> (23.09 cm), V<sub>2</sub> (31.17 cm), V<sub>1</sub> (33.72 cm), V<sub>17</sub> (34.22 cm) and V<sub>14</sub> (34.98 cm).

#### **4.1.1.1.2 Plant spread**

Significant differences were observed with regard to plant spread in different *Ascocentrum* hybrids/varieties (Appendix III, IV and Fig. 2). During April 2016, maximum plant spread was recorded in V<sub>27</sub> (57.33 cm) which was on par with V<sub>28</sub> (54.67cm), V<sub>22</sub> (52.66 cm), V<sub>23</sub> (52 cm), V<sub>29</sub> (51.83 cm) and V<sub>15</sub> (46.5 cm), and followed by V<sub>21</sub> (46.33 cm) and V<sub>3</sub> (46.33 cm). The highest value during the month of August 2016 was again observed in V<sub>27</sub> (58.83 cm), which was on par with V<sub>28</sub> (56.23 cm), V<sub>22</sub> (53.77 cm), V<sub>23</sub> (53.4 cm) and V<sub>29</sub> (53.33 cm). Almost a similar kind of result was observed during December. 2016,

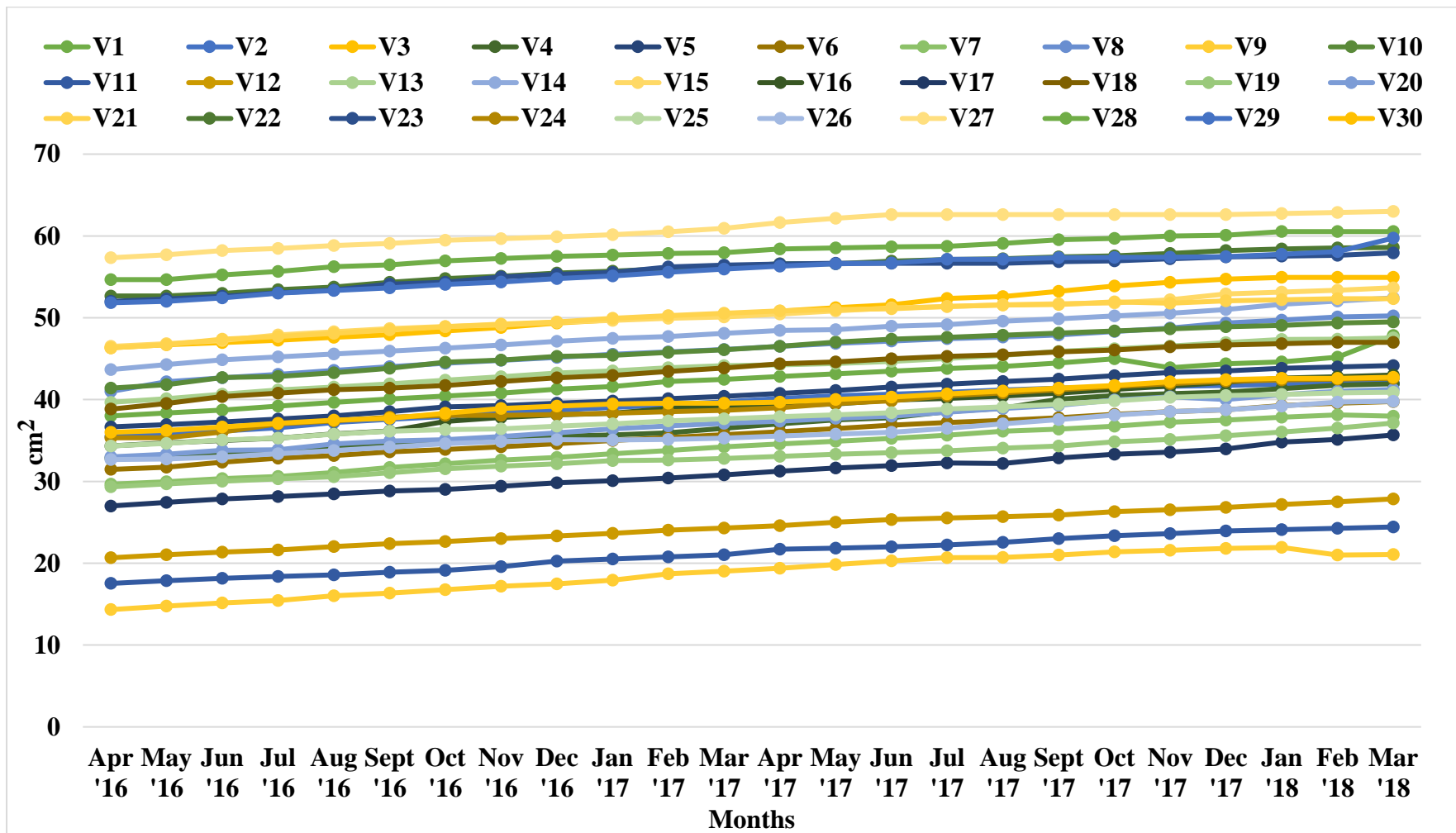


Fig. 2. Plant spread of *Ascozentrum* hybrids/varieties during the period 2016-18

wherein maximum plant spread was noticed in V<sub>27</sub> (59.9 cm), which was on par with V<sub>28</sub> (57.5 cm), V<sub>22</sub> (55.47 cm), V<sub>23</sub> (55.3 cm) and V<sub>29</sub> (54.8 cm), and followed by V<sub>21</sub> (49.5 cm). During April 2017, the highest value was found in V<sub>27</sub> (61.63 cm), which was on par with V<sub>28</sub> (58.4 cm), V<sub>22</sub> (56.6 cm), V<sub>23</sub> (56.57 cm) and V<sub>29</sub> (56.3 cm). Maximum plant spread was recorded in V<sub>27</sub> which was constant during August 2017 (62.6 cm) and December 2017 (62.6 cm) and varied during March 2017 (63.0 cm) however this was on par with V<sub>28</sub> (60.53 cm), V<sub>29</sub> (59.73 cm) V<sub>22</sub> (58.60 cm) and V<sub>23</sub> (57.93 cm).

The minimum plant spread was observed in V<sub>9</sub> (14.33 cm) during April 2016, which was on par with V<sub>11</sub> (17.53 cm). The same trend was noticed in the month of August 2016 also where V<sub>9</sub> (16.03 cm) was on par with V<sub>11</sub> (18.57 cm). During December 2016, the lowest value of plant spread was in V<sub>9</sub> (17.47 cm), which was on par with V<sub>11</sub> (20.27 cm) and V<sub>12</sub> (23.33 cm). The lowest plant spread was again observed in V<sub>9</sub> during April 2017 and August 2017 19.4 cm and 20.7 cm, respectively, which were on par with V<sub>11</sub> (21.7 cm, 22.57 cm) and V<sub>12</sub> (24.6 cm, 25.7 cm) in Apr. 2017 and August 2017, respectively. During December 2017, the lowest value was recorded in V<sub>9</sub> (21.8 cm) and was on par with V<sub>11</sub> (23.93 cm) and V<sub>12</sub> (26.83 cm). Minimum plant spread was observed during the Mar. 2018 in V<sub>9</sub> (21.07 cm), which was on par with V<sub>11</sub> (24.43 cm) followed by V<sub>12</sub> (27.87 cm) (Table 8).

#### **4.1.1.1.3 Shoot girth**

The data regarding the shoot characters of *Ascocentrum* hybrids/varieties are presented in Appendix V, VI and Fig. 3. *Ascocentrum* hybrids/varieties did not exhibit significant variation with respect to shoot girth during most of the months. In April 2016, maximum shoot girth (5.7 cm) was observed in V<sub>23</sub> *Mok. Omayaiy Yellow*, which was on par with V<sub>25</sub> (5.6 cm) and V<sub>24</sub> (5.47 cm). Appreciable differences were not found during August 2016 and December 2016. The maximum values were observed in V<sub>24</sub> (5.7 cm and 5.9 cm, respectively) which were on par with V<sub>25</sub> (5.83 cm and 6.1 cm) and V<sub>23</sub> (5.97 cm and 6.13 cm) for Aug. 2016 and Dec. 2016 respectively. During April 2016, maximum shoot

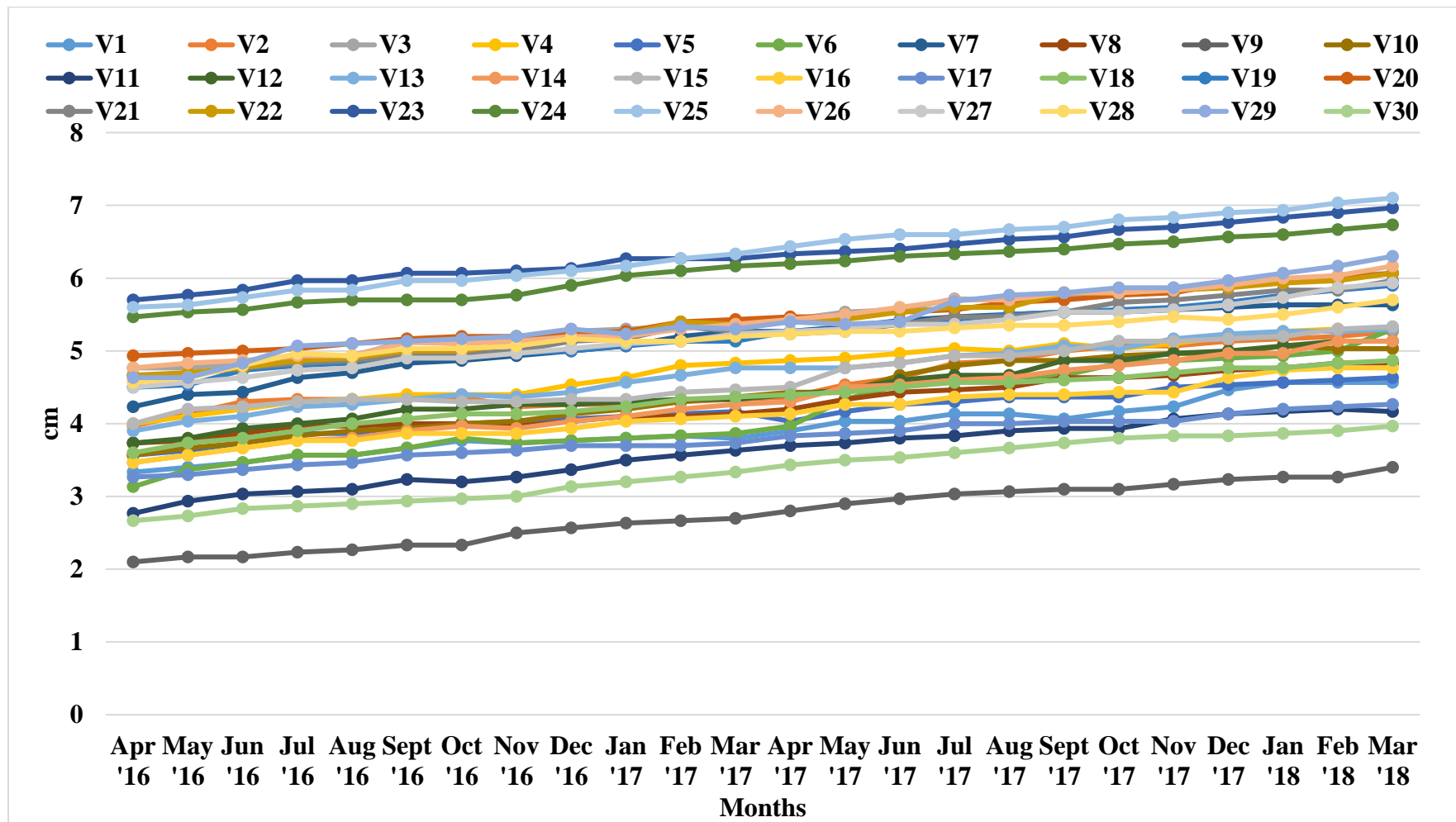


Fig. 3. Shoot girth of *Ascozentrum* hybrids/varieties during the period 2016-18

girth was observed in V<sub>25</sub> (6.43 cm), which was on par with V<sub>23</sub> (6.33 cm), V<sub>24</sub> (6.2 cm), V<sub>20</sub> (5.47 cm), V<sub>26</sub> (5.43 cm), V<sub>3</sub> (5.43 cm), V<sub>22</sub> (5.43 cm), V<sub>29</sub> (5.4 cm) and V<sub>19</sub> (5.27 cm).

However, significant variation was noted during August 2017 and December 2017. The maximum value of shoot girth was observed in V<sub>24</sub> (6.37 cm, in August 2017 and 6.57 cm, December 2017 respectively), which were on par with V<sub>25</sub> (6.53 cm, 6.77cm) and V<sub>23</sub> (6.67 cm, 6.9 cm). During March 2018 least significant variation was exhibited and maximum shoot girth (Table 8) was measured in the hybrid/variety V<sub>25</sub> (7.10 cm). This was on par with V<sub>23</sub> (6.97 cm), V<sub>24</sub> (6.73 cm), V<sub>29</sub> (6.30 cm), V<sub>26</sub> (6.17 cm), V<sub>3</sub> (6.07 cm), V<sub>20</sub> (6.07cm), V<sub>22</sub> (6.07cm), V<sub>21</sub> (5.97 cm), V<sub>27</sub> (5.93 cm), V<sub>19</sub> (5.9 cm), V<sub>28</sub> (5.7 cm), V<sub>7</sub> (5.63 cm), V<sub>15</sub> (5.33 cm), V<sub>4</sub> (5.33 cm), V<sub>6</sub> (5.33 cm) and V<sub>2</sub> (5.27 cm).

The minimum shoot girth was found during April 2016 in V<sub>9</sub> (2.10 cm). The minimum shoot girth was observed in V<sub>9</sub> throughout the months *viz.*, Aug. 2016 (2.27 cm), Dec. 2016 (2.57 cm), April 2017 (2.8 cm), August 2017 (3.07 cm), December 2017 (3.23 cm) and March 2018 (3.23 cm) which was on par with V<sub>17</sub> throughout the entire study period *viz.*, August 2016 (2.90 cm), December 2016 (3.13 cm), April 2017 (3.43 cm), August 2017 (3.67 cm), December 2017 (3.83 cm) and March 2018 (3.97 cm) is given in Table 8.

#### **4.1.1.1.4 Shoot diameter**

The data regarding the diameter of shoots *Ascocentrum* hybrids are presented in Appendix VII, VIII and Fig. 4. The diameter of shoot was recorded at monthly intervals for two years (April 2016 to March 2018). Least significant was variation observed among the thirty varieties during the period of study. In the month of April 2016, V<sub>23</sub> (*Mok. Omayaiy Yellow*) recorded maximum diameter (1.82 cm), which was on par with V<sub>25</sub> (*Mok. Sayan × Ascda. DOUNG PORN*) (1.78 cm). V<sub>25</sub> (1.74 cm), V<sub>24</sub> (1.74 cm) and V<sub>25</sub> (1.57 cm). During August 2016, V<sub>23</sub> (*Mok. Omayaiy Yellow*) recorded maximum diameter (1.90 cm), which was on par with V<sub>25</sub> (*Mok. Sayan × Ascda. DOUNG PORN*) (1.86 cm) and V<sub>24</sub> (1.82 cm).

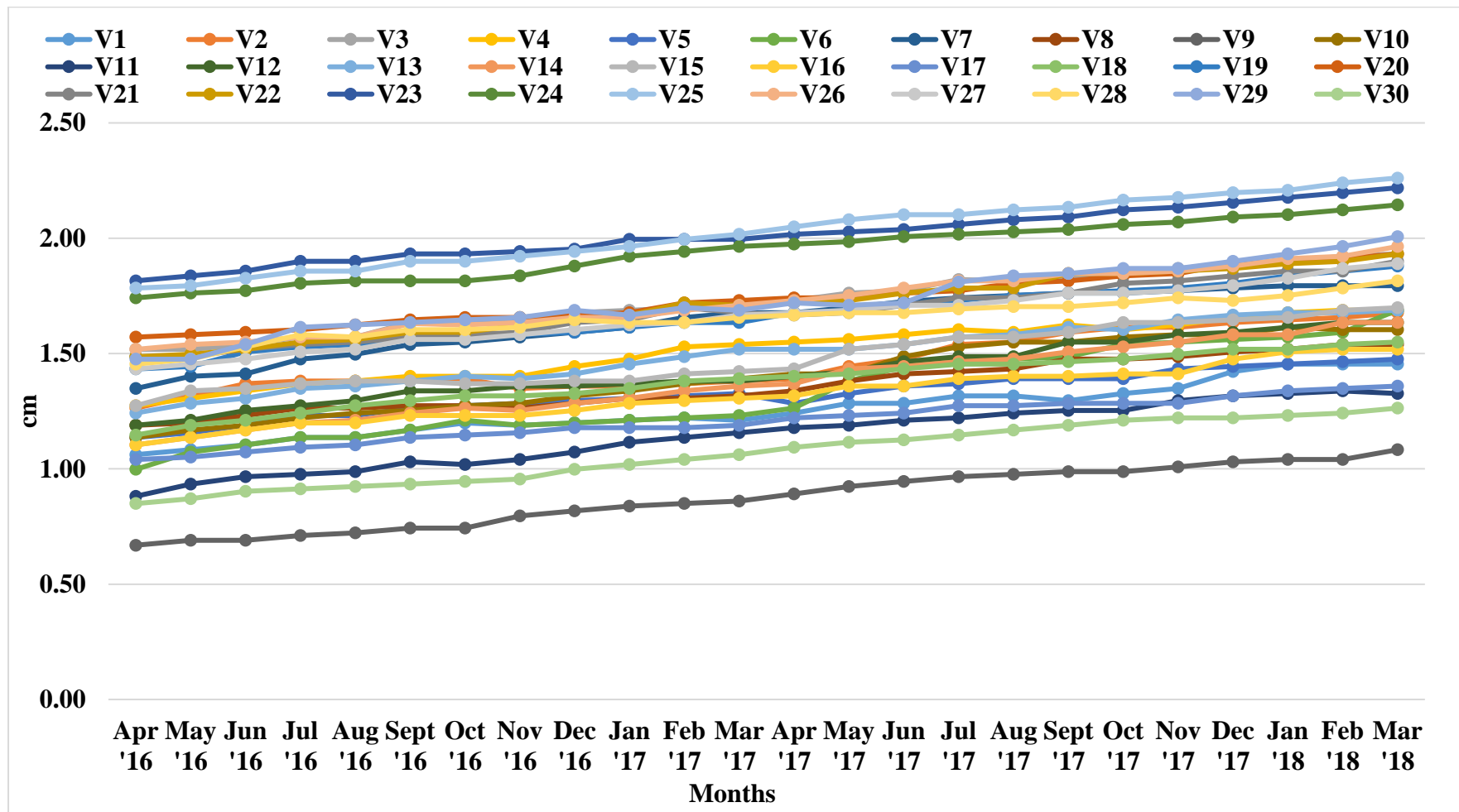


Fig. 4. Shoot diameter *Ascocentrum* hybrids/varieties during the period 2016-18

Maximum diameter was found in V<sub>23</sub> during Dec. 2016 (1.95 cm), which was on par with V<sub>25</sub> (*Mok. Sayan* × *Ascda. Doung Porn*) (1.94 cm) and V<sub>24</sub> (1.89 cm). The maximum diameter was recorded in V<sub>25</sub>. During Apr. 2017 (2.05 cm), Aug. 2017 (2.12 cm), Dec. 2017 (2.20 cm) and Mar. 2018 (2.26 cm) (Table 8).

The minimum shoot diameter was observed in V<sub>9</sub> throughout the study period April 2016 (0.67 cm), August 2016 (0.72 cm), December 2016 (0.82 cm), April 2017 (0.89 cm), August 2017 (0.98 cm), December 2017 (1.03 cm) and March 2018 (1.08 cm) and was on par with V<sub>17</sub> throughout the entire months [April 2016 (0.85 cm), August 2016 (0.92 cm), December 2017 (1.00 cm), April 2017 (1.09 cm), August 2017 (1.17 cm), December 2017 (1.22 cm) and March 2018 (1.26 cm)].

#### **4.1.1.1.5 Internodal length**

The data regarding the internodal length of *Ascocentrum* hybrids/varieties are presented in Appendix IX, X and Fig. 5. *Ascocentrum* hybrids/varieties showed variations with respect to internodal length. The maximum internodal length was observed in V<sub>23</sub> *Mok. Omayaiy Yellow* (3.73 cm) during Apr. 2016, which was on par with V<sub>24</sub> (3.47 cm), and V<sub>25</sub> (3.47 cm). The least internodal length was recorded in V<sub>9</sub> (0.27 cm), which was on par with V<sub>11</sub> (0.60 cm), V<sub>10</sub> (0.63 cm), V<sub>8</sub> (0.69 cm), V<sub>6</sub> (0.70 cm) and V<sub>7</sub> (0.70 cm). During Aug. 2016, the highest value was recorded in V<sub>23</sub> (4.10 cm), which was on par with V<sub>25</sub> (3.8 cm), V<sub>24</sub> (3.77 cm) V<sub>28</sub> (3.63 cm) and V<sub>21</sub> (3.63 cm). Whereas, the lowest internodal length was observed in V<sub>9</sub> (0.73 cm).

In the month of December 2016, the maximum value was observed in V<sub>23</sub> (4.37 cm), while, the lowest value was recorded in V<sub>9</sub> (1.01 cm), which was on par with V<sub>17</sub> (1.13 cm), V<sub>6</sub> (1.17 cm), V<sub>10</sub> (1.20 cm), V<sub>7</sub> (1.33 cm), V<sub>8</sub> (1.33 cm), V<sub>11</sub> (1.33 cm) V<sub>12</sub> (1.50 cm), V<sub>5</sub> (1.57 cm), V<sub>14</sub> (1.60 cm) and V<sub>13</sub> (1.67 cm). Similarly, the hybrid/variety V<sub>23</sub> showed highest internodal length during the rest of the period *viz.*, April 2017 (4.63 cm), August 2017 (4.93 cm), December 2017 (4.37 cm) and March 2018 (4.37 cm), while the lowest internodal length was



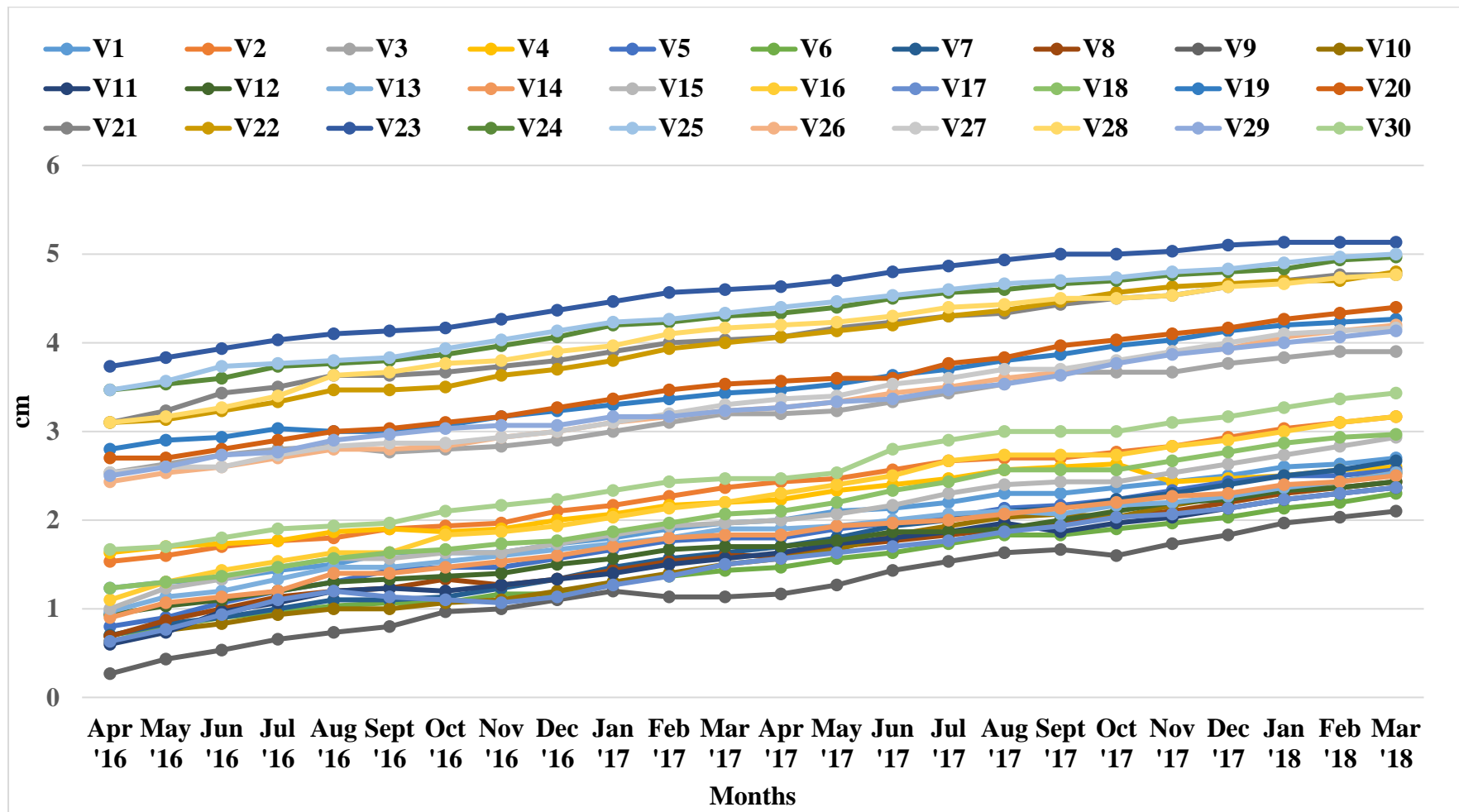


Fig. 5. Internodal length of *Ascozentrum* hybrids/varieties during the period 2016-18

recorded in V<sub>9</sub> for the month of April 2017 (1.17 cm), August 2017 (1.63 cm), December 2017 (1.83 cm) and March 2018 (2.10 cm).

#### **4.1.1.2 Leaf characters**

The data pertaining to the leaf characters of different *Ascocentrum* hybrids/varieties are presented in Appendix XI-XVIII and Fig. 6-12.

##### **4.1.1.2.1 Number of leaves**

Marked variation was noticed in the number of leaves produced by the hybrids (Appendix XI, XII and Fig. 6). During the month of April 2016, the maximum number of leaves per plants were counted in V<sub>11</sub> (54.00) which was on par with V<sub>23</sub> (53.80). The minimum number of leaves was observed in V<sub>16</sub> (5.40) which was on par with V<sub>9</sub> (5.80), V<sub>11</sub> (8.40), V<sub>2</sub> (9.40), V<sub>7</sub> (9.40), V<sub>17</sub> (9.60), V<sub>14</sub> (11.40), V<sub>1</sub> (11.40), V<sub>10</sub> (11.60), V<sub>18</sub> (11.80) and V<sub>15</sub> (12.20). During August 2016 highest number of leaves was found in V<sub>23</sub> (57.20) which was on par with V<sub>25</sub> (57.20). The least number of leaves was recorded in V<sub>16</sub> (7.60). During December 2016, the maximum number of leaves was observed in V<sub>25</sub> (61.00) which was on par with V<sub>23</sub> (60.60) and V<sub>24</sub> (53.60). The minimum number of leaves were counted in V<sub>16</sub> (10.20) which was on par with V<sub>9</sub> (10.80), V<sub>11</sub> (13.40), V<sub>2</sub> (14.40), V<sub>17</sub> (14.80), V<sub>7</sub> (16.00), V<sub>1</sub> (16.00), V<sub>10</sub> (16.60), V<sub>14</sub> (16.80), V<sub>18</sub> (17.20), V<sub>15</sub> (18.00) and V<sub>13</sub> (18.40). Differences were found among the varieties during April 2017. Whereas, the maximum number of leaves was found in V<sub>25</sub> (64.40), which was on par with V<sub>23</sub> (64.20) and V<sub>24</sub> (57.00).

The least number of leaves was observed in V<sub>16</sub> (11.80). During August 2017, the highest number of leaves was recorded in V<sub>25</sub> (68.20), which was on par with V<sub>23</sub> (67.80) and V<sub>24</sub> (60.00). The minimum number of leaves was observed in V<sub>16</sub> (14.00). Marked variation was observed during December 2017 and V<sub>25</sub> (71.80) was observed to have the maximum number of leaves and which was on par with V<sub>23</sub> (70.60). The minimum number of leaves were found in V<sub>16</sub> (16.60). In March 2018 (Table 9), the maximum number of leaves was found in V<sub>25</sub> (74.80) which was on par with V<sub>23</sub> (72.80).

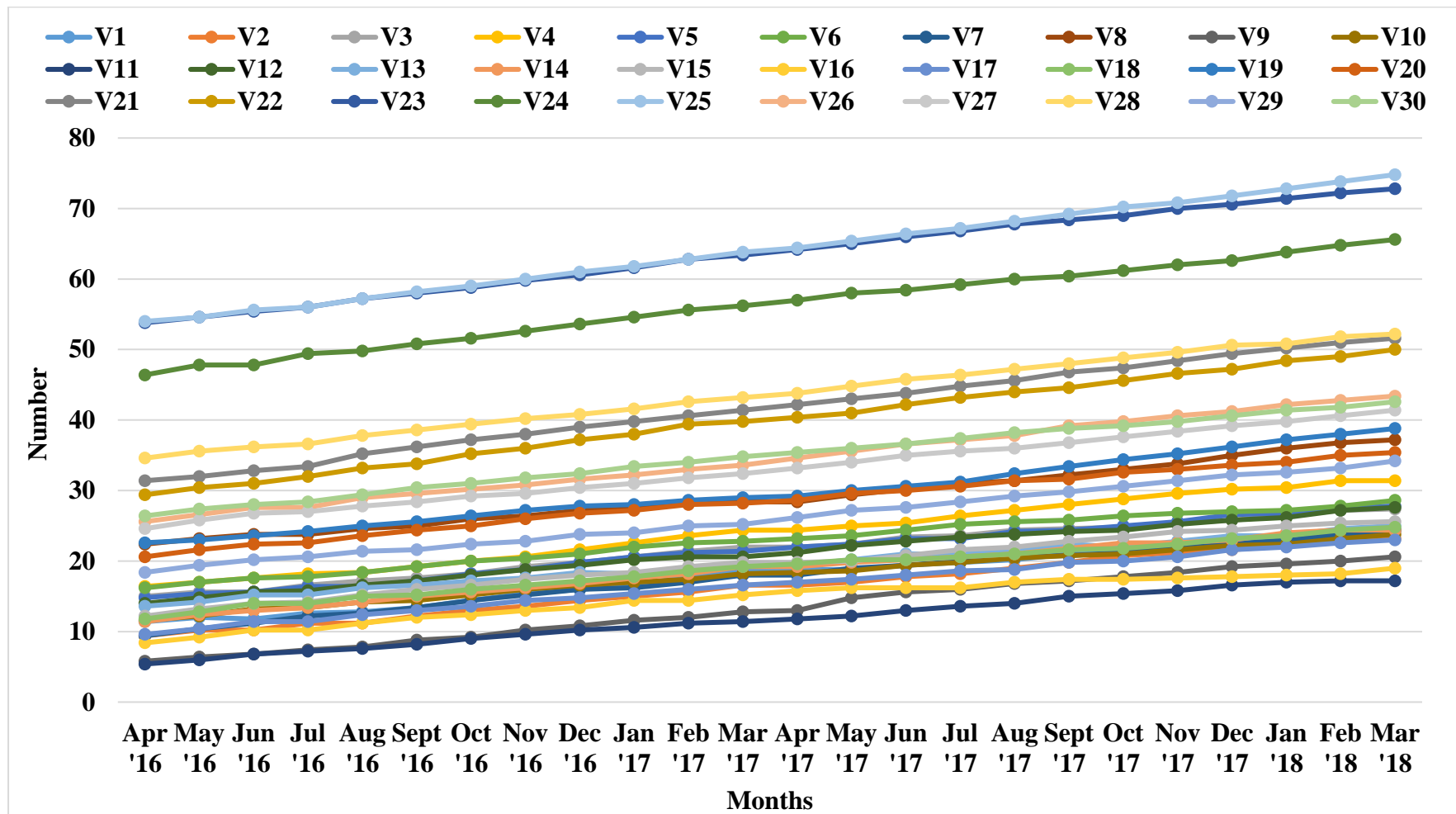


Fig. 6. Number of leaves of *Ascozentrum* hybrids/varieties during the period 2016-18

**Table 9. Quantitative morphological leaf characters of intergeneric hybrids of *Ascocentrum* during March 2017 and 2018**

Var. No.	Number of leaves	Leaf length (cm)		Leaf breadth (cm)		Leaf area (cm <sup>2</sup> )	
	Mar.'18	Mar.'17	Mar.'18	Mar.'17	Mar.'18	Mar.'17	Mar.'18
V <sub>1</sub>	24.40	27.66	27.00	3.00	3.10	54.01	53.19
V <sub>2</sub>	23.20	26.50	25.68	3.10	3.14	52.54	51.49
V <sub>3</sub>	27.40	37.40	34.44	3.76	3.90	66.88	63.09
V <sub>4</sub>	31.40	25.82	26.96	2.94	3.16	51.61	53.16
V <sub>5</sub>	27.80	24.94	27.92	3.10	3.25	50.52	54.22
V <sub>6</sub>	28.60	25.22	24.84	2.38	2.32	50.65	50.14
V <sub>7</sub>	24.40	23.74	24.82	3.08	2.90	48.96	50.30
V <sub>8</sub>	37.20	27.02	25.98	2.74	2.65	53.10	51.55
V <sub>9</sub>	20.60	13.02	15.02	0.82	1.04	34.34	37.00
V <sub>10</sub>	23.80	24.68	25.68	2.62	2.74	50.03	51.36
V <sub>11</sub>	19.00	14.62	15.56	2.16	2.20	36.85	38.08
V <sub>12</sub>	27.60	18.56	20.28	2.66	2.80	42.11	44.39
V <sub>13</sub>	25.00	26.00	27.22	2.76	2.72	51.79	53.35
V <sub>14</sub>	24.40	27.36	25.70	2.74	2.64	53.54	51.36
V <sub>15</sub>	25.60	28.14	25.90	2.56	2.52	54.50	51.58
V <sub>16</sub>	17.20	22.66	21.98	2.96	2.76	47.52	46.58
V <sub>17</sub>	23.00	19.12	16.70	2.40	2.54	42.75	39.66
V <sub>18</sub>	24.80	26.82	23.56	3.00	2.88	52.93	48.66
V <sub>19</sub>	38.80	20.06	21.02	3.42	3.12	44.30	45.45
V <sub>20</sub>	35.40	20.74	23.06	3.30	3.22	45.14	48.12
V <sub>21</sub>	51.60	27.86	27.28	2.94	2.92	54.25	53.50
V <sub>22</sub>	50.00	31.34	30.90	3.26	3.36	58.87	58.33
V <sub>23</sub>	72.80	31.72	30.84	4.04	3.94	59.61	58.44
V <sub>24</sub>	65.60	22.96	23.64	3.92	3.88	48.22	49.09
V <sub>25</sub>	74.80	21.60	21.42	3.66	3.48	46.37	46.08
V <sub>26</sub>	43.40	21.48	20.18	3.60	3.62	46.20	44.52
V <sub>27</sub>	41.40	37.80	38.74	3.98	3.80	67.47	68.63
V <sub>28</sub>	52.20	35.14	32.90	3.20	3.38	63.77	60.93
V <sub>29</sub>	34.20	29.40	27.96	3.24	3.14	56.35	54.45
V <sub>30</sub>	42.60	20.90	20.80	3.24	3.36	45.33	45.24
<b>C.D.</b>	<b>0.658</b>	<b>0.407</b>	<b>0.489</b>	<b>0.072</b>	<b>0.168</b>	<b>0.37</b>	<b>0.443</b>
<b>SE(M)</b>	<b>0.235</b>	<b>0.145</b>	<b>0.174</b>	<b>0.026</b>	<b>0.06</b>	<b>0.132</b>	<b>0.158</b>
<b>C.V.</b>	<b>8.906</b>	<b>6.372</b>	<b>7.688</b>	<b>2.867</b>	<b>6.758</b>	<b>4.105</b>	<b>4.931</b>

#### **4.1.1.2.2 Leaf length**

The *Ascocentrum* hybrids/varieties showed considerable variations in leaf length during the period of observation (Appendix XIII, XIV, Fig. 7 and 8). In the month of April 2016 longest leaf was found in V<sub>27</sub> (35.44 cm) which was on par with V<sub>3</sub> (35.20 cm) and V<sub>28</sub> (32.80 cm). The minimum leaf length was found in V<sub>9</sub> (8.60 cm) which was on par with V<sub>11</sub> (10.90 cm). During August 2016 maximum leaf length was recorded in V<sub>27</sub> (37.06 cm) which was on par with V<sub>3</sub> (37.06 cm) and V<sub>28</sub> (34.62 cm), whereas, the minimum leaf length was observed in V<sub>11</sub> (13.36 cm) which was on par with V<sub>9</sub> (10.86 cm). Almost similar results were found in Dec. 2017 also wherein longest leaf was observed in V<sub>27</sub> (*Kag. Youthong Beauty*) which was on par V<sub>3</sub> (*Ascda. Kultana* × *V. Bitz's Heartthrob*) and V<sub>28</sub> (*Kag. Christie Low*). The minimum leaf length was found in V<sub>9</sub> (*Vasco. Aroonsri Beauty*) which was on par with V<sub>11</sub> (*Vasco. Blue Bay White*).

In Apr. 2017 maximum leaf length was found in V<sub>27</sub> (36.60 cm) which was on par with V<sub>3</sub> (32.38 cm) while the minimum leaf length of was measured in V<sub>9</sub> (10.72 cm) which was on par with V<sub>11</sub> (12.30 cm), V<sub>17</sub> (14.30 cm), V<sub>12</sub> (14.30 cm), V<sub>27</sub> (38.66 cm) and V<sub>3</sub> (34.18 cm). In August 2017, longest leaf was recorded in V<sub>27</sub> (38.66 cm) and which was on par with V<sub>3</sub> (34.18 cm). Minimum leaf length was recorded in V<sub>9</sub> (12.94 cm) which was on par with V<sub>11</sub> (14.32 cm) and V<sub>17</sub> (16.06 cm). In Dec. 2017 maximum leaf length was recorded in V<sub>27</sub> (38.74 cm) which was on par with V<sub>3</sub> (34.44 cm) and the lowest leaf length was recorded in V<sub>9</sub> (14.62 cm) which was on par with V<sub>11</sub> (15.16 cm) and V<sub>17</sub> (16.64 cm). At the end of the study period in March 2018 longest leaf was found in V<sub>27</sub> (38.74 cm) which was on par with V<sub>3</sub> (34.44 cm) and minimum leaf length was recorded in V<sub>9</sub> (15.02 cm) which was on par with V<sub>11</sub> (15.56 cm), V<sub>17</sub> (16.70 cm), V<sub>26</sub> (20.18 cm) and V<sub>12</sub> (20.28 cm) (Table 9).

#### **4.1.1.2.3 Leaf breadth**

Appreciable differences were noticed among the varieties with respect to leaf breadth (Appendix XV, XVI, Fig. 9 and 10). During April 2016 maximum

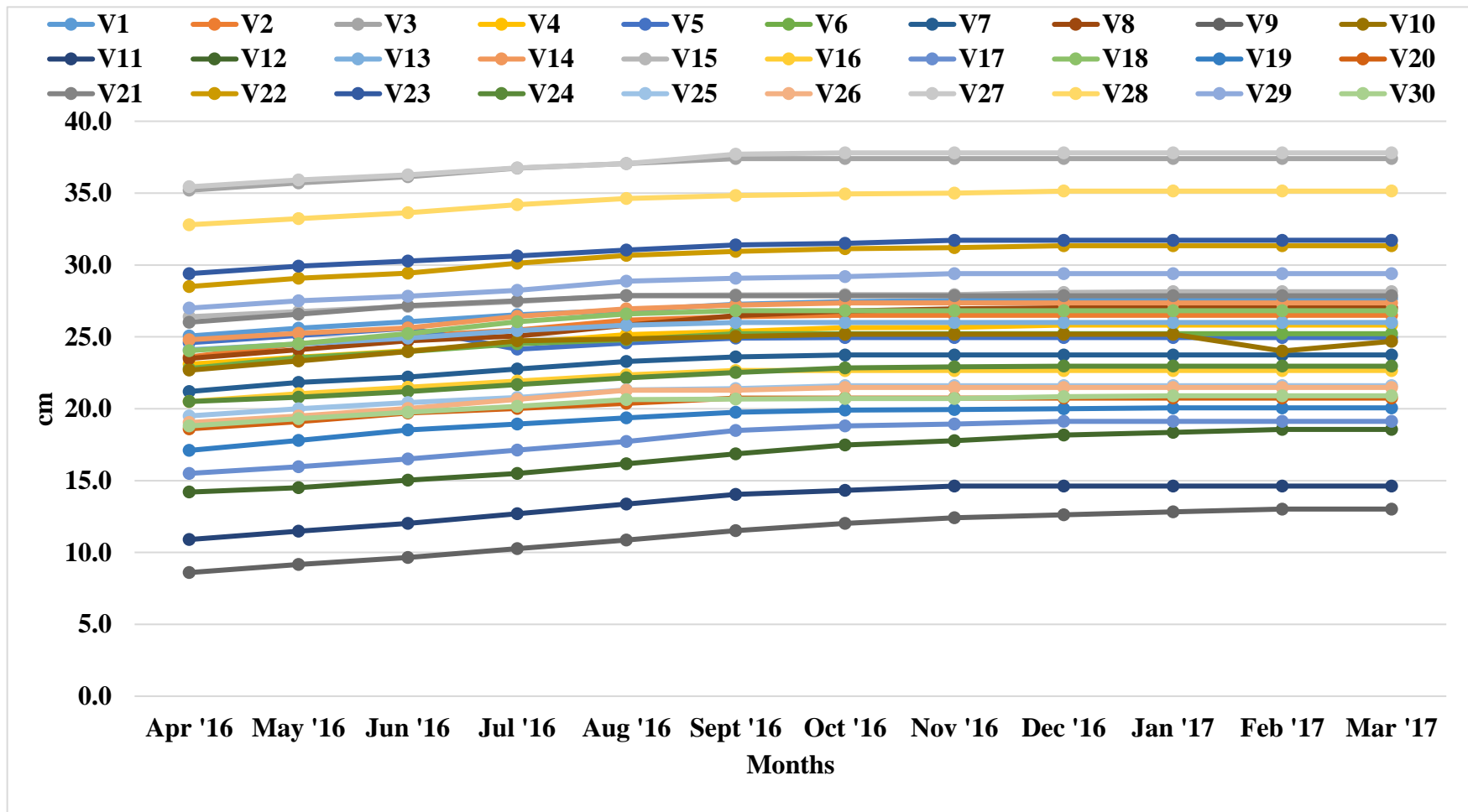


Fig. 7. Leaf length of *Ascocentrum* hybrids/varieties during the period 2016-17

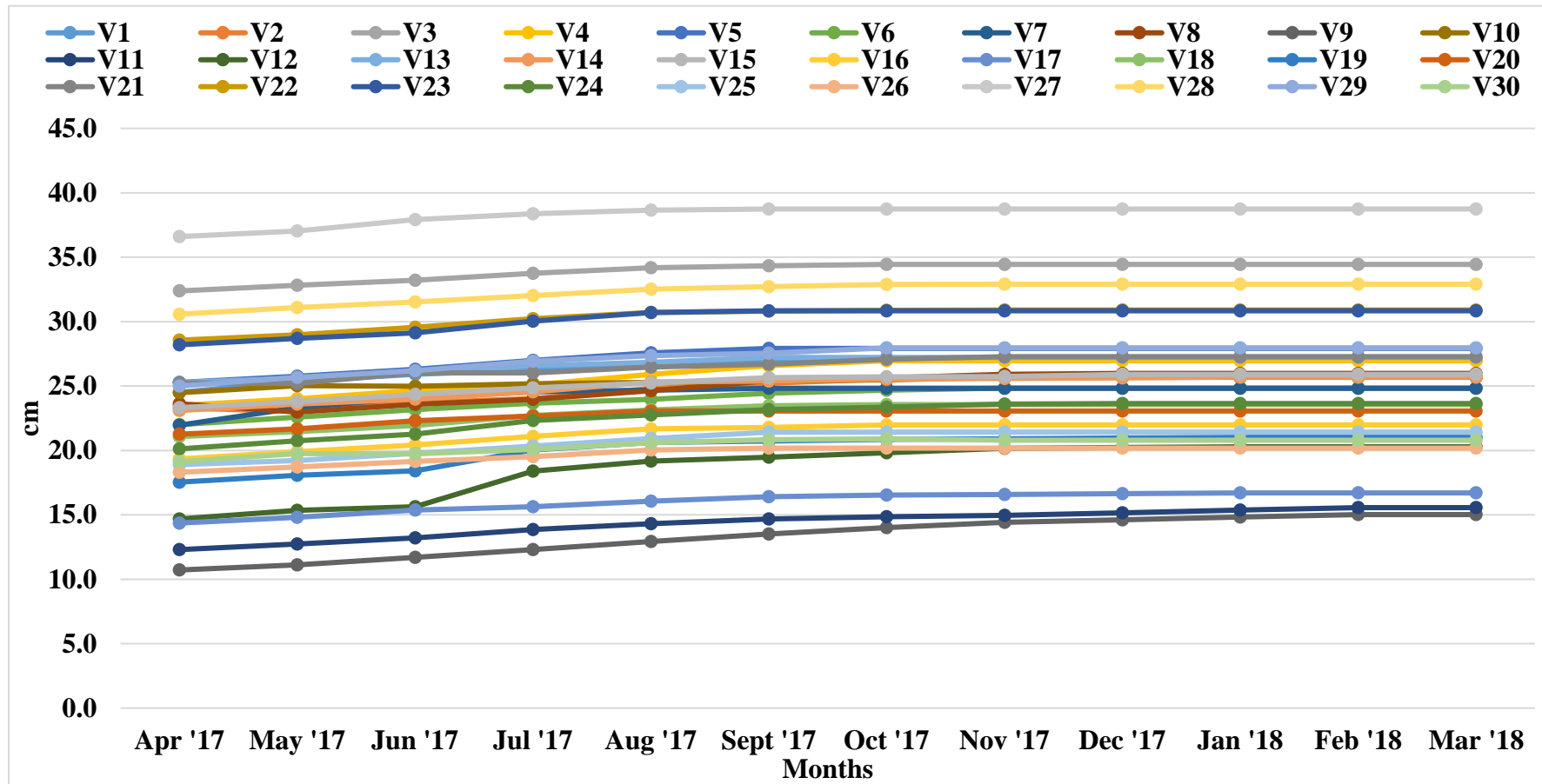


Fig. 8. Leaf length of *Ascozentrum* hybrids/varieties during the period 2017-18

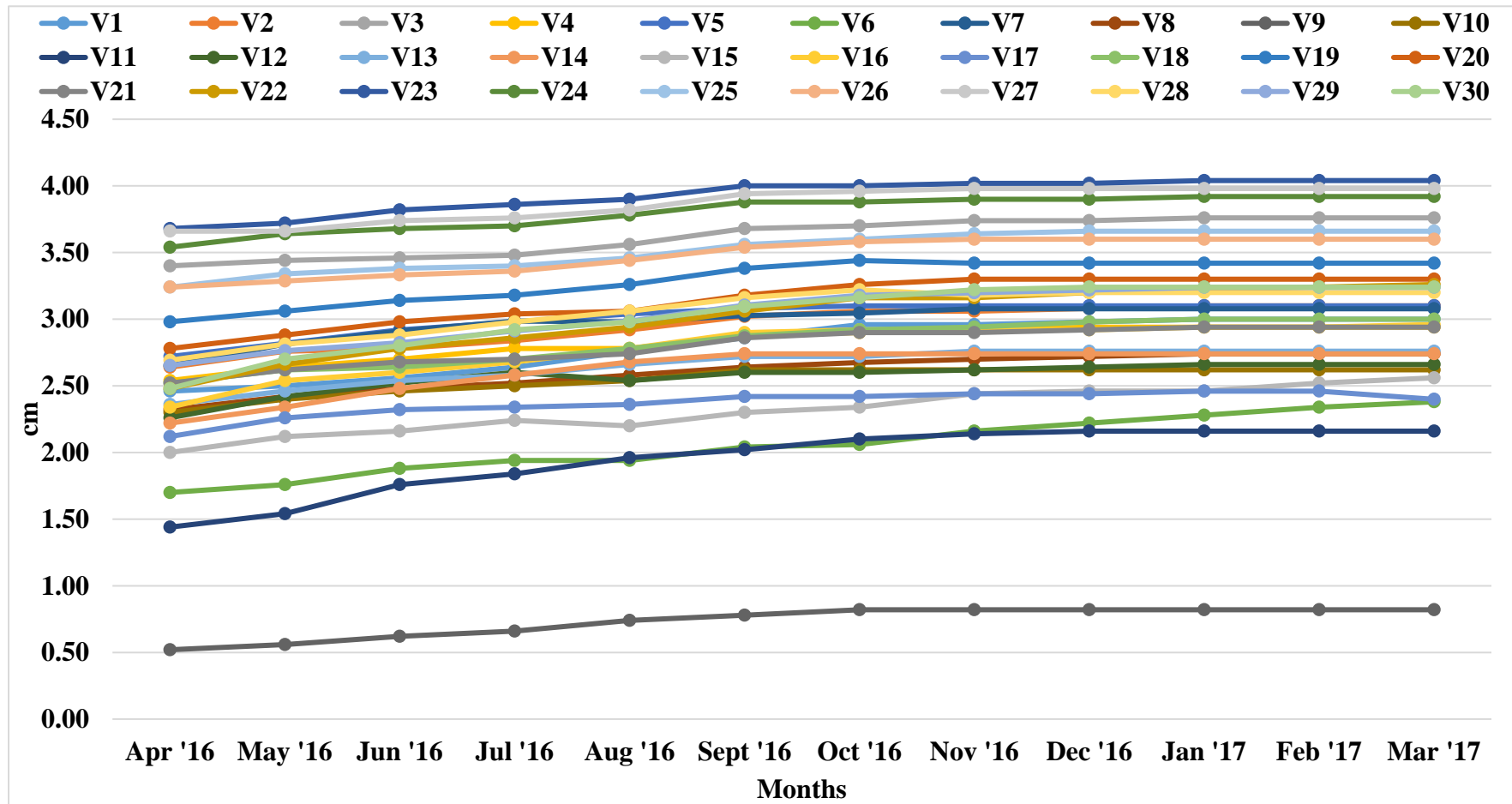


Fig. 9. Leaf breadth of *Ascozentrum* hybrids/varieties during the period 2016-17



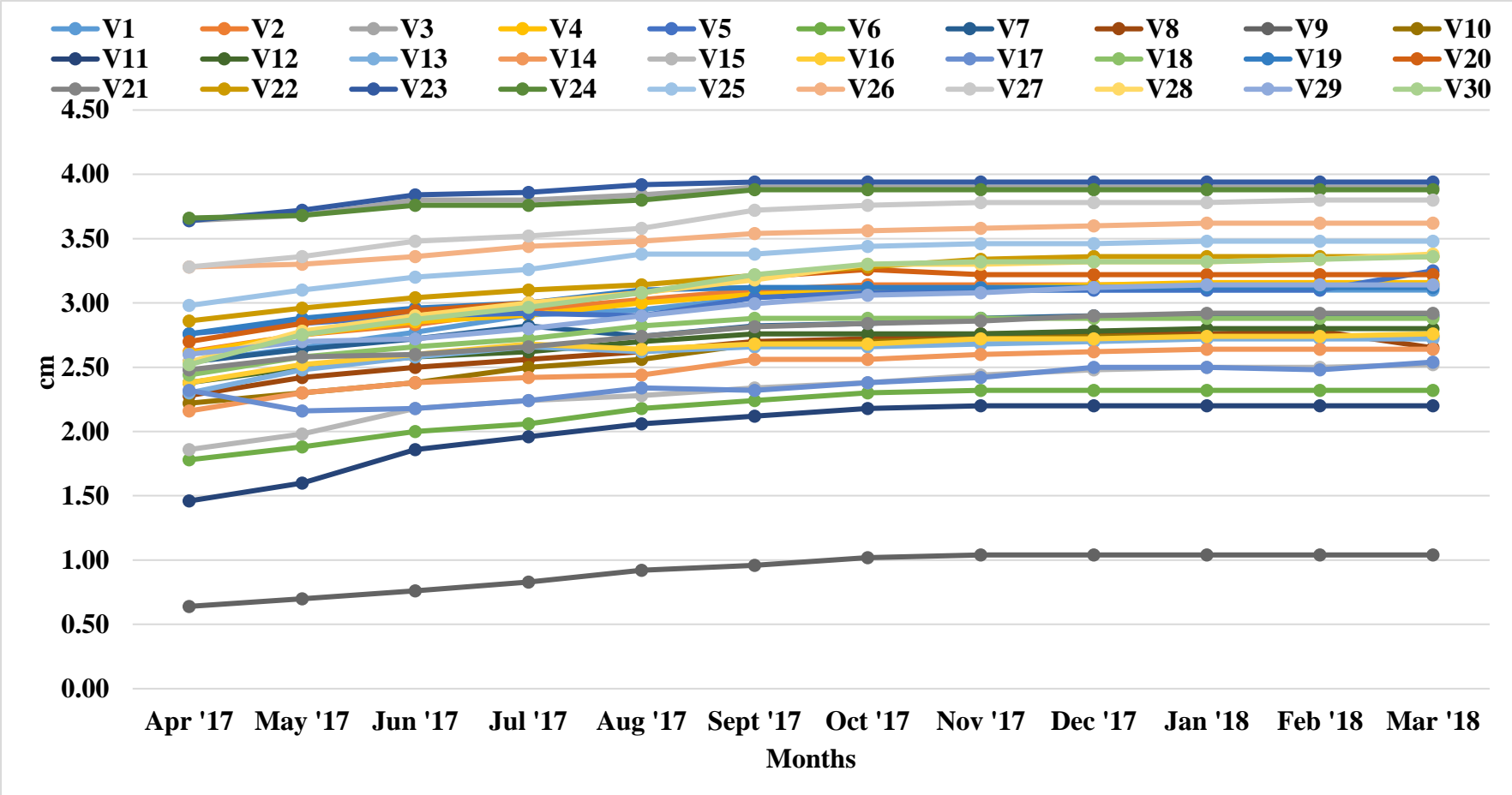


Fig. 10. Leaf breadth of *Ascocentrum* hybrids/varieties during the period 2017-18

leaf breadth was recorded in V<sub>23</sub> (3.68 cm) which was on par with V<sub>27</sub> (3.66 cm), V<sub>24</sub> (3.54 cm) and V<sub>3</sub> (3.40 cm). Throughout the study period, V<sub>9</sub> (*Vasco*. Aroonsri Beauty) was observed to have the lowest leaf breadth among all thirty hybrids/varieties. Throughout the months of study viz., April 2016 (0.52 cm), August 2016 (0.74 cm), December 2016 (0.82 cm) and March, April 2017 (0.64 cm), August 2017 (0.92 cm), December 2017 (1.04 cm) and March 2018 (1.04 cm) it was found to be significantly different from all other varieties. In August 2016 V<sub>23</sub> (3.90 cm) showed maximum leaf breadth and which was on par with V<sub>27</sub> (3.82 cm), V<sub>24</sub> (3.78 cm) and V<sub>3</sub> (3.56 cm). During December 2016 maximum leaf breadth was recorded in V<sub>23</sub> (4.02 cm) and was on par with V<sub>27</sub> (3.98 cm), V<sub>24</sub> (3.90 cm) and V<sub>3</sub> (3.74 cm).

In April 2017 maximum leaf breadth was recorded in V<sub>24</sub> (3.66cm) and was on par with V<sub>3</sub> (3.64cm), V<sub>23</sub> (3.64cm), V<sub>26</sub> (3.28cm) and V<sub>27</sub> (3.28cm). During August 2017, the highest value of leaf breadth was found in V<sub>23</sub> (3.92 cm). In December 2017 maximum breadth of the leaf was recorded in V<sub>23</sub> (3.94 cm) which was on par with V<sub>3</sub> (3.90 cm), V<sub>24</sub> (3.88 cm), V<sub>27</sub> (3.78 cm) and V<sub>26</sub> (3.60 cm). By the end of the study, during March 2018 (Table 9), maximum leaf breadth was recorded in V<sub>23</sub> (3.94 cm) which was on par with V<sub>3</sub> (3.90 cm), V<sub>24</sub> (3.88 cm), V<sub>27</sub> (3.78 cm) and V<sub>26</sub> (3.62 cm).

#### **4.1.1.2.4 Leaf area**

Marked significant differences were observed in the leaf area of different intergeneric hybrids of *Ascocentrum* (Appendix XVII, XVIII, Fig. 11 and 12). Maximum leaf area was recorded throughout the study period was V<sub>27</sub>. (*Kag*. Youthoung Beauty). During April 2016 maximum leaf area was recorded in V<sub>27</sub> (64.31 cm<sup>2</sup>) which was on par with V<sub>3</sub> (63.91 cm<sup>2</sup>) and V<sub>28</sub> (60.58 cm<sup>2</sup>) whereas, minimum leaf area was recorded in V<sub>9</sub> (28.52 cm<sup>2</sup>) which was on par with V<sub>11</sub> (31.79 cm<sup>2</sup>). In the month of August 2016, maximum leaf area was found in V<sub>27</sub> (66.46 cm<sup>2</sup>) and which was on par with V<sub>3</sub> (66.38 cm<sup>2</sup>) and V<sub>28</sub> (63.05 cm<sup>2</sup>). Almost similar kind of result was found in December 2016 whereas, in V<sub>27</sub> (67.47 cm<sup>2</sup>) largest leaf area recorded and was on par with V<sub>3</sub> (66.88 cm<sup>2</sup>) and V<sub>28</sub> (63.77

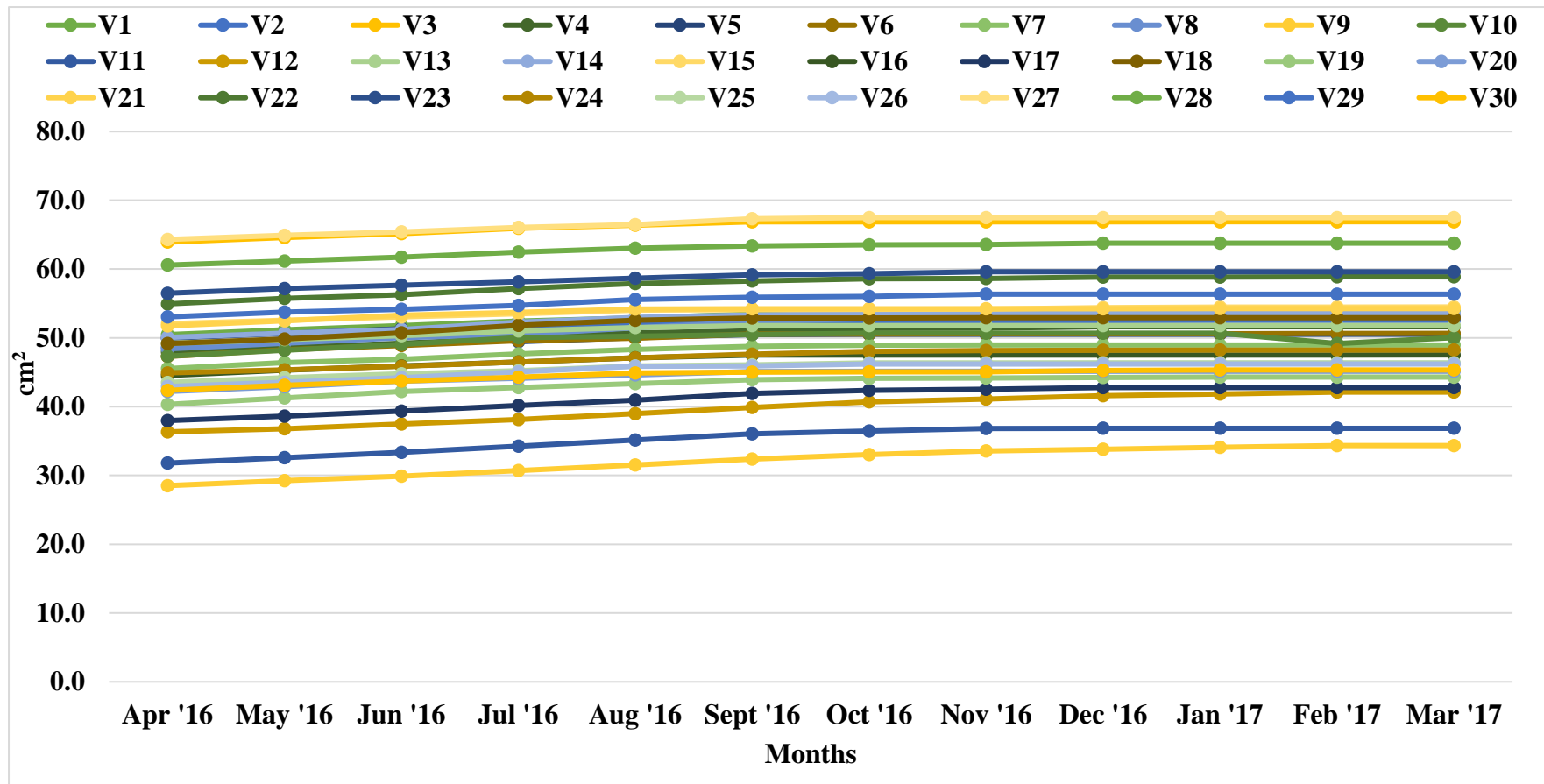


Fig. 11. Leaf area of *Ascozentrum* hybrids/varieties during the period 2016-17

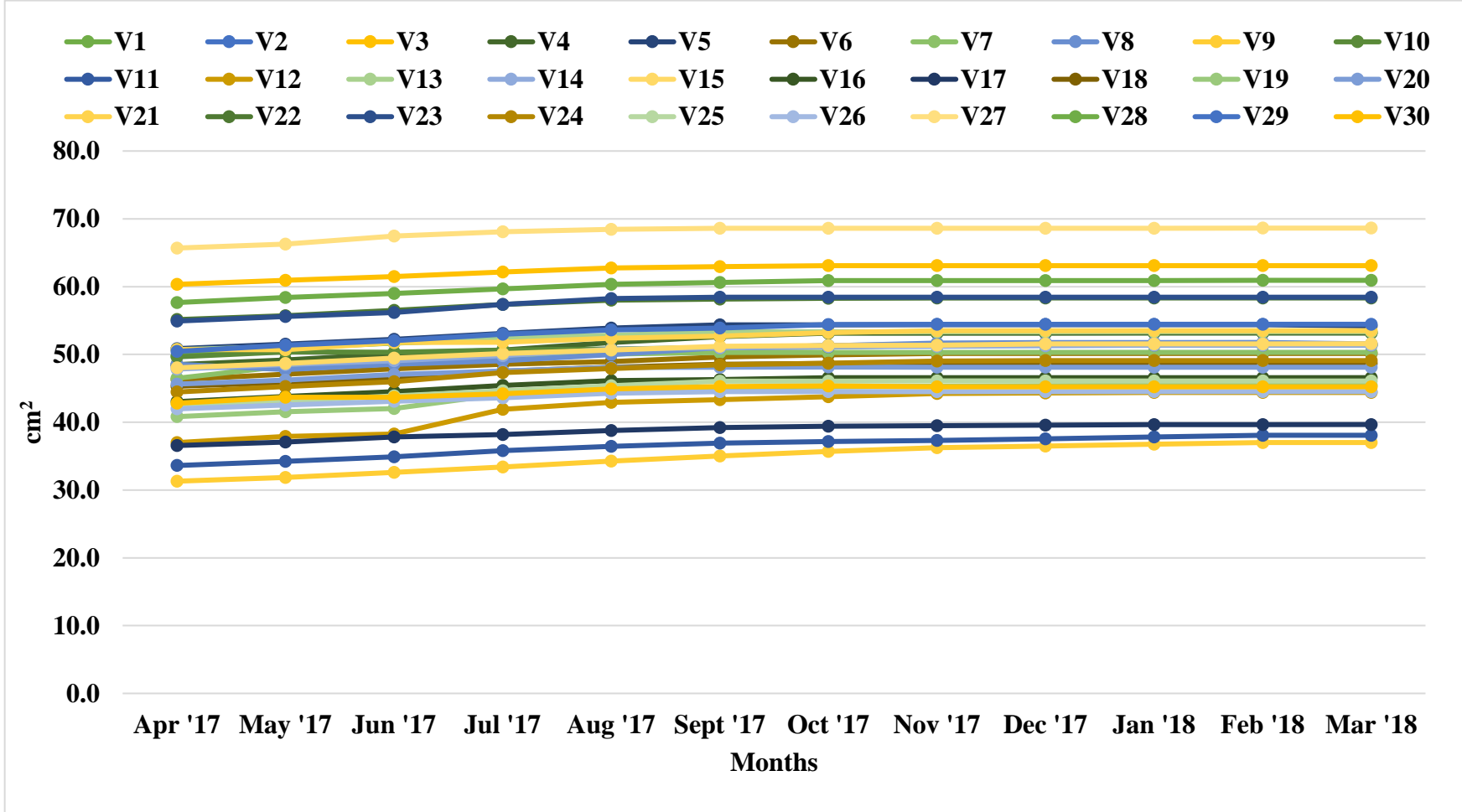


Fig. 12. Leaf area of *Ascocentrum* hybrids/varieties during the period 2017-18

cm<sup>2</sup>). Where the minimum leaf area was recorded in V<sub>9</sub> (33.82 cm<sup>2</sup>) and was on par with V<sub>11</sub> (36.85 cm<sup>2</sup>).

In April 2017 and August 2017, the largest leaf area was recorded in V<sub>27</sub> (65.69 cm<sup>2</sup> and 68.46 cm<sup>2</sup>, respectively), which was on par with V<sub>3</sub> (60.34 cm<sup>2</sup> and 62.73 cm<sup>2</sup> respectively). The smallest leaf area found during Apr. 2017, was in V<sub>9</sub> (31.30 cm<sup>2</sup>) and during Aug. 2017 also was in V<sub>9</sub> (34.27cm<sup>2</sup>). At the end of the year in Dec. 2017, maximum leaf area was found in V<sub>27</sub> (68.62 cm<sup>2</sup>) which was on par with V<sub>3</sub> (63.09 cm<sup>2</sup>). Whereas, minimum leaf area was found in V<sub>9</sub> (36.49 cm<sup>2</sup>) which was on par with V<sub>11</sub> (37.56 cm<sup>2</sup>) and V<sub>17</sub> (39.57 cm<sup>2</sup>). By the end of the study period in March 2018 (Table 9), maximum leaf area was observed in V<sub>27</sub> (68.63 cm<sup>2</sup>) followed by V<sub>3</sub> (63.09 cm<sup>2</sup>) whereas the minimum leaf area was found in V<sub>9</sub> (37.00 cm<sup>2</sup>) which was on par with V<sub>17</sub> (39.66 cm<sup>2</sup>) and V<sub>11</sub> (38.08 cm<sup>2</sup>).

#### **4.1.1.3 Root characters**

Data pertaining to aerial root characters of *Ascocentrum* hybrids/varieties are presented in Appendix XIX, XXIV, Fig. 13 and 14.

##### **4.1.1.3.1 Number of roots**

Noticeable differences were observed in the number of roots produced by *Ascocentrum* hybrids/varieties throughout the study period (Appendix XIX, XX and Fig. 13). During the month of April 2016, the maximum number of roots per plant was recorded in V<sub>4</sub> (18.80) which was on par with V<sub>23</sub> (16.00). Whereas, the minimum number of roots was recorded in V<sub>9</sub> (1.40) which was on par with V<sub>18</sub> (4.60). During August 2016, the maximum number of roots was found in V<sub>4</sub> (19.80) followed by V<sub>23</sub> (17.00) and the minimum number of roots was observed in V<sub>9</sub> (3.20) which was on par with V<sub>18</sub> (6.00). During December 2016, the maximum number of roots was recorded in V<sub>4</sub> (22.00) which was on par with V<sub>23</sub> (19.40). The minimum number of leaves were observed in V<sub>16</sub> (4.80) which was on par with V<sub>9</sub> (7.00) and V<sub>11</sub> (8.00).

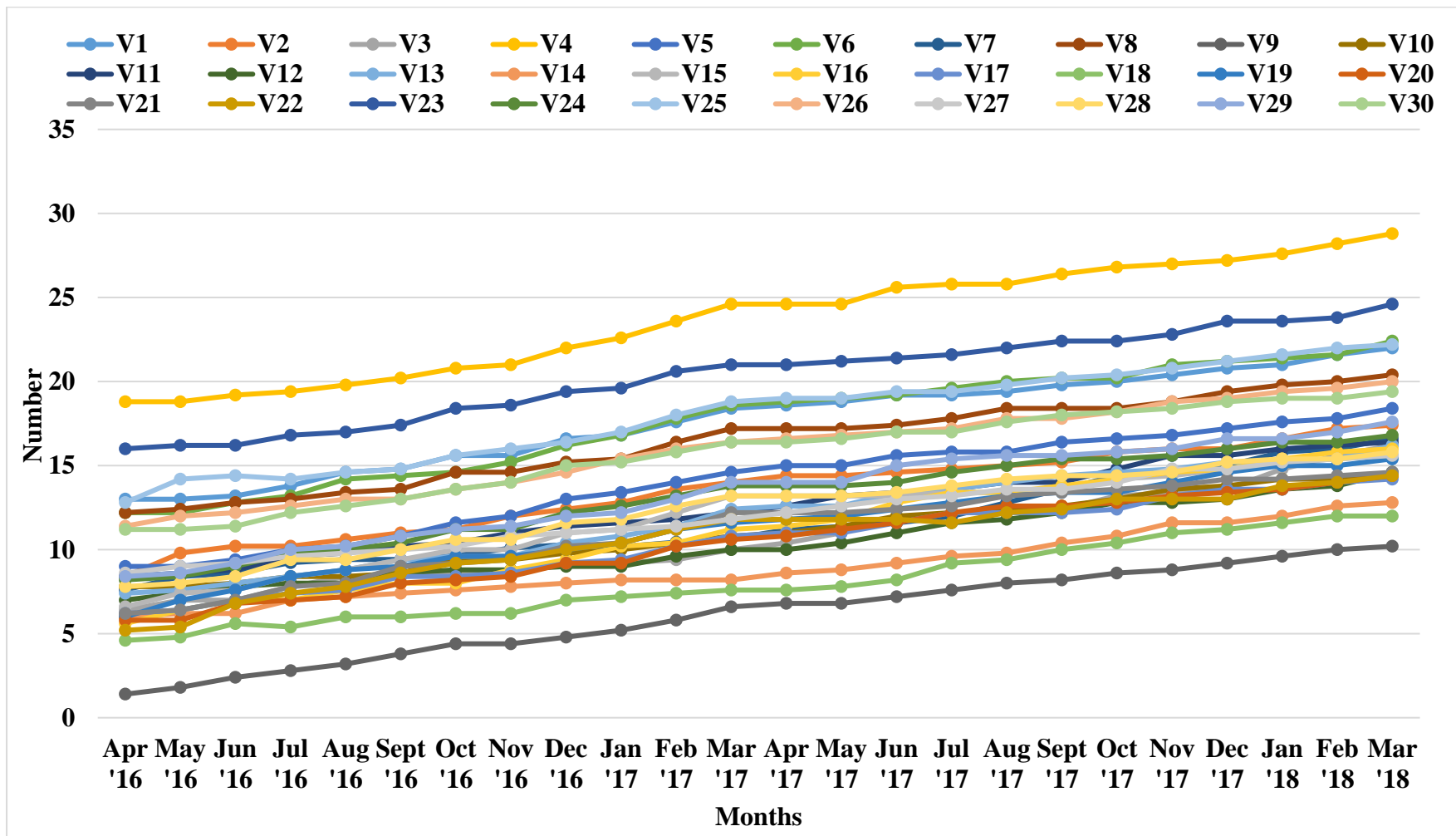


Fig. 13. Number of roots of *Ascozentrum* hybrids/varieties during the period 2016-18

In April 2017 maximum number of roots was found in V<sub>4</sub> (24.60) which was on par with V<sub>23</sub> (21.00). The least number of roots were counted in V<sub>9</sub> (6.80). However, there was no significant difference between V<sub>18</sub> (7.60), V<sub>14</sub> (8.60), V<sub>12</sub> (10.00), V<sub>3</sub> (10.40), V<sub>20</sub> (10.80), V<sub>17</sub> (11.00), V<sub>10</sub> (11.00), V<sub>7</sub> (11.18) and V<sub>16</sub> (11.40). During August 2017 highest number of roots was recorded in V<sub>25</sub> (25.80) which was on par with V<sub>23</sub> (22.00). While the least number of roots was observed in V<sub>9</sub> (8.00). Marked variation was observed during December 2017, the highest number of roots was observed in V<sub>4</sub> (27.20) which was on par with V<sub>23</sub> (23.60) while the minimum number of roots was found in V<sub>16</sub> (9.20). At the end of the study period in March 2018, the maximum number of roots was found in V<sub>4</sub> (28.80) which was significantly superior to all thirty hybrids/varieties (Table 8). The least number of roots was found in V<sub>9</sub> (10.20) which was statistically on par with V<sub>18</sub> (12.00), V<sub>14</sub> (12.80), V<sub>17</sub> (14.20), V<sub>22</sub> (14.40), V<sub>20</sub> (14.40), V<sub>10</sub> (14.40), V<sub>21</sub> (14.60) and V<sub>12</sub> (14.60).

#### **4.1.1.3.2 Length of roots**

The *Ascocentrum* hybrids/varieties showed considerable variations with respect to root length throughout the study period (Appendix XXI and XXII). In the month of April 2016, longest roots were found in V<sub>16</sub> (171.66 cm) which was on par with V<sub>18</sub> (169.33 cm), V<sub>15</sub> (165.00 cm), V<sub>13</sub> (155.33 cm), V<sub>14</sub> (154.00 cm), V<sub>7</sub> (154.00 cm), V<sub>4</sub> (151.00 cm) and V<sub>8</sub> (151.00 cm). The minimum length of the roots was found in V<sub>9</sub> (25.73 cm) which was on par with V<sub>27</sub> (39.23cm) V<sub>26</sub> (41.33 cm) V<sub>21</sub> (45.23 cm) and V<sub>28</sub> (47.66 cm). During the month August 2016, longest root length was observed in V<sub>27</sub> (184.0 cm) whereas minimum root length was observed in V<sub>9</sub> (30.23 cm.). In December 2016, maximum root length was observed in V<sub>27</sub> (191.33 cm). The minimum root length of was observed in V<sub>9</sub> (38.39 cm) there was no significant difference between V<sub>26</sub> (47.00 cm), V<sub>27</sub> (48.87 cm) and V<sub>16</sub> (14.30 cm).

In the month of April 2017 maximum root length was found in V<sub>16</sub> (195.0 cm). The minimum root length was observed in V<sub>9</sub> (38.38 cm) which was statistically on par with V<sub>26</sub> (50.30 cm), V<sub>27</sub> (51.48 cm) and V<sub>19</sub> (56.93 cm) and

V<sub>21</sub> (57.94 cm). In August 2017 longest roots were recorded in V<sub>16</sub> (199.0 cm) and shortest roots in V<sub>9</sub> (40.71 cm). During the month of December 2017 maximum root length was recorded in V<sub>16</sub> (202.87 cm). Whereas, the lowest root length was recorded in V<sub>9</sub> (43.39 cm) which was statistically on par with V<sub>26</sub> (54.10 cm) V<sub>27</sub> (58.27 cm). V<sub>27</sub> (58.27 cm), V<sub>21</sub> (65.10 cm) and V<sub>21</sub> (66.27 cm). By the end of the study period in March 2018 (Table 8), longest roots were found in V<sub>16</sub> (205.3 cm) and it was on par with V<sub>18</sub> (196.2 cm), V<sub>6</sub> (194.33 cm), V<sub>15</sub> (193.67 cm), V<sub>14</sub> (193.33 cm), V<sub>13</sub> (192.00 cm) V<sub>7</sub> (191.67 cm), V<sub>15</sub> (189.80 cm) and V<sub>6</sub> (184.67 cm). Whereas, minimum root length was recorded in V<sub>9</sub> (45.23 cm) which was on par with V<sub>26</sub> (57.82 cm), V<sub>27</sub> (61.09 cm), V<sub>19</sub> (68.17cm), V<sub>21</sub> (68.40 cm).

#### 4.1.1.3.3 *Girth of roots*

*Ascocentrum* hybrids did not exhibit variation among the varieties with respect to shoot girth of aerial roots (Appendix XXIII, XXIV and Fig. 14). During Apr. 2016 maximum root girth (5.70 cm) was observed in V<sub>23</sub> *Mok. Omayyai* Yellow (2.73 cm) which was on par with V<sub>25</sub> (2.70 cm), V<sub>24</sub> (2.70 cm), V<sub>22</sub> (2.60 cm), V<sub>20</sub> (2.47 cm), V<sub>21</sub> (2.47 cm), V<sub>26</sub> (2.37 cm), V<sub>19</sub> (2.33 cm), V<sub>28</sub> (2.33 cm), V<sub>12</sub> (2.30 cm) and V<sub>3</sub> (2.27 cm). All varieties were significantly superior to V<sub>9</sub> among all varieties lowest root girth was observed in V<sub>9</sub> (0.77 cm). The same trend was continued throughout the study period, hybrid V<sub>9</sub> recorded minimum root girth in August 2016 (1.07cm), December 2016 (1.40 cm), April 2017 (1.63 cm), August 2017 (1.77 cm), and December 2017 (1.93 cm) and at the end of the study, in March 2018 (Table 8.) (1.93 cm) as well. In August 2016 maximum root girth was observed in V<sub>25</sub> (3.13 cm) V<sub>23</sub> (3.00 cm) respectively, which was on par with V<sub>24</sub> (2.96 cm), V<sub>22</sub> (2.90 cm) and V<sub>20</sub> (2.87 cm), V<sub>26</sub> (2.70 cm). Maximum root girth during December 2016 was observed in V<sub>25</sub> (3.37 cm) while lower values were observed *viz.*, V<sub>9</sub> (1.40 cm) followed by V<sub>11</sub> (1.77 cm), V<sub>16</sub> (1.83 cm) and V<sub>17</sub> (1.90 cm). Almost similar trend was continued during April 2017 and August 2017. During December 2017 maximum root girth was observed in V<sub>25</sub> (3.87 cm) which was on par with V<sub>20</sub> (3.73 cm), V<sub>24</sub> (3.66 cm), V<sub>26</sub> (3.60 cm), V<sub>22</sub>



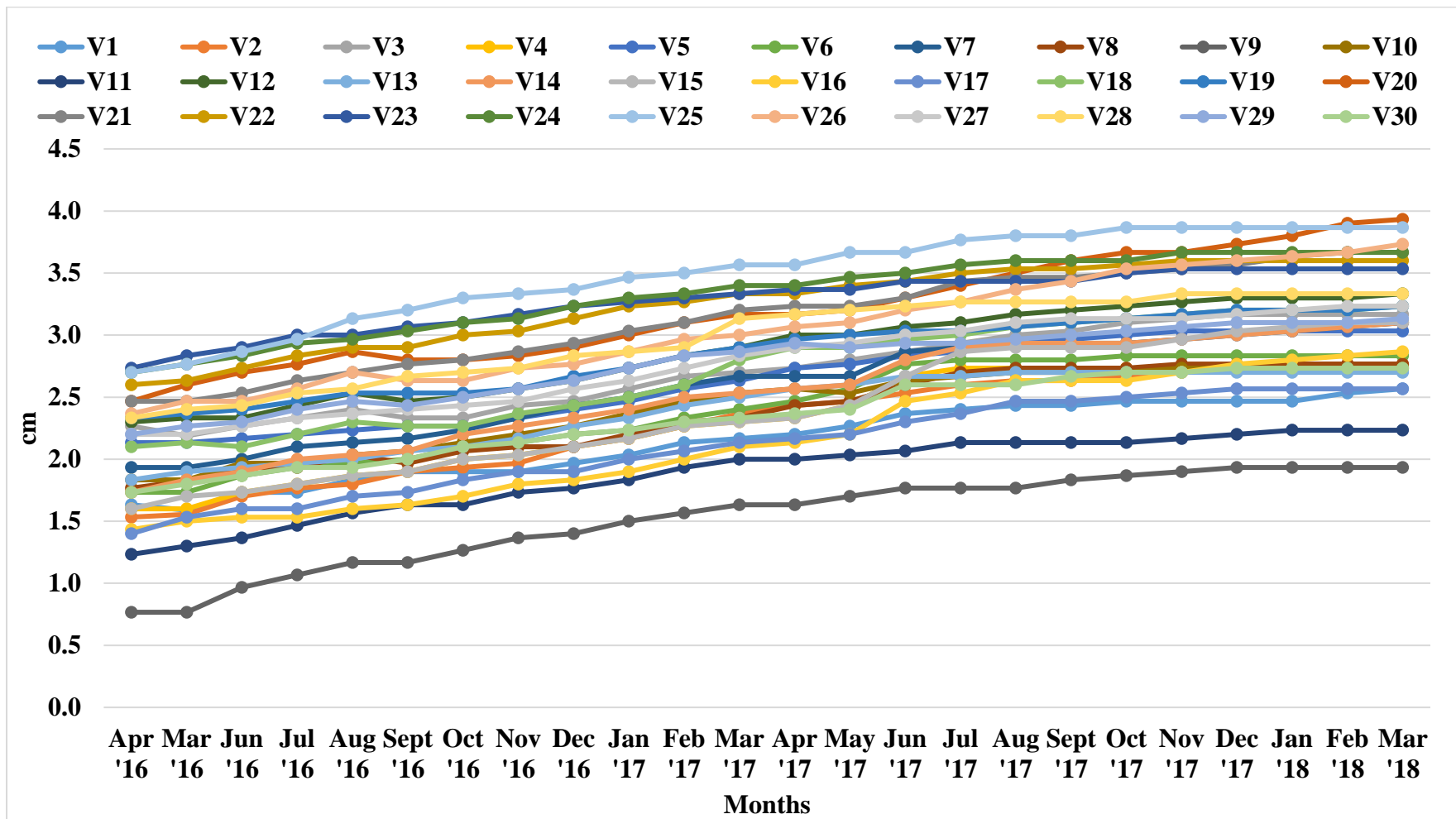


Fig. 14. Root girth of *Ascozentrum* hybrids/varieties during the period 2016-18

(3.60 cm), V<sub>21</sub> (3.57 cm), V<sub>23</sub> (3.53 cm) and V<sub>28</sub> (3.33 cm). At the end of the study (March 2018), no significant variation was exhibited among the hybrids/varieties (Table 8).

#### **4.1.1.4 Floral characters**

Data relating to the flowering characteristics (Plate 2) of selected thirty *Ascocentrum* hybrids/varieties are given in Table 10.

##### **4.1.1.4.1 Spike characters**

Data pertaining to the quantitative spike characters (Plate 3-7) with respect to number of spikes produced per year per plant, spike length, rachis length, flower stalk or peduncle length and girth of spike at base are presented in Table 10.

#### **A. Number of spikes produced per year**

High variation was observed in all the hybrids/varieties with respect to spike characters (Table 10 and Fig. 18). Variety V<sub>25</sub> (7.86) was observed to produce the maximum number of spikes per plant per year which was on par with V<sub>23</sub> (6.33) and V<sub>27</sub> (6.00). Whereas, V<sub>16</sub> (1.00) produced the least number of spike per plant per year which was statistically on par with V<sub>1</sub>, V<sub>2</sub>, V<sub>3</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>17</sub>, V<sub>19</sub>, V<sub>20</sub>, V<sub>21</sub>, V<sub>22</sub>, V<sub>26</sub>, V<sub>28</sub> and V<sub>30</sub>.

#### **B. Length of spike**

Varietal differences were clearly evident with regard to spike length (Table 10 and Fig. 19). The hybrids/varieties varied more significantly with respect to spike length, variety V<sub>23</sub> (54.70 cm) had longest spikes which was on par with V<sub>24</sub> (47.53 cm). Whereas, V<sub>9</sub> (17.33 cm) recorded spike length which was on par with V<sub>1</sub>, V<sub>2</sub>, V<sub>3</sub>, V<sub>6</sub>, V<sub>7</sub>, V<sub>11</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>15</sub>, V<sub>16</sub> and V<sub>17</sub>.

#### **C. Length of rachis**

Variation was noticed with regard to rachis length (Table 10 and Fig. 19) and it was observed to be higher in V<sub>30</sub> (28.80 cm) which was on par with V<sub>8</sub>, V<sub>10</sub>,



(A) Selected hanging orchids in flowering phase



(B) Selected prostrate orchids in flowering phase

**Plate 2. Flowering phase of intergeneric hybrids of *Ascocentrum***



*Ascda. Udomochai* (V<sub>1</sub>)



*Ascda. Kraillerk White* × *V. Sanderiana* (V<sub>2</sub>)



*Ascda. Yip Sum Wah* × *V. JVB* (V<sub>3</sub>)



*Ascda. Kultana* × *V. Bitzs Heartthrob* (V<sub>4</sub>)



*Ascda. Suksamran Sunlight* (V<sub>5</sub>)



*Ascda. Sirichi Fragrance* (V<sub>6</sub>)

**Plate 3. Intergeneric hybrids of *Ascocenda***



*Vasco. Pine River Blue* (V<sub>7</sub>)



*Vasco. Pine River Pink* (V<sub>8</sub>)



*Vasco. Pine Rivers Fuchsia Delight* (V<sub>10</sub>)



*Vasco. Aroonsri Beauty* (V<sub>9</sub>)



*Vasco. Blue Bay White* (V<sub>11</sub>)

**Plate 4. Intergeneric hybrids of *Vascostylis***



*Mok. Walter Oumae Pink (V<sub>12</sub>) Mok. Calypso × V. Dr. Anek (V<sub>13</sub>) Mok. Rassmatozz (V<sub>14</sub>)*



*Mok. Khaw Phiak Suan × Ascda.  
Bicentennial Yellow Spot (V<sub>15</sub>)*

*Mok. Khaw Phiak Suan × Ascda.  
Jiraprapa (V<sub>16</sub>)*

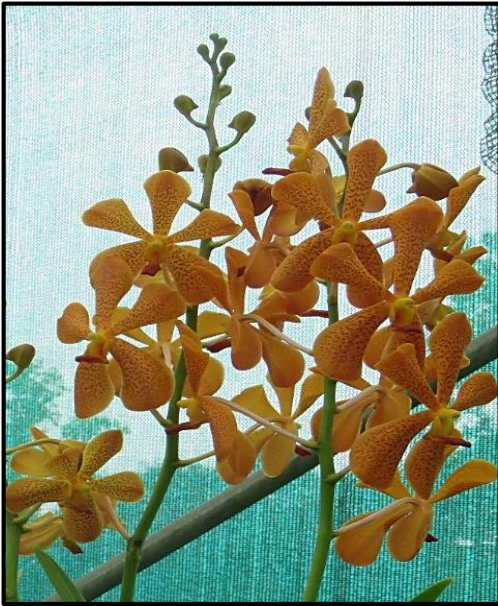


*Mok. Sayan × Ascda.  
Bangkuntein Gold (V<sub>17</sub>)*

*Mok. Calypso Pink (V<sub>18</sub>)*

*Mok. Calypso Jumbo (V<sub>19</sub>)*

**Plate 5. Intergeneric hybrids of *Mokara***



*Mok. Sayan × Ascda. Doung Porn (V<sub>25</sub>)*



*Mok. Omayaiy Orange (V<sub>24</sub>)*



*Mok. Chark Kuan Pink (V<sub>26</sub>)*



*Mok. Chao Praya Sunset  
Yellow Spot (V<sub>20</sub>)*



*Mok. Chao Praya Sunset  
Orange (V<sub>21</sub>)*



*Mok. Sunspot (V<sub>22</sub>)*



*Mok. Omayaiy Yellow (V<sub>23</sub>)*

**Plate 6. Intergeneric hybrids of *Mokara***



*Kag. Youthong Beauty (V<sub>27</sub>)*



*Kag. Christie Low (V<sub>28</sub>)*



*Kag. Boon Ruby (V<sub>29</sub>)*



*Kag. Samrong (V<sub>30</sub>)*

**Plate 7. Intergeneric hybrids of *Kagawara***



V<sub>18</sub>, V<sub>19</sub>, V<sub>21</sub>, V<sub>23</sub>, V<sub>24</sub>, V<sub>25</sub>, V<sub>26</sub>, V<sub>27</sub>, V<sub>28</sub> and V<sub>29</sub>. Rachis length was lowest in V<sub>16</sub> (8.67 cm) which was on par with V<sub>1</sub>, V<sub>2</sub>, V<sub>3</sub>, V<sub>5</sub>, V<sub>7</sub>, V<sub>9</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>15</sub> and V<sub>17</sub>.

#### **D. Length of flower stalk/peduncle length**

Noticeable variations were found with respect to stalk/peduncle length of spikes of selected *Ascocentrum* hybrids/varieties (Table 10 and Fig. 19). With regard to peduncle length also V<sub>23</sub> (27.60 cm) was observed to have a higher value which was on par with V<sub>25</sub> (24.40 cm) and V<sub>24</sub> (23.47 cm), However V<sub>3</sub> (8.47 cm) was observed to have the lowest value. This was closely followed by V<sub>2</sub> (6.63 cm), V<sub>24</sub> (6.53 cm), V<sub>23</sub> (6.43 cm) and V<sub>25</sub> (6.10 cm).

#### **E. Girth of the spike at the base**

Significant variation was observed with respect to the girth of spikes (Table 10) among the hybrids/varieties. With respect to spike girth, V<sub>21</sub> (2.80 cm) was observed to have the highest value which was on par with V<sub>24</sub> (2.73 cm), V<sub>25</sub> (2.73 cm), V<sub>19</sub> (2.70 cm), V<sub>23</sub> (2.567 cm) and V<sub>2</sub> (2.57 cm), whereas, it was low in V<sub>11</sub> (1.17 cm), V<sub>9</sub> (1.30 cm), V<sub>6</sub> (1.40 cm) and V<sub>10</sub> (1.40 cm).

#### **4.1.1.4.2 Floret characters**

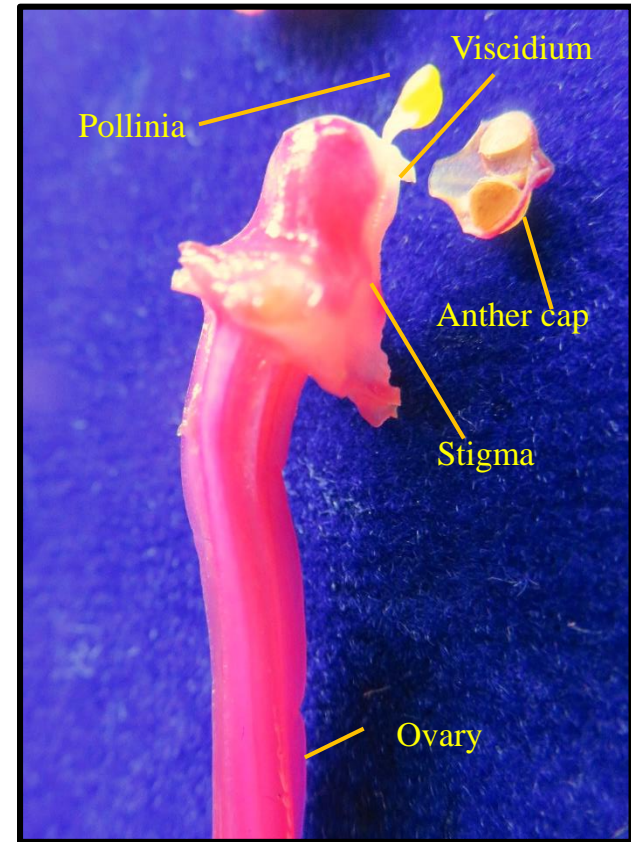
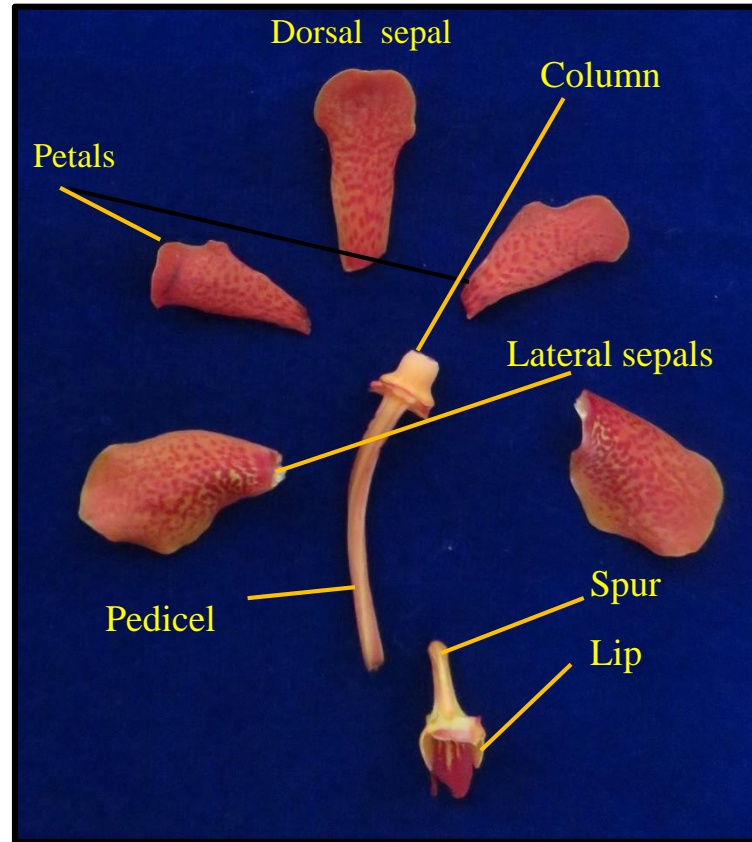
Data pertaining to the quantitative floret characters (Plate 8) with regard to the number of florets/spike, internodal length, pedicel length, length of floret, width of floret, length of lip/labellum and width of lip are presented in Table 10 and also explained Appendix XXV.

#### **A. Number of florets per spike**

High significant variations were noticed with regard to the number of florets per spike in the selected intergeneric hybrids of *Ascocentrum* (Table 10 and Fig. 20). Variety V<sub>11</sub> (40.67) was observed to produce more numbers florets per spike which was on par with V<sub>6</sub> (37.67), whereas, V<sub>15</sub> (5.67) produced the least number of florets/spike, but was on par with V<sub>2</sub>, V<sub>3</sub>, V<sub>4</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>14</sub>, V<sub>16</sub>, V<sub>18</sub>, V<sub>20</sub>, V<sub>21</sub>, V<sub>24</sub> and V<sub>26</sub>.



Lebellum or lip



Column

Plate 8. Floral parts of a typical monopodial orchid (*Kag. Youthong Beauty*)

**Table 10. Floral characters of *Ascoentrum* hybrids/varieties**

Var. No.	Spike characters				Flower characters							
	No./yr./ plant	Spike length (cm)	Rachis length (cm)	Peduncle length (cm)	Spike girth (cm)	No. of florets/spike	Internodal length (cm)	Pedicel length (cm)	Length of flower (cm)	Width of flower (cm)	Lip length (cm)	Lip width (cm)
V <sub>1</sub>	3.00 (1.99)	24.33 (5.02)	12.17 (3.62)	12.17 (3.62)	1.80 (1.67)	20.67 (4.63)	1.73 (1.65)	3.23 (2.06)	3.70 (2.17)	3.47 (2.11)	2.33 (1.83)	1.77 (1.66)
V <sub>2</sub>	2.00 (1.72)	25.00 (5.08)	12.50 (3.66)	12.50 (3.66)	2.57 (1.89)	9.67 (3.26)	1.50 (1.58)	6.63 (2.76)	6.67 (2.77)	6.33 (2.71)	3.43 (2.11)	2.70 (1.92)
V <sub>3</sub>	2.67 (1.88)	22.50 (4.81)	10.87 (3.43)	11.63 (3.51)	2.50 (1.87)	8.00 (2.99)	2.17 (1.78)	8.47 (3.08)	9.33 (3.21)	9.00 (3.16)	4.80 (2.41)	3.20 (2.05)
V <sub>4</sub>	3.00 (1.99)	31.87 (5.73)	15.00 (4.00)	16.87 (4.22)	2.17 (1.78)	9.00 (3.16)	1.80 (1.67)	5.73 (2.59)	6.30 (2.70)	5.90 (2.63)	2.37 (1.84)	2.50 (1.87)
V <sub>5</sub>	3.00 (1.99)	27.33 (5.32)	12.33 (3.64)	15.00 (4.00)	2.20 (1.79)	16.67 (4.20)	5.00 (2.44)	3.37 (2.09)	6.43 (2.73)	6.23 (2.69)	2.53 (1.88)	1.37 (1.54)
V <sub>6</sub>	5.00 (2.45)	24.40 (5.03)	14.50 (3.94)	9.90 (3.28)	1.40 (1.55)	37.67 (6.21)	1.27 (1.51)	2.80 (1.95)	2.77 (1.94)	2.70 (1.92)	3.10 (2.03)	2.17 (1.78)
V <sub>7</sub>	2.67 (1.91)	22.67 (4.86)	14.00 (3.86)	8.67 (3.11)	1.57 (1.60)	14.67 (3.96)	1.47 (1.57)	2.40 (1.84)	2.53 (1.88)	2.43 (1.85)	2.63 (1.91)	1.63 (1.62)
V <sub>8</sub>	3.67 (2.16)	30.00 (5.56)	17.50 (4.25)	12.50 (3.66)	1.67 (1.63)	33.33 (5.85)	1.37 (1.54)	2.73 (1.93)	2.77 (1.94)	2.70 (1.92)	2.63 (1.91)	1.53 (1.59)
V <sub>9</sub>	3.00 (1.99)	17.33 (4.28)	11.60 (3.55)	5.73 (2.59)	1.30 (1.52)	17.00 (4.23)	1.33 (1.53)	3.00 (2.00)	2.80 (1.95)	2.53 (1.88)	2.57 (1.89)	1.20 (1.48)
V <sub>10</sub>	2.33 (1.82)	34.33 (5.93)	23.00 (4.89)	11.33 (3.49)	1.40 (1.55)	32.67 (5.78)	1.07 (1.44)	2.53 (1.88)	2.73 (1.93)	2.37 (1.83)	1.80 (1.67)	1.33 (1.53)
V <sub>11</sub>	3.00 (1.99)	24.33 (5.03)	15.50 (4.06)	8.83 (3.13)	1.17 (1.47)	40.67 (6.45)	0.70 (1.30)	2.23 (1.80)	2.27 (1.81)	2.13 (1.77)	2.43 (1.85)	1.87 (1.69)
V <sub>12</sub>	2.33 (1.82)	25.00 (5.09)	12.00 (3.60)	13.00 (3.74)	1.97 (1.72)	8.33 (3.05)	1.77 (1.66)	3.70 (2.17)	6.83 (2.80)	5.67 (2.58)	2.90 (1.97)	1.23 (1.49)
V <sub>13</sub>	2.33 (1.81)	24.80 (5.07)	11.97 (3.59)	12.83 (3.72)	2.10 (1.76)	10.67 (3.41)	1.97 (1.72)	3.67 (2.16)	5.83 (2.61)	5.80 (2.60)	3.17 (2.04)	1.53 (1.59)
V <sub>14</sub>	4.33 (2.29)	29.20 (5.50)	14.43 (3.93)	14.77 (3.97)	2.10 (1.76)	7.00 (2.83)	1.90 (1.70)	5.00 (2.45)	6.60 (2.76)	6.47 (2.73)	2.37 (1.84)	1.60 (1.61)
V <sub>15</sub>	3.67 (2.14)	24.33 (5.00)	11.33 (3.49)	13.00 (3.72)	1.63 (1.62)	5.67 (2.56)	2.77 (1.94)	3.80 (2.19)	6.40 (2.72)	6.30 (2.70)	2.60 (1.90)	1.30 (1.52)
V <sub>16</sub>	1.00 (1.41)	20.00 (4.58)	8.67 (3.09)	11.33 (3.51)	2.30 (1.82)	7.00 (2.83)	2.67 (1.91)	4.87 (2.42)	5.70 (2.59)	5.23 (2.50)	1.83 (1.68)	1.23 (1.49)

Table 10. continued

Var. no.	Spike characters					Flower characters						
	No./yr./plant	Spike length (cm)	Rachis length (cm)	Peduncle length (cm)	Spike girth (cm)	No. of florets/spike	Internodal length (cm)	Pedicel length (cm)	Length of flower (cm)	Width of flower (cm)	Lip length (cm)	Lip width (cm)
V <sub>17</sub>	3.00 (1.99)	24.00 (5.00)	13.00 (3.74)	11.00 (3.46)	1.67 (1.63)	13.67 (3.83)	1.83 (1.68)	3.77 (2.18)	7.47 (2.91)	6.23 (2.69)	2.70 (1.92)	1.90 (1.70)
V <sub>18</sub>	4.00 (2.23)	28.00 (5.39)	17.60 (4.31)	10.40 (3.38)	2.00 (1.73)	11.33 (3.51)	2.63 (1.91)	4.47 (2.34)	4.93 (2.44)	4.83 (2.41)	2.73 (1.93)	2.33 (1.83)
V <sub>19</sub>	1.63 (1.62)	35.53 (6.03)	19.03 (4.46)	16.50 (4.18)	2.70 (1.92)	24.33 (5.01)	2.53 (1.88)	5.97 (2.64)	6.73 (2.78)	6.33 (2.71)	2.90 (1.98)	2.70 (1.92)
V <sub>20</sub>	2.33 (1.82)	33.97 (5.89)	16.03 (4.12)	17.93 (4.32)	2.33 (1.83)	10.67 (3.40)	2.37 (1.84)	5.90 (2.63)	5.70 (2.59)	6.50 (2.74)	2.30 (1.82)	2.00 (1.73)
V <sub>21</sub>	1.27 (1.50)	35.43 (6.00)	19.00 (4.45)	16.43 (4.15)	2.80 (1.95)	10.43 (3.38)	2.57 (1.89)	5.57 (2.56)	7.57 (2.93)	7.37 (2.89)	3.20 (2.05)	2.73 (1.93)
V <sub>22</sub>	2.87 (1.95)	34.00 (5.89)	16.03 (4.12)	17.97 (4.34)	2.50 (1.87)	13.77 (3.83)	1.87 (1.69)	4.67 (2.38)	6.60 (2.76)	6.20 (2.68)	2.53 (1.88)	1.53 (1.59)
V <sub>23</sub>	6.33 (2.70)	54.70 (7.45)	27.10 (5.29)	27.60 (5.33)	2.57 (1.89)	19.33 (4.49)	2.87 (1.97)	6.43 (2.72)	8.10 (3.02)	7.57 (2.93)	2.63 (1.91)	1.67 (1.63)
V <sub>24</sub>	5.00 (2.43)	47.53 (6.96)	24.07 (5.00)	23.47 (4.92)	2.73 (1.93)	12.10 (3.62)	3.67 (2.16)	6.53 (2.74)	7.77 (2.96)	7.43 (2.90)	2.40 (1.84)	1.70 (1.64)
V <sub>25</sub>	7.87 (2.97)	42.60 (6.60)	18.20 (4.38)	24.40 (5.04)	2.73 (1.93)	28.73 (5.44)	3.77 (2.18)	6.10 (2.66)	7.00 (2.83)	6.73 (2.78)	2.40 (1.84)	1.50 (1.58)
V <sub>26</sub>	1.33 (1.52)	35.17 (6.00)	18.27 (4.39)	16.90 (4.18)	2.40 (1.84)	7.87 (2.97)	2.33 (1.83)	5.83 (2.61)	7.00 (2.83)	6.67 (2.77)	2.40 (1.84)	1.50 (1.58)
V <sub>27</sub>	6.00 (2.62)	39.50 (6.34)	24.37 (5.03)	15.13 (3.96)	2.03 (1.74)	18.43 (4.38)	2.67 (1.92)	5.87 (2.62)	6.23 (2.69)	6.30 (2.70)	2.20 (1.79)	1.50 (1.58)
V <sub>28</sub>	3.10 (2.02)	42.87 (6.62)	25.53 (5.14)	17.33 (4.24)	1.90 (1.70)	24.20 (5.01)	1.93 (1.71)	4.60 (2.37)	3.80 (2.19)	3.93 (2.22)	2.63 (1.91)	3.77 (2.03)
V <sub>29</sub>	3.33 (2.08)	28.87 (5.46)	17.10 (4.25)	11.77 (3.57)	1.67 (1.63)	20.53 (4.64)	1.73 (1.65)	4.33 (2.31)	3.43 (2.11)	3.33 (2.08)	2.37 (1.84)	1.30 (1.51)
V <sub>30</sub>	2.00 (1.72)	40.87 (6.47)	28.80 (5.46)	12.07 (3.61)	1.67 (1.63)	30.07 (5.57)	1.47 (1.57)	3.60 (2.14)	3.33 (2.08)	3.13 (2.03)	1.87 (1.69)	1.87 (1.69)
C.D.@5%	0.418	0.712	0.547	0.715	0.071	0.60	0.133	0.125	0.106	0.117	0.097	0.303
SE(m)±	0.147	0.251	0.193	0.252	0.025	0.211	0.047	0.044	0.037	0.041	0.034	0.107
C.V.	12.654	7.762	8.029	11.339	2.497	8.828	4.613	3.256	2.577	2.891	3.111	11.025

The data in parenthesis indicate square root transformed value

## **B. Internodal length**

Variation was also noticed with respect to internodal length between the florets (Table 10 and Fig. 21). Internodal length was highest in V<sub>5</sub> (5.00 cm) and was lowest in V<sub>11</sub> (0.70 cm).

## **C. Length of the pedicel (stalk of single floret)**

A good amount of variation was noticed with respect to the length of the pedicel (Table 10). Hybrid V<sub>3</sub> (8.47 cm) was observed to have highest pedicel length followed by V<sub>2</sub> (6.63 cm), V<sub>24</sub> (6.53 cm), V<sub>23</sub> (6.43 cm) and V<sub>25</sub> (6.10 cm), whereas, it was the lowest in V<sub>11</sub> (2.23 cm) which was on par with V<sub>7</sub> (2.40 cm), V<sub>10</sub> (2.53 cm), V<sub>8</sub> (2.73 cm) and V<sub>6</sub> (2.80 cm).

## **D. Length and breadth of floret**

Slight variation was also noticed in *Ascocentrum* hybrids/varieties with respect to the length and breadth of florets (Table 10). Hybrid V<sub>3</sub> (*Ascda.* Kultana × *V.* Bitz's Heartthrob) also recorded the highest length of floret (9.33 cm) and breadth of flower (9.00 cm), whereas, lowest length and breadth of flower (2.27 cm and 2.13 cm, respectively) was observed in V<sub>11</sub> (*Vasco.* Blue Bay White).

## **E. Length and width of the labellum/lip**

Difference was noticed with respect to the length of labellum among varieties (Table 10). The width of labellum also showed marked variation among the varieties. Slight variation was detected regarding lip length and it ranged from 1.80 cm (V<sub>10</sub>) to 4.80 cm (V<sub>3</sub>). In which V<sub>3</sub> found to have the highest value even though it was on par with V<sub>2</sub>, V<sub>21</sub>, V<sub>13</sub>, V<sub>6</sub>, V<sub>12</sub>, V<sub>19</sub>, V<sub>18</sub>, V<sub>17</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>23</sub>, V<sub>28</sub>, V<sub>15</sub>, V<sub>9</sub>, V<sub>22</sub>, V<sub>5</sub> and V<sub>11</sub>. While the lowest value was observed in V<sub>10</sub> which on par with V<sub>30</sub> and V<sub>16</sub>. Variation was observed with respect lip width and many of the hybrids/varieties were observed to have near to similar and lip width, which ranged from 1.20 cm (V<sub>9</sub>) to 3.77 cm (V<sub>28</sub>). High value was observed in V<sub>28</sub> and which was not found significantly different than V<sub>3</sub>, V<sub>21</sub>, V<sub>19</sub>, V<sub>2</sub>, V<sub>4</sub> and V<sub>18</sub>.

However, the lowest value was observed in V<sub>9</sub> which was not found significantly different from rest of the varieties except V<sub>9</sub> and V<sub>21</sub>.

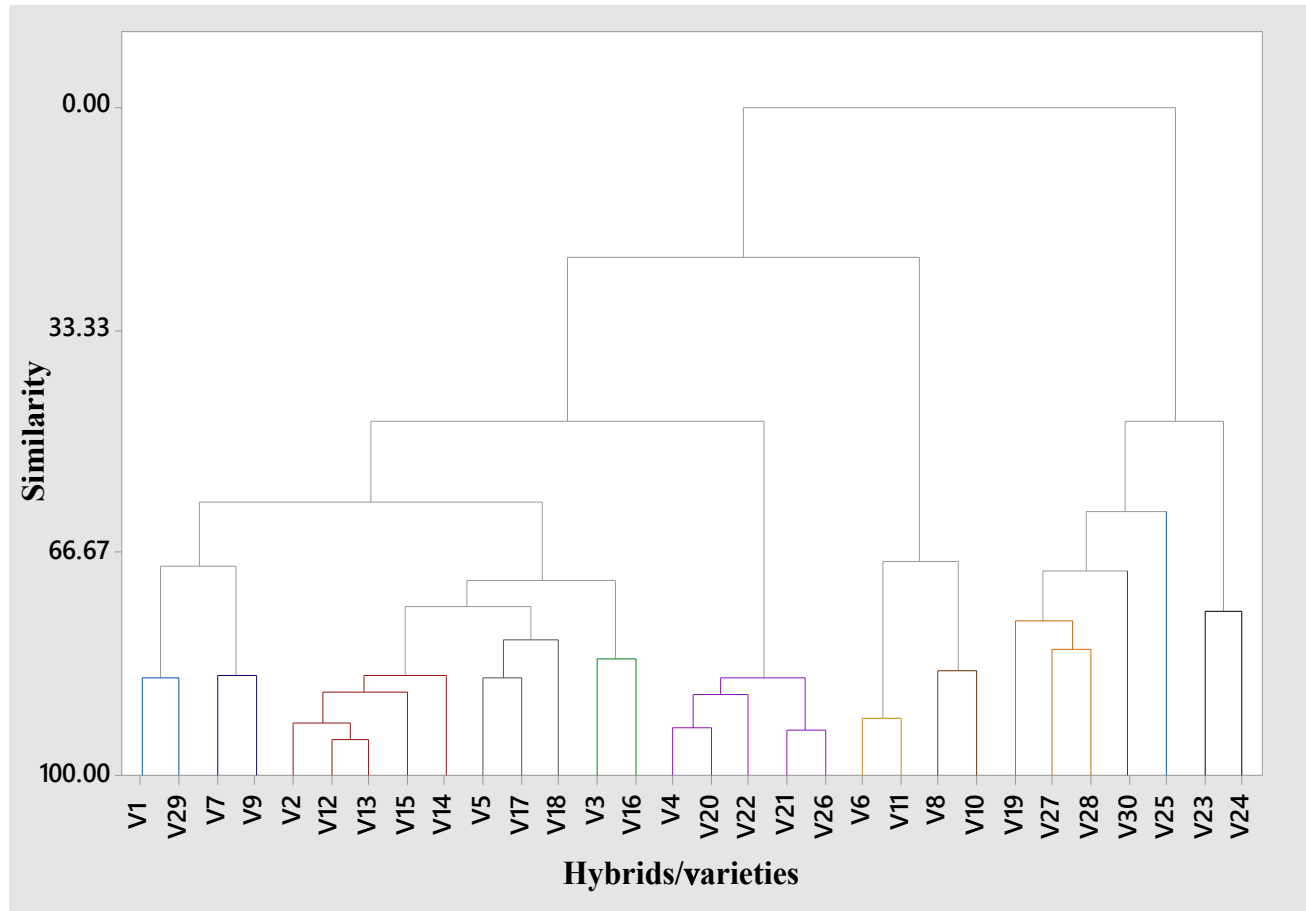
#### 4.1.1.4.3 Cluster analysis for floral characters

The data pertaining to the cluster analysis are presented in Table 11 to 13 and depicted in Fig. 15. Cluster analysis with 14 different floral characters revealed 12 clusters at 75 per cent similarity. Cluster 2 and 5 observed to have five members each whereas, cluster 11 and 12 with only one member in each (Table 11 and Fig. 15). It was observed that cluster 2 and cluster 5 were less similar with each other with an inter-cluster distance of 6.27, whereas, the highest inter-cluster distance was observed in cluster 6 and cluster 10 (41.47) (Table 12).

**Table 11. Members of different clusters in *Ascocentrum* hybrids**

Sl. No.	Cluster	No. of members	Members of the cluster
1	Cluster1	2	V <sub>1</sub> , V <sub>29</sub>
2	Cluster2	5	V <sub>2</sub> , V <sub>12</sub> , V <sub>13</sub> , V <sub>14</sub> , V <sub>15</sub> ,
3	Cluster3	2	V <sub>3</sub> , V <sub>16</sub>
4	Cluster4	5	V <sub>4</sub> , V <sub>20</sub> , V <sub>21</sub> , V <sub>22</sub> , V <sub>26</sub>
5	Cluster5	3	V <sub>5</sub> , V <sub>17</sub> , V <sub>18</sub>
6	Cluster6	2	V <sub>6</sub> , V <sub>11</sub>
7	Cluster7	2	V <sub>7</sub> , V <sub>9</sub>
8	Cluster8	2	V <sub>8</sub> , V <sub>10</sub>
9	Cluster9	3	V <sub>19</sub> , V <sub>27</sub> , V <sub>28</sub>
10	Cluster10	2	V <sub>23</sub> , V <sub>24</sub>
11	Cluster11	1	V <sub>25</sub>
12	Cluster12	1	V <sub>30</sub>

Cluster mean values are presented in Table 13. Cluster 10 was observed to have high mean values for spike length (51.12 cm), flower length (7.94 cm) and flower width (7.50 cm). Cluster 11 was observed with high mean values for the number of spikes per plant per year and internodal length (3.77 cm) and also have highest flower length (7.00 cm) and flower width (6.73 cm) next to cluster 10. Whereas, cluster 6 which included V<sub>6</sub> (*Ascda.* Sirichi Fragrance) and V<sub>11</sub> (*Vasco* Blue Bay White) was found to have the lowest internodal length with the highest value for number of florets per spike. This cluster also observed to have low



**Fig. 15.** Dendrogram showing clustering of 30 intergeneric hybrids of *Ascozentrum*

**Table 12. Inter cluster distance between 12 clusters in *Ascocentrum* hybrids/varieties**

Cluster No.	Cluster 1	Cluster 2	Cluster 3	Cluster 4	Cluster 5	Cluster 6	Cluster 7	Cluster 8	Cluster 9	Cluster 10	Cluster 11	Cluster 12
Cluster1	0.00											
Cluster2	13.28	0.00										
Cluster3	16.32	6.24	0.00									
Cluster4	14.77	10.63	16.11	0.00								
Cluster5	7.80	6.27	10.18	10.47	0.00							
Cluster6	19.04	31.85	33.42	32.24	26.16	0.00						
Cluster7	9.78	12.56	12.79	19.71	10.06	23.98	0.00					
Cluster8	14.81	27.35	30.74	24.47	21.50	11.61	22.82	0.00				
Cluster9	16.33	22.53	27.47	14.60	18.26	25.50	25.11	14.86	0.00			
Cluster10	31.33	32.32	37.73	21.93	30.62	41.47	39.35	30.84	17.08	0.00		
Cluster11	23.34	29.91	34.43	22.35	25.99	27.47	32.91	19.15	12.89	17.45	0.00	
Cluster12	22.27	31.58	36.11	25.14	26.49	23.68	30.35	12.67	11.57	23.87	18.53	0.00

**Table 13. Mean values of floral characters for clusters in *Ascocentrum* hybrids/varieties**

Variable	Cluster 1	Cluster 2	Cluster 3	Cluster 4	Cluster 5	Cluster 6	Cluster 7	Cluster 8	Cluster 9	Cluster 10	Cluster 11	Cluster 12
No. of spikes/ yr./ plant	3.17	2.93	1.84	2.16	3.33	4.00	2.84	3.00	3.58	5.67	7.87	2.00
Spike length (cm)	26.60	25.67	21.25	34.09	26.44	24.37	20.00	32.17	39.30	51.12	42.60	40.87
Rachis length (cm)	14.64	12.45	9.77	16.87	14.31	15.00	12.80	20.25	22.98	25.59	18.20	28.80
Peduncle length (cm)	11.97	13.22	11.48	17.22	12.13	9.37	7.20	11.92	16.32	25.54	24.40	12.07
Spike girth (cm)	1.74	2.07	2.40	2.44	1.96	1.29	1.44	1.54	2.21	2.65	2.73	1.67
No. of florets / spike	20.60	8.27	7.50	10.35	13.89	39.17	15.84	33.00	22.32	15.72	28.73	30.07
Internodal length (cm)	1.73	1.98	2.42	2.19	3.15	0.99	1.40	1.22	2.38	3.27	3.77	1.47
Pediceal length (cm)	3.78	4.56	6.67	5.54	3.87	2.52	2.70	2.63	5.48	6.48	6.10	3.60
Length of flower (cm)	3.57	6.47	7.52	6.63	6.28	2.52	2.67	2.75	5.59	7.94	7.00	3.33
Width of flower (cm)	3.40	6.11	7.12	6.53	5.76	2.42	2.48	2.54	5.52	7.50	6.73	3.13
Lip length (cm)	2.35	2.89	3.32	2.56	2.65	2.77	2.60	2.22	2.58	2.52	2.40	1.87
Lip width (cm)	1.54	1.67	2.22	2.05	1.87	2.02	1.42	1.43	2.66	1.69	1.50	1.87



flower length and width (2.52 cm and 2.42 cm, respectively). This indicated that these varieties produced flowers in the dense bunch.

#### **4.1.2 Phenological characters**

The data pertaining to the phenological characters *viz.*, interval of leaf production, leaf longevity, spike emergence to the opening of first floret, spike emergence to 75 per cent opening, spike emergence to complete (100 %) opening, spike longevity on the plant and life of individual floret on plant, were measured and expressed in days and presented in Table 14.

##### **4.1.2.1 Interval of leaf production**

A wide range of variation was observed among the hybrids/varieties in the character interval of leaf production. It ranged from 32.17 days to 108.44 days. The lowest value, *i.e.* below 40 days (Table 14), was observed in V<sub>25</sub> (32.17 days), V<sub>23</sub> (33.89 days) and V<sub>22</sub> (35.80 days). Whereas, two hybrids *i.e.*, V<sub>16</sub> and V<sub>17</sub> observed to have an interval of leaf production more than 100 days (104.91 and 108.44 days).

##### **4.1.2.2 Leaf longevity**

Noticeable variation was observed with respect to leaf longevity. Leaf longevity ranged from 129.02 to 292.20 days. Twelve number hybrids were found to have leaf longevity above 200 days *viz.*, V<sub>1</sub> (201.67 days), V<sub>4</sub> (202.70 days), V<sub>8</sub> (231.61 days), V<sub>10</sub> (261.47 days), V<sub>11</sub> (247.10 days), V<sub>12</sub> (277.04 days), V<sub>14</sub> (216.86 days), V<sub>15</sub> (230.29 days), V<sub>17</sub> (229.16 days), V<sub>19</sub> (235.06 days), V<sub>22</sub> (206.42 days) and V<sub>24</sub> (220.66), and it was highest in V<sub>9</sub> (292.20 days). Whereas, V<sub>20</sub> (129.02 days) was observed to have short leaf longevity (Table 14).

##### **4.1.2.3 Days from spike emergence to first floret open**

Significant variation was also observed with respect to the opening of the first floret (Table 14). The variety V<sub>10</sub> was early with respect to floret opening

and had taken 14.99 days to open the first floret. Whereas, hybrid V<sub>3</sub> had taken maximum days to open the first floret (40.18 days).

**Table 14. Phenological leaf and floral characters of *Ascozentrum* hybrids**

Var. No.	Leaf characters (days)		Floral characters				
			Days from spike emergence to			Spike longevity (days)	Life of individual floret (days)
	Interval of leaf production	Leaf longevity	First floret open	75% florets open	100% floret open		
V <sub>1</sub>	83.34	201.67	20.44	30.43	37.02	19.17	18.22
V <sub>2</sub>	80.71	186.93	19.00	28.00	36.40	20.70	16.80
V <sub>3</sub>	88.71	156.13	40.18	41.21	42.50	18.54	18.32
V <sub>4</sub>	73.02	202.70	20.99	24.49	27.52	22.73	19.33
V <sub>5</sub>	82.98	141.70	30.43	38.91	44.57	22.67	19.96
V <sub>6</sub>	77.00	172.30	25.55	34.57	41.71	23.50	18.44
V <sub>7</sub>	97.95	155.21	20.64	26.69	31.55	27.37	20.55
V <sub>8</sub>	83.93	231.61	34.02	44.81	50.99	28.50	28.30
V <sub>9</sub>	78.01	292.20	17.60	22.50	31.50	17.67	15.64
V <sub>10</sub>	85.31	261.47	14.99	18.50	28.50	18.67	15.79
V <sub>11</sub>	79.01	247.10	22.99	28.98	29.80	16.96	14.40
V <sub>12</sub>	79.63	277.04	24.55	34.20	45.60	29.50	25.70
V <sub>13</sub>	98.19	140.18	22.10	32.70	42.30	25.80	20.26
V <sub>14</sub>	81.42	216.86	19.88	30.47	39.08	24.44	19.80
V <sub>15</sub>	97.08	230.29	22.98	28.28	28.94	22.32	19.22
V <sub>16</sub>	104.91	184.51	24.58	32.77	30.51	23.77	22.33
V <sub>17</sub>	108.44	229.16	21.48	28.50	28.21	20.67	19.90
V <sub>18</sub>	63.06	143.69	16.40	26.50	38.40	24.76	20.50
V <sub>19</sub>	49.44	235.06	17.40	30.00	42.70	28.75	24.40
V <sub>20</sub>	59.82	129.02	18.80	34.08	48.80	32.58	27.50
V <sub>21</sub>	41.46	145.85	19.50	33.50	45.80	31.80	25.90
V <sub>22</sub>	35.80	206.42	19.40	32.80	43.50	33.02	23.56
V <sub>23</sub>	33.89	191.61	23.40	40.11	50.28	34.80	28.80
V <sub>24</sub>	43.42	220.66	17.66	30.20	43.21	32.47	23.40
V <sub>25</sub>	32.17	161.78	25.40	39.80	51.42	37.40	29.04
V <sub>26</sub>	49.45	160.06	26.40	38.50	52.40	36.77	31.50
V <sub>27</sub>	56.68	144.33	28.40	38.23	51.56	35.89	30.40
V <sub>28</sub>	50.82	204.92	24.30	33.20	47.15	31.90	27.10
V <sub>29</sub>	60.49	188.95	23.40	31.33	46.53	29.79	25.44
V <sub>30</sub>	59.09	249.09	23.11	32.11	45.18	28.70	26.44

#### **4.1.2.4 Days from spike emergence to opening 75 per cent florets**

Visible differences were observed with respect to the opening of 75 per cent florets (Table 14). The variety of V<sub>10</sub> was observed to take minimum days to open 75 per cent florets (Table 14) and had taken 18.50 days. Whereas, variety V<sub>8</sub> (44.81 days) had taken maximum days to open 75 per cent florets.

#### **4.1.2.5 Days from spike emergence to opening of 100 per cent florets**

Noticeable differences were observed with respect to the opening of all the florets (100 %). The hybrid V<sub>4</sub> was earliest with respect to the opening of all the floret and had taken 27.52 days (Table 14). Whereas, V<sub>26</sub> hybrid had taken the maximum days to open all florets (52.40 days).

#### **4.1.2.6 Spike longevity on the plant (days)**

Variation was observed with respect to spike longevity. It ranged from 16.96 days to 37.40 days (Table 14). Three varieties *viz.*, V<sub>25</sub> (37.40 days), V<sub>26</sub> (36.77 days) and V<sub>27</sub> (35.89 days) were observed to have spike longevity more than 35 days. Variety V<sub>11</sub> was observed with the minimum spike longevity on the plant (16.96 days).

#### **4.1.2.7 Life of individual floret on the spike (days)**

Evident variation was observed with respect to the life of individual floret on the spike (Table 14). It ranged from 14.40 days to 31.50 days. It was highest in V<sub>26</sub> (31.50 days), and lowest in V<sub>11</sub> (14.40 days).

### **4.1.3 Post-harvest characters**

The data pertaining to the post-harvest characters *viz.*, fresh weight of spike, wilting of the first floret, life span each floret, spike longevity in a vase, water uptake and physiological loss in weight are presented in Table 15 and depicted in Fig. 22-24.

#### 4.1.3.1 Fresh weight of the spike (g)

Significant difference was observed in fresh weight of spike among *Vanda* hybrids/varieties (Table 15 and Fig. 22). Fresh weight of spike was highest in V<sub>23</sub> (53.29 g) which was on par with V<sub>25</sub> (45.67 g) and V<sub>24</sub> (45.67 g). And was significantly superior to remaining other varieties/hybrids. The hybrid V<sub>17</sub> (*Mok. Sayan* × *Ascda. Bangkuntein Gold*) recorded lowest fresh weight of the spike (9.81 g) even though it was on par with variety V<sub>1</sub> (18.43 g), V<sub>4</sub> (19.33 g), V<sub>6</sub> (12.64 g), V<sub>7</sub> (12.88 g), V<sub>9</sub> (12.50 g), V<sub>13</sub> (18.60 g), V<sub>14</sub> (16.00 g), V<sub>15</sub> (12.62 g), V<sub>16</sub> (13.68 g), V<sub>18</sub> (18.00 g) and V<sub>27</sub> (19.78 g).

#### 4.1.3.2 Wilting of first floret (days)

Less variation was noted among the varieties/ hybrids with regard to the time taken for wilting of the first floret (Table 15 and Fig. 23). The period taken for wilting of the first floret was maximum in V<sub>27</sub> (*Kag. Youthong Beauty*) (25.00 days) which was on par with V<sub>8</sub> (19.00 days), V<sub>12</sub> (22.33 days), V<sub>13</sub> (18.67 days), V<sub>23</sub> (21.80 days), V<sub>24</sub> (19.57 days) and V<sub>25</sub> (19.91 days). Variety V<sub>27</sub> was significantly superior over remaining hybrids. Whereas, V<sub>17</sub> (*Mok. Sayan* × *Ascda. Bangkuntein Gold*) (10.91 days) took the minimum days for wilting of first floret. However, it was on par with V<sub>1</sub> (12.03 days), V<sub>2</sub> (13.67 days), V<sub>3</sub> (14.00 days), V<sub>4</sub> (13.33 days), V<sub>6</sub> (14.00 days), V<sub>7</sub> (16.31 days), V<sub>9</sub> (13.11 days), V<sub>10</sub> (12.67 days), V<sub>11</sub> (13.89 days), V<sub>14</sub> (14.00 days), V<sub>15</sub> (16.92 days), V<sub>16</sub> (14.67 days), V<sub>18</sub> (15.50 days), V<sub>19</sub> (17.00 days), V<sub>21</sub> (14.84 days), V<sub>29</sub> (14.63 days) and V<sub>30</sub> (13.18 days).

#### 4.1.3.3 Floret life span (days)

Variation was not observed with respect to life span of floret (Table 15 and Fig. 23). The variety V<sub>26</sub> (*Mok. Chark Kuan Pink*) (28.44 days) was observed to have the maximum value which was on par with V<sub>27</sub> (27.67 days), V<sub>8</sub> (27.50 days), V<sub>12</sub> (27.17 days), V<sub>20</sub> (23.19 days), V<sub>21</sub> (23.14 days) and V<sub>30</sub> (21.75 days). Whereas, V<sub>11</sub> (*Vasco. Blue Bay White*) (13.76 days) had the minimum floret life even though it was on par with 21 other hybrids/varieties.

#### 4.1.3.4 Spike longevity in a vase (days)

Noticeable variation was noticed with respect to spike longevity in *Ascozentrum* varieties/hybrids (Table 15 and Fig. 23). Variety V<sub>26</sub> (31.65 days) recorded significantly maximum spike longevity among all the selected varieties/hybrids which was on par with V<sub>27</sub> (31.33 days), V<sub>25</sub> (31.00 days), V<sub>20</sub> (29.11 days), V<sub>12</sub> (28.80 days), V<sub>28</sub> (27.47 days), V<sub>8</sub> (26.93 days), V<sub>24</sub> (26.87 days), V<sub>23</sub> (26.80 days), V<sub>30</sub> (26.28 days), V<sub>7</sub> (26.19 days) and V<sub>29</sub> (26.00 days). The variety V<sub>11</sub> (*Vasco*. Blue Bay White) (16.87 days) recorded the minimum value but was on par with 11 other hybrids/varieties.

#### 4.1.3.5 Water uptake (ml)

Less variation was observed with respect to water uptake (Table 15 and Fig. 24). Variety V<sub>23</sub> was observed to uptake more amount of water (14.00 ml) which was on par with 20 other varieties. The variety V<sub>9</sub> (*Vasco*. Aroonsri Beauty) (5.29 ml) was observed to absorb the minimum amount of water which was on par with 23 other hybrids/varieties.

**Table 15. Post-harvest floral characters of *Ascozentrum* hybrids/varieties**

Var. No.	Fresh wt. of spike (g)	Wilting of first floret (days)	Floret life span (days)	Spike longevity in vase	Water uptake (ml)	Physiological loss in wt.(g)
V <sub>1</sub>	18.43 (4.394)	12.03 (3.573)	16.31 (4.155)	18.33 (4.382)	7.00 (2.799)	10.06 (3.268)
V <sub>2</sub>	21.03 (4.688)	13.67 (3.823)	15.83 (4.1)	18.67 (4.434)	6.33 (2.698)	12.59 (3.683)
V <sub>3</sub>	25.07 (5.106)	14.00 (3.872)	16.17 (4.142)	18.00 (4.358)	13.00 (3.74)	13.20 (3.767)
V <sub>4</sub>	19.33 (4.494)	13.33 (3.782)	17.00 (4.242)	21.47 (4.732)	5.33 (2.515)	6.89 (2.801)
V <sub>5</sub>	26.06 (5.2)	17.76 (4.269)	18.92 (4.396)	24.01 (4.984)	8.57 (3.087)	10.23 (3.327)
V <sub>6</sub>	12.64 (3.678)	14.00 (3.863)	14.96 (3.982)	20.17 (4.595)	6.34 (2.691)	10.49 (3.377)
V <sub>7</sub>	12.88 (3.72)	16.31 (4.155)	19.12 (4.486)	26.19 (5.213)	7.13 (2.82)	5.89 (2.611)
V <sub>8</sub>	26.88 (5.273)	19.00 (4.452)	27.50 (5.338)	26.93 (5.284)	7.10 (2.833)	9.57 (3.226)
V <sub>9</sub>	12.50 (3.671)	13.11 (3.719)	14.71 (3.949)	17.17 (4.258)	5.29 (2.482)	5.14 (2.467)

**Table 14. continued**

<b>Var. No.</b>	<b>Fresh wt. of spike (g)</b>	<b>Wilting of first floret (days)</b>	<b>Floret life span (days)</b>	<b>Spike longevity in vase</b>	<b>Water uptake (ml)</b>	<b>Physiological loss in wt.(g)</b>
V <sub>10</sub>	22.90 (4.875)	12.67 (3.678)	15.50 (4.056)	18.67 (4.434)	10.00 (3.266)	11.13 (3.438)
V <sub>11</sub>	21.44 (4.72)	13.89 (3.846)	13.76 (3.787)	16.87 (4.217)	11.06 (3.459)	13.91 (3.861)
V <sub>12</sub>	24.17 (5.004)	22.33 (4.829)	27.17 (5.302)	28.80 (5.454)	7.93 (2.977)	11.31 (3.491)
V <sub>13</sub>	18.60 (4.426)	18.67 (4.426)	20.67 (4.646)	22.67 (4.855)	7.00 (2.804)	7.33 (2.875)
V <sub>14</sub>	16.00 (4.099)	14.00 (3.872)	18.33 (4.397)	20.40 (4.626)	8.00 (2.976)	6.60 (2.739)
V <sub>15</sub>	12.62 (3.687)	16.92 (4.223)	16.84 (4.217)	21.41 (4.729)	9.34 (3.193)	7.98 (2.9)
V <sub>16</sub>	13.68 (3.826)	14.67 (3.958)	18.99 (4.465)	22.65 (4.857)	9.69 (3.203)	9.85 (3.228)
V <sub>17</sub>	9.81 (3.285)	10.91 (3.449)	17.33 (4.255)	20.07 (4.563)	8.70 (3.044)	5.18 (2.479)
V <sub>18</sub>	18.00 (4.324)	15.50 (4.044)	17.00 (4.212)	21.01 (4.681)	9.73 (3.265)	8.45 (3.039)
V <sub>19</sub>	25.67 (5.153)	17.00 (4.239)	21.50 (4.743)	24.00 (4.999)	11.13 (3.453)	10.56 (3.383)
V <sub>20</sub>	37.17 (6.163)	18.33 (4.38)	23.19 (4.917)	29.11 (5.485)	11.00 (3.458)	10.20 (3.325)
V <sub>21</sub>	28.02 (5.374)	14.84 (3.93)	23.14 (4.911)	25.28 (5.122)	7.00 (2.794)	14.18 (3.776)
V <sub>22</sub>	40.33 (6.367)	18.33 (4.393)	20.67 (4.629)	25.30 (5.105)	13.67 (3.826)	19.97 (4.498)
V <sub>23</sub>	53.29 (7.354)	21.80 (4.771)	20.77 (4.626)	26.80 (5.263)	14.00 (3.828)	27.16 (5.267)
V <sub>24</sub>	45.67 (6.829)	19.57 (4.533)	18.54 (4.381)	26.87 (5.273)	13.67 (3.798)	19.58 (4.534)
V <sub>25</sub>	45.67 (6.809)	19.91 (4.562)	19.77 (4.556)	31.00 (5.656)	12.60 (3.639)	21.12 (4.703)
V <sub>26</sub>	28.96 (5.465)	17.30 (4.269)	28.44 (5.425)	31.65 (5.713)	13.67 (3.811)	11.13 (3.477)
V <sub>27</sub>	19.78 (4.476)	25.00 (5.097)	27.67 (5.345)	31.33 (5.676)	8.99 (3.141)	14.00 (3.599)
V <sub>28</sub>	33.33 (5.835)	18.20 (4.377)	21.00 (4.674)	27.47 (5.333)	14.00 (3.827)	11.59 (3.408)
V <sub>29</sub>	23.67 (4.962)	14.63 (3.952)	20.07 (4.59)	26.00 (5.194)	11.73 (3.56)	14.29 (3.885)
V <sub>30</sub>	26.84 (5.266)	13.18 (3.764)	21.75 (4.767)	26.28 (5.222)	10.67 (3.322)	6.06 (2.64)
<b>C.D.</b>	<b>0.791</b>	<b>0.632</b>	<b>0.677</b>	<b>0.496</b>	<b>0.847</b>	<b>1.07</b>
<b>SE(m)</b>	<b>0.279</b>	<b>0.223</b>	<b>0.239</b>	<b>0.175</b>	<b>0.299</b>	<b>0.377</b>
<b>C.V.</b>	<b>9.764</b>	<b>9.324</b>	<b>9.142</b>	<b>6.113</b>	<b>16.107</b>	<b>19.022</b>

The data in parenthesis indicate square root transformed values

#### **4.1.3.6 Physiological loss in weight (g)**

Noticeable variation was observed with respect to a physiological loss in weight (Table 15 and Fig. 22). *Mok.* Omayaiy Yellow (27.16 g) was observed to have minimum PLW which was on par with V<sub>25</sub> (21.12 g), V<sub>22</sub> (19.97 g) and V<sub>24</sub> (19.58 g). Whereas least was observed in *Vasco.* Aroonsri Beauty (5.14 days) was observed with the lowest value, it was on par with 26 other hybrids.

#### **4.1.4 Qualitative morphological characters**

##### **4.1.4.1 Plant characters**

Qualitative characters of the plants with respect to nature of growth, shoot characters, leaf and leaf sheath characters and root characters are presented in Tables 16 to 18.

##### **4.1.4.1.1 Nature of growth**

There are two types of nature of growth pattern in selected monopodial orchid varieties hanging and prostrate. Most of the selected *Ascocentrum* hybrids/varieties were grown in hanging baskets. V<sub>1</sub>, V<sub>2</sub>, V<sub>3</sub>, V<sub>4</sub>, V<sub>5</sub>, V<sub>6</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>9</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>14</sub>, V<sub>15</sub>, V<sub>16</sub>, V<sub>17</sub>, V<sub>18</sub>, V<sub>19</sub> and V<sub>30</sub> hybrids/varieties were included in hanging type. Whereas some tall climbing varieties were found to have a prostrate nature of growth in which included the varieties like V<sub>20</sub>, V<sub>21</sub>, V<sub>22</sub>, V<sub>23</sub>, V<sub>24</sub>, V<sub>25</sub>, V<sub>26</sub>, V<sub>27</sub>, V<sub>28</sub> and V<sub>29</sub> (Table 16 and Plate 1).

##### **4.1.4.1.2 Shoot characters**

Qualitative characters of the shoot with respect to the type of shoot, shoot colour, nature of shoot and branching of shoot are presented in Table 16.

Considerable variation was noticed among the varieties with respect to qualitative shoot characters. Medium-sized stout shoot was found in V<sub>1</sub>, V<sub>2</sub>, V<sub>4</sub>, V<sub>5</sub>, V<sub>6</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>14</sub>, V<sub>15</sub>, V<sub>16</sub> and V<sub>17</sub>. Slender shoots were observed in *Vasco.* Aroonsri Beauty and *Mok.* Calypso Pink. All other varieties were found to have sturdy shoots.

**Table 16. Qualitative shoot characters of *Ascocentrum* hybrids/varieties**

Var. No.	Nature of growth	Shoot characters		
		Nature	Colour	Branching
V <sub>1</sub>	Hanging	Medium sized	Brown	Absent
V <sub>2</sub>	Hanging	Medium sized	Brown	Absent
V <sub>3</sub>	Hanging	Sturdy	Dark brown	Absent
V <sub>4</sub>	Hanging	Medium sized	Brown	Absent
V <sub>5</sub>	Hanging	Medium sized	Brown	Absent
V <sub>6</sub>	Hanging	Medium sized	Brown	Absent
V <sub>7</sub>	Hanging	Medium sized	Brown	Absent
V <sub>8</sub>	Hanging	Medium sized	Brown	Absent
V <sub>9</sub>	Hanging	Slender	Greenish brown	Present
V <sub>10</sub>	Hanging	Medium sized	Brown	Present
V <sub>11</sub>	Hanging	Medium sized	Brown	Absent
V <sub>12</sub>	Hanging	Medium sized	Brown	Absent
V <sub>13</sub>	Hanging	Medium sized	Brown	Absent
V <sub>14</sub>	Hanging	Medium sized	Brown	Present
V <sub>15</sub>	Hanging	Medium sized	Brown	Present
V <sub>16</sub>	Hanging	Medium sized	Brown	Absent
V <sub>17</sub>	Hanging	Medium sized	Brown	Absent
V <sub>18</sub>	Hanging	Slender	Brown	Absent
V <sub>19</sub>	Prostrate	Sturdy	Dark brown	Present
V <sub>20</sub>	Prostrate	Sturdy	Dark brown	Absent
V <sub>21</sub>	Prostrate	Sturdy	Dark brown	Absent
V <sub>22</sub>	Prostrate	Sturdy	Dark brown	Present
V <sub>23</sub>	Prostrate	Sturdy	Dark brown	Present
V <sub>24</sub>	Prostrate	Sturdy	Dark brown	Present
V <sub>25</sub>	Prostrate	Sturdy	Dark brown	Present
V <sub>26</sub>	Prostrate	Sturdy	Dark brown	Absent
V <sub>27</sub>	Prostrate	Sturdy	Dark brown	Absent
V <sub>28</sub>	Prostrate	Sturdy	Dark brown	Absent
V <sub>29</sub>	Prostrate	Sturdy	Dark brown	Absent
V <sub>30</sub>	Hanging	Sturdy	Dark brown	Absent

A slight variation was observed with respect to the colour of shoots, Brown coloured shoots were observed in V<sub>1</sub>, V<sub>2</sub>, V<sub>4</sub>, V<sub>5</sub>, V<sub>6</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>14</sub>, V<sub>15</sub>, V<sub>16</sub>, V<sub>17</sub> and V<sub>18</sub>. Greenish brown coloured shoots were observed in V<sub>9</sub> (*Vasco*. Aroonsri Beauty), whereas the rest of varieties were observed to have dark brown coloured shoots. Branching of the shoots was observed in V<sub>9</sub> (*Vasco*. Aroonsri Beauty), V<sub>10</sub> (*Vasco*. Pine Rivers Fuchsia Delight), V<sub>14</sub> (*Mok*.



Rassmatozz), V<sub>15</sub> (*Mok. Khaw Phiak Suan* × *Ascda. Bicentennial Yellow Spot*), V<sub>19</sub>, V<sub>22</sub>, V<sub>23</sub>, V<sub>24</sub> and V<sub>25</sub> (Table 16).

#### 4.1.4.2 Leaf characters

Qualitative leaf characters with respect to leaf shape, leaf surface, leaf margin, leaf orientation, pigmentation, leaf base, nature of leaf sheath and colour of sheath are presented in Table 17 and also explained in Appendix XXV.

The leaf shape of variety V<sub>1</sub>, V<sub>3</sub> and V<sub>30</sub> were observed as deeply channelled towards the base. In V<sub>4</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>28</sub>, V<sub>29</sub> were found to have similar leaf shape which were semiterete leaf. The variety was V<sub>2</sub>, V<sub>13</sub>, V<sub>16</sub>, V<sub>14</sub>, V<sub>15</sub>, V<sub>22</sub> and V<sub>23</sub> were found to observe to have strap shaped leaf which was also found to be observe slightly channelled towards the base of the leaf. Whereas, V<sub>5</sub>, V<sub>6</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>12</sub>, V<sub>17</sub>, V<sub>25</sub> and V<sub>26</sub> had quarter terete shaped leaves. Liner leaves found in V<sub>18</sub> and V<sub>19</sub>. Terete leaves were observed V<sub>9</sub> was the only variety (Table 17 and Appendix XXV).

The leaf surface of V<sub>1</sub>, V<sub>2</sub>, V<sub>4</sub>, V<sub>5</sub>, V<sub>6</sub>, V<sub>7</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>15</sub>, V<sub>19</sub>, V<sub>20</sub> and V<sub>30</sub> was observed to be smooth and rigid. Varieties V<sub>3</sub>, V<sub>8</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>21</sub>, V<sub>23</sub> and V<sub>24</sub> were observed with leathery leaves, whereas in V<sub>9</sub> leaf surface was leathery and fleshy. Smooth and glabrous leaf surface was observed in the rest of the varieties.

Leaf margin of the all thirty selected *Ascocentrum* hybrids/varieties was entire (Table 17 and Appendix XXV). At the apex, V<sub>4</sub> and V<sub>9</sub> were found to have acute shape whereas emarginate leaf apex was found in the rest of the varieties. In almost all varieties colour of leaf was observed to be green except in V<sub>9</sub>, V<sub>11</sub>, V<sub>27</sub> and V<sub>30</sub> which had dark green coloured leaves. Leaf pigmentation was found in V<sub>19</sub>, V<sub>20</sub>, V<sub>21</sub>, V<sub>22</sub>, V<sub>23</sub>, V<sub>24</sub>, V<sub>25</sub>, V<sub>26</sub>, V<sub>27</sub>, V<sub>28</sub> and V<sub>29</sub>.

Three types of leaf orientation were found in the selected *Ascocentrum* hybrids/varieties straight, arching and horizontal (Table 17 and Appendix XXV). Straight or erect type of leaves were found in V<sub>1</sub>, V<sub>2</sub>, V<sub>3</sub>, V<sub>4</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>17</sub>, V<sub>18</sub>, V<sub>20</sub>, V<sub>21</sub>, V<sub>23</sub> and V<sub>26</sub>. Arching type of leaf orientation was found in V<sub>6</sub>, V<sub>9</sub>, V<sub>27</sub>,

**Table 17. Qualitative leaf characters of *Ascocentrum* hybrids/varieties**

Var. No.	Leaf shape	Leaf texture	Leaf margin	Leaf apex	Leaf colour	Leaf pigmentation	Leaf orientation	Leaf base	Nature of leaf sheath	Sheath colour
V <sub>1</sub>	Deeply channelled at base	Smooth, rigid	Entire	Emarginated	Green	Absent	Straight	Sheathed	Membranous, thick	Green
V <sub>2</sub>	Strap, channelled at base	Smooth, rigid	Entire	Emarginated	Green	Absent	Straight	Sheathed	Membranous, thick	Green
V <sub>3</sub>	Deeply channelled at base	Leathery	Entire	Emarginated	Green	Absent	Straight	Sheathed	Membranous	Green
V <sub>4</sub>	Semi terete	Smooth, rigid	Entire	Acute	Green	Absent	Straight	Sheathed	Membranous, thick	Green
V <sub>5</sub>	Quarter terete	Smooth, rigid	Entire	Emarginated	Green	Absent	Horizontal	Sheathed	Membranous, thick	Green
V <sub>6</sub>	Quarter terete	Smooth, rigid	Entire	Emarginated	Green	Absent	Arching	Sheathed	Membranous, thick	Green
V <sub>7</sub>	Semi terete	Smooth, rigid	Entire	Emarginated	Green	Absent	Horizontal	Sheathed	Membranous, thick	Green
V <sub>8</sub>	Semi terete	Leathery	Entire	Emarginated	Green	Absent	Horizontal	Sheathed	Membranous, thick	Green
V <sub>9</sub>	Terete	Fleshy, Leathery	Entire	Acute	Dark green	Absent	Arching	Sheathed	Membranous, fleshy	Green
V <sub>10</sub>	Quarter terete	Leathery	Entire	Emarginated	Green	Absent	Horizontal	Sheathed	Membranous, thick	Green
V <sub>11</sub>	Quarter terete	Leathery	Entire	Emarginated	Dark green	Absent	Horizontal	Sheathed	Membranous, thick	Dark green
V <sub>12</sub>	Quarter terete	Smooth, rigid	Entire	Emarginated	Green	Absent	Straight	Sheathed	Membranous	Green
V <sub>13</sub>	Strap, channelled at base	Smooth, rigid	Entire	Emarginated	Green	Absent	Straight	Sheathed	Membranous	Green
V <sub>14</sub>	Strap, channelled at base	Smooth glabrous	Entire	Emarginated	Green	Absent	Horizontal	Sheathed	Membranous	Green
V <sub>15</sub>	Strap, channelled at base	Smooth, rigid	Entire	Emarginated	Green	Absent	Horizontal	Sheathed	Membranous, thick	Green
V <sub>16</sub>	Strap, channelled at base	Smooth, glabrous	Entire	Emarginated	Green	Absent	Horizontal	Sheathed	Membranous, thick	Green
V <sub>17</sub>	Quarter terete	Smooth, glabrous	Entire	Emarginated	Green	Absent	Straight	Sheathed	Membranous, thick	Green
V <sub>18</sub>	Liner, quarter terete	Smooth, glabrous	Entire	Emarginated	Green	Absent	Straight	Sheathed	Membranous	Dark green

**Table 17. continued**

Var. No.	Leaf shape	Leaf texture	Leaf margin	Leaf apex	Leaf colour	Leaf pigmentation	Leaf orientation	Leaf base	Nature of leaf sheath	Sheath colour
V <sub>19</sub>	Liner, channelled at tip	Smooth, rigid	Entire	Emarginated	Green	Present	Horizontal	Sheathed	Membranous, thick	Green
V <sub>20</sub>	Strap	Smooth, rigid	Entire	Emarginated	Green	Present	Straight	Sheathed	Membranous, thick	Green
V <sub>21</sub>	Strap	Leathery	Entire	Emarginated	Green	Present	Straight	Sheathed	Membranous, thick	Green
V <sub>22</sub>	Strap, channelled at base	Smooth, glabrous	Entire	Emarginated	Green	Present	Horizontal	Sheathed	Membranous, thick	Green
V <sub>23</sub>	Strap, channelled at base	Leathery	Entire	Emarginated	Green	Present	Straight	Sheathed	Membranous, thick	Green
V <sub>24</sub>	Strap	Leathery	Entire	Emarginated	Green	Present	Horizontal	Sheathed	Membranous, thick	Green
V <sub>25</sub>	Quarter terete	Smooth, glabrous	Entire	Emarginated	Green	Present	Horizontal	Sheathed	Membranous, thick	Green
V <sub>26</sub>	Quarter terete	Smooth, glabrous	Entire	Emarginated	Green	Present	Straight	Sheathed	Membranous, thick	Green
V <sub>27</sub>	Strap	Smooth, glabrous	Entire	Emarginated	Dark green	Present	Arching	Sheathed	Membranous, thick	Dark green
V <sub>28</sub>	Semi terete	Smooth, glabrous	Entire	Emarginated	Green	Present	Arching	Sheathed	Membranous, thick	Green
V <sub>29</sub>	Semi terete	Smooth, glabrous	Entire	Emarginated	Green	Present	Horizontal	Sheathed	Membranous, thick	Green
V <sub>30</sub>	Deeply channelled at base	Smooth, rigid	Entire	Emarginated	Dark green	Absent	Horizontal	Sheathed	Membranous, thick	Dark green

and V<sub>28</sub>. However, the rest of the varieties had horizontally orientated leaves. The leaf base was observed to be sheathed in all the varieties. The membranous nature of sheath was observed in V<sub>12</sub>, V<sub>13</sub>, V<sub>14</sub> and V<sub>18</sub> wherein, it was membranous and thick in rest of the varieties. Green sheath colour was shown in all varieties except in V<sub>11</sub>, V<sub>18</sub>, V<sub>27</sub> and V<sub>30</sub> which had the dark green colour of sheaths.

**Table 18. Qualitative root characters of *Ascocentrum* hybrids/ varieties**

<b>Var. No.</b>	<b>Origin of roots</b>	<b>Branching of roots</b>	<b>Nature of roots</b>	<b>Root colour</b>
V <sub>1</sub>	Basal	Present	Cylindrical	Greyish brown
V <sub>2</sub>	Basal	Present	Cylindrical	Greenish grey
V <sub>3</sub>	Basal	Present	Cylindrical	Greenish grey
V <sub>4</sub>	Basal	Present	Cylindrical	Greenish grey
V <sub>5</sub>	Basal	Present	Cylindrical	Greenish grey
V <sub>6</sub>	Along the stem	Present	Cylindrical	Greenish grey
V <sub>7</sub>	Basal	Present	Cylindrical	Greyish brown
V <sub>8</sub>	Along the stem	Present	Cylindrical	Greenish grey
V <sub>9</sub>	Along the stem	Present	Cylindrical	Greenish grey
V <sub>10</sub>	Along the stem	Present	Cylindrical	Greenish grey
V <sub>11</sub>	Basal	Present	Cylindrical	Greenish grey
V <sub>12</sub>	Basal	Absent	Cylindrical	Greenish grey
V <sub>13</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>14</sub>	Basal	Present	Cylindrical	Greenish grey
V <sub>15</sub>	Along the stem	Present	Cylindrical	Greyish brown
V <sub>16</sub>	Along the stem	Present	Cylindrical	Greyish brown
V <sub>17</sub>	Basal, Along the stem	Present	Cylindrical	Greyish brown
V <sub>18</sub>	Along the stem	Present	Cylindrical	Greenish grey
V <sub>19</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>20</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>21</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>22</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>23</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>24</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>25</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>26</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>27</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>28</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>29</sub>	Along the stem	Absent	Cylindrical	Greenish grey
V <sub>30</sub>	Along the stem	Absent	Cylindrical	Greenish grey

#### **4.1.4.3 Root characters**

Detailed qualitative root characters with respect to root origin, root branching, root nature and root colour are presented in Table 18.

Slight variation was found among the varieties with respect to qualitative root characters. Root origin towards basal region was found in V<sub>1</sub>, V<sub>2</sub>, V<sub>3</sub>, V<sub>4</sub>, V<sub>5</sub>, V<sub>7</sub>, V<sub>11</sub>, V<sub>12</sub> and V<sub>14</sub>. However, in the rest of the varieties, root origin was found along the stem. Root branching was observed in V<sub>1</sub>, V<sub>2</sub>, V<sub>3</sub>, V<sub>4</sub>, V<sub>5</sub>, V<sub>6</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>9</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>14</sub>, V<sub>15</sub>, V<sub>16</sub>, V<sub>17</sub> and V<sub>18</sub>. Cylindrical roots were observed in the entire selected thirty varieties. Greyish brown coloured roots were observed in V<sub>1</sub>, V<sub>7</sub>, V<sub>15</sub>, V<sub>16</sub> and V<sub>17</sub> whereas the greenish-grey coloured fresh and healthy roots were observed in rest of the hybrids/varieties.

#### **4.1.4.4 Floral characters**

The qualitative details about the floral characters with respect to their blooming period/flowering seasons, spike characters, floret characters, petal characters, labellum/lip characters, column characters and spur characters are presented in Tables 19 to 24 and also explained in Appendix XXV.

##### **4.1.4.4.1 Spike characters**

Variations were observed with respect to flowering and spike characters of *Ascocentrum* hybrids/varieties (Table 19 and Plate 2-7). Detailed observations were recorded on the number of spike per plant at a time, spike orientation, colour of spike, colour of inflorescence and orientation of florets on spike.

Five blooming period were observed in throughout the period of study in V<sub>25</sub> (Nov.-Dec., Feb.-Mar., Apr.-May., Jun.-Jul. and Aug.-Sept.), whereas four were noticed in V<sub>22</sub> (Jun.-Jul., Nov.-Dec., Apr.-May and Aug. -Sept) and V<sub>27</sub> (Nov. Dec., Jan.-Feb., Apr.-May. and Jun.-Jul.).

Three blooming period were noticed V<sub>1</sub> (May-Jun., Apr.-May and Nov.-Dec). V<sub>2</sub> (Mar.-Apr., Sep.-Oct. and Jun.-Jul), V<sub>6</sub> (Mar.-Apr., May-Jun. and Aug.-

**Table 19. Qualitative flowering/spike characters of *Ascocentrum* hybrids/ varieties**

Var. No.	Blooming period/flowering season	Spikes per plant at a time	Spike orientation	Nature of inflorescence	Spike colour	Colour of inflorescence	Orientation of flowers on the spike
V <sub>1</sub>	May-Jun., Apr.-May, Nov.-Dec.	Single	Erect	Dense	Green	Strong orange	Facing in all directions
V <sub>2</sub>	Mar.-Apr., Sep.-Oct., Jun.-Jul.	Single	Arching	Lax	Green	White and greyish yellow shaded with maroon dots	Facing in all directions
V <sub>3</sub>	Apr.-May, Jun.-Jul.	Double	Arching	Lax	Green	Whitish violet, violet shaded, blue dotted	Facing in all directions
V <sub>4</sub>	Apr.-Mar., Jan.-Feb.	Single	Arching	Lax	Green	Reddish orange, scarlet	Facing in all directions
V <sub>5</sub>	Feb. - Apr., Aug.-Oct.	Single	Arching	Lax	Green	Pale yellow	Facing in all directions
V <sub>6</sub>	Mar.- Apr., May-Jun., Aug.- Sep.	Triple	Erect	Dense	Green	Orangish yellow, dotted	Facing in all directions
V <sub>7</sub>	Mar.-Apr. May. -Jun.	Double	Erect	Lax	Green	Purplish blue	Facing in all directions
V <sub>8</sub>	Jun.-Jul.	Double	Erect	Lax	Green	Fresh pink	Facing in all directions
V <sub>9</sub>	Mar.-Apr.	Double	Erect	Lax	Green, brown streaked	Dark pink	Facing in all directions
V <sub>10</sub>	Jun.-Jul., Nov. Dec.	Triple	Erect	Lax	Green	Purplish pink	Facing in all directions
V <sub>11</sub>	May- Jun.	Double	Arching	Dense	Green	White	Facing in all directions
V <sub>12</sub>	Apr.-May, Jun.-Jul.	Single	Erect	Lax	Green	Pinkish white shades, dark pink dotted	Facing in all directions
V <sub>13</sub>	Jun.-Jul., Mar.-Apr., Nov.-Dec.	Single	Arching	Lax	Green	Dark purple, netted	Facing in all directions
V <sub>14</sub>	Jan.-Feb., Mar.-Apr.	Single	Erect	Lax	Green	Orangish cream yellow, dotted	Facing in all directions
V <sub>15</sub>	May.-Jun., Jul.-Sept.	Single	Deflexed	Lax	Green	Sulfur yellow big dotted	Facing in all directions

**Table 19. continued**

Var. No.	Blooming period/flowering season	Spikes per plant at a time	Spike orientation	Nature of inflorescence	Spike colour	Colour of inflorescence	Orientation of flowers on a spike
V <sub>16</sub>	Mar.-Apr., May-Jun.	Single	Erect	Lax	Green	Sulfur yellow, minute dotted	Facing in all directions
V <sub>17</sub>	Jan.-Feb., Mar.-Apr., Nov.-Dec.	Single	Erect	Lax	Green	Yellow	Facing in all directions
V <sub>18</sub>	Mar.-Apr.	Single	Erect	Lax	Green	Dark pink	Facing in all directions
V <sub>19</sub>	Jun.-Jul., Oct.-Nov.	Double	Erect	Lax	Green	Deep dark pink	Facing in all direction
V <sub>20</sub>	Nov.-Dec., Mar.-Apr. Jun.-Jul.	Single	Erect	Lax	Green	Sulfur yellow, small dotted	Facing in all direction
V <sub>21</sub>	Sept. Oct.	Single	Erect	Lax	Green	Yellowish orange, minute dotted	Facing in all direction
V <sub>22</sub>	Jun.-Jul., Nov.-Dec., Apr.-May. Aug.-Sept.	Triple	Erect	Lax	Green	Orangish yellow, big dotted	Facing in all direction
V <sub>23</sub>	Mar.-Apr., May-Jun, Jul.-Aug.	Multiple	Erect	Dense	Green	Pinkish yellow, minute rarely dotted	Facing in all direction
V <sub>24</sub>	Mar.-Apr., May-Jun., Jul.-Aug.	Triple	Erect	Lax	Green	Yellowish orange, rarely, minute dotted	Facing in all direction
V <sub>25</sub>	Nov.-Dec., Feb.-Mar., Apr.-May., Jun.-Jul., Aug.-Sept.	Multiple	Erect	Dense	Green	Yellowish with small dark red spots	Facing in all direction
V <sub>26</sub>	May-Jun., Jul.-Aug.	Single	Erect	Lax	Green	Pinkish cream saded with dark maroon spots	Facing in all direction
V <sub>27</sub>	Nov. Dec., Jan.-Feb., Apr.-May., Jun.-Jul.	Double	Erect	Dense	Green	Dark brownish orange, big dotted	Facing in all direction
V <sub>28</sub>	Jun.-Jul.	Double	Erect	Lax	Green	Dark red, rarely spotted	Facing in all direction
V <sub>29</sub>	Apr.-May, Jun.-Aug.	Single	Erect	Lax	Green	Red, small dotted	Facing in all direction
V <sub>30</sub>	May-Jun, Jul.-Aug.	Double, branched	Erect	Dense	Green	Pleasant dark red	Facing in all direction

Sep), V<sub>13</sub> (Jun.-Jul., Mar.-Apr. and Nov.-Dec.), V<sub>17</sub> (Jan.-Feb., Mar.-Apr. and Nov.-Dec.), V<sub>20</sub> (Nov.-Dec., Mar.-Apr. and Jun.-Jul.), V<sub>24</sub> and V<sub>23</sub> (Mar.-Apr., May-Jun. and Jul.-Aug. in each).

Two blooming period were noticed in V<sub>4</sub> (Apr.-Mar. and Jan.-Feb.), V<sub>5</sub> (Feb.-Apr. and Aug.-Oct.), V<sub>10</sub> (Jun.-Jul. and Nov. Dec.) V<sub>14</sub>, (Jan.-Feb. and Mar.-Apr.) V<sub>15</sub> (May.-Jun. and Jul.-Sept.), V<sub>19</sub> (Jun.-Jul. and Oct.-Nov.), V<sub>29</sub> (Apr.-May and Jun.-Aug), V<sub>3</sub>, V<sub>12</sub> (Apr.-May and Jun.-Jul. in each), V<sub>7</sub>, V<sub>16</sub> (Mar.-Apr. and May. -Jun. in each) and V<sub>26</sub>, V<sub>30</sub> (May-Jun and Jul.-Aug. in each), whereas only a single flowering season throughout the period of study was noticed in V<sub>9</sub> (Mar.-Apr.), V<sub>11</sub> (May- Jun.), V<sub>18</sub> (Mar.-Apr.), V<sub>21</sub> (Sept. Oct.), V<sub>8</sub> and V<sub>28</sub> (Jun.-Jul.in each) (Table 19 and Plate 2-7).

The multiple numbers of spikes per plant at a time (Plate 2-7) were observed in V<sub>23</sub> and V<sub>25</sub>. Whereas three spikes per plant at a time were observed in V<sub>6</sub>, V<sub>10</sub>, V<sub>22</sub> and V<sub>24</sub>. Two spikes per plant at a time were noted in V<sub>3</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>9</sub>, V<sub>11</sub>, V<sub>19</sub>, V<sub>27</sub> and V<sub>28</sub>. Whereas rest of the varieties had only single spike per plant at a time.

Spike orientation was found deflexed in V<sub>15</sub> and arching in V<sub>2</sub>, V<sub>3</sub>, V<sub>4</sub>, V<sub>5</sub>, V<sub>11</sub> and V<sub>13</sub>. Whereas in rest of the varieties flowering spike was found straight/erect oriented. Nature of inflorescences was found dense in V<sub>1</sub>, V<sub>7</sub>, V<sub>11</sub>, V<sub>23</sub>, V<sub>27</sub> and V<sub>30</sub> whereas lax nature of inflorescences found in rest of the varieties. Green with the brown-streaked colour of spike was observed in V<sub>9</sub> (*Vasco*. Aroonsri Beauty) whereas in the rest of the varieties had green coloured spikes (Appendix XXV and Plate 3-7).

The predominant colour of the inflorescence was observed and recorded in the selected thirty intergeneric hybrids/varieties of *Ascocentrum* (Plate 2 to 7). In V<sub>1</sub> strong orange coloured inflorescences were observed. In V<sub>2</sub> white and greyish-yellow shaded inflorescences were observed. In V<sub>3</sub> inflorescence was whitish violet, violet shaded, blue dotted.



Inflorescences colours in other varieties *viz.*, V<sub>4</sub> (reddish orange/scarlet), V<sub>5</sub> (pale yellow), V<sub>6</sub> (orangish yellow, dotted), V<sub>7</sub> (purplish blue), V<sub>8</sub> (fresh pink), V<sub>9</sub> (dark pink), V<sub>10</sub> (purplish pink), V<sub>11</sub> (white), V<sub>12</sub> (pinkish white shades, dark pink dotted), V<sub>13</sub> (dark purple, netted), V<sub>14</sub> (orangish cream yellow, dotted), V<sub>15</sub> (sulfur yellow big dotted), V<sub>16</sub> (sulfur yellow, minute dotted), V<sub>17</sub> (yellow), V<sub>18</sub> (dark pink), V<sub>19</sub> (deep dark pink) V<sub>20</sub> (sulfur yellow, small dotted) V<sub>21</sub> (yellowish orange, minute dotted), V<sub>22</sub> (orangish yellow, big dotted), V<sub>23</sub> (pinkish yellow, minute rarely dotted), V<sub>24</sub> (yellowish orange, rarely, minutely dotted), V<sub>25</sub> (yellowish with small dark red spots), V<sub>26</sub> (pinkish cream shaded with dark maroon spots), V<sub>27</sub> (dark brownish orange, big dotted), V<sub>28</sub> (dark red, rarely spotted), V<sub>29</sub> (red, small dotted) and V<sub>30</sub> (pleasant dark red) was also recorded (Plate 3 to 7).

Much variation was not exhibited with respect to the orientation of florets on a spike. The orientation of flowers on a spike was observed to be facing in all directions in all selected thirty varieties.

#### **4.1.4.4.2 Floret characters**

The selected intergeneric hybrid varieties varied in all floret aspects and wide range of variation could be observed with respect to the colour of florets, flower fragrance and pigmentation. The details regarding qualitative floral characters are presented in Table 20, depicted in Plate 8 and also explained in Appendix XXV.

Colour of single floret was observed by RHS colour chart and recorded in all selected hybrids/varieties (Plate 3-7). Variation was found with respect to flower colour in the selected varieties which differed in each variety. V<sub>1</sub> (orange group 25, strong orange A), V<sub>2</sub> (grayed-yellow 160, moderate yellow A and White group N155, pinkish white B), V<sub>3</sub> (violet-blue group 94, Brilliant purplish blue C), V<sub>4</sub> (grayed-orange group 169, strong reddish orange A), V<sub>5</sub> (yellow group 13, vivid yellow A and B), V<sub>6</sub> (orange group 28, vivid yellowish pink A), V<sub>7</sub> (violet-blue group N87, brilliant purple C), V<sub>8</sub> (red purple group N57, deep purplish pink

**Table 20. Qualitative floret characters of *Asocentrum* hybrids/ varieties**

<b>Var. No.</b>	<b>Colour of florets</b>	<b>Pigmentation</b>	<b>Flower fragrance</b>
V <sub>1</sub>	Orange group 25, strong orange A	Absent	Present
V <sub>2</sub>	Grayed-yellow 160, moderate yellow A & White group N155, pinkish white B	Absent	Present
V <sub>3</sub>	Violet-blue group 94, brilliant purplish blue C	Absent	Present
V <sub>4</sub>	Grayed-orange group 169, strong reddish orange A	Absent	Present
V <sub>5</sub>	Yellow group 13. vivid yellow A & B	Absent	Present
V <sub>6</sub>	Orange group 28, vivid yellowish pink A	Absent	Present
V <sub>7</sub>	Violet-blue group N87, Brilliant purple C	Absent	Present
V <sub>8</sub>	Red purple group N57, deep purplish pink C & Red-purple group 61, Deep purplish pink D	Absent	Present
V <sub>9</sub>	Red-purple group N74, vivid reddish purple B	Absent	Present
V <sub>10</sub>	Purple group NN78, light reddish purple D	Absent	Present
V <sub>11</sub>	White group NN155, white D	Present	Present
V <sub>12</sub>	Purple group N78, strong reddish purple B & Red purple group N74, moderate purplish pink D	Present	Absent
V <sub>13</sub>	Purple group N78, strong reddish purple B & Pink group NN74, strong reddish purple A	Present	Absent
V <sub>14</sub>	Orange-red group 32, vivid reddish orange A & Yellow -orange group 20, light yellow B	Present	Absent
V <sub>15</sub>	Yellow group 9. brilliant yellow C & Grayed-red group 179, moderate red A	Absent	Present
V <sub>16</sub>	Yellow group 6, brilliant greenish yellow C	Absent	Present
V <sub>17</sub>	Yellow orange group 17, light yellow D	Absent	Present

**Table 20. continued**

<b>Var. No.</b>	<b>Colour of florets</b>	<b>Pigmentation</b>	<b>Flower fragrance</b>
V <sub>18</sub>	Purple group NN78, light reddish purple D	Absent	Present
V <sub>19</sub>	Red purple group N74, vivid reddish purple A	Present	Absent
V <sub>20</sub>	Yellow orange group 14, vivid yellow B	Present	Absent
V <sub>21</sub>	Greyed-orange group N163, strong orange yellow C	Present	Absent
V <sub>22</sub>	Orange group 25, strong orange B & Yellow orange group 22, strong orange yellow A	Present	Absent
V <sub>23</sub>	Yellow orange group 22, strong orange yellow A & Orange group 26, light yellowish pink D	Present	Absent
V <sub>24</sub>	Yellow-orange group 22, strong orange yellow A & Yellow orange group 14, vivid yellow B	Present	Absent
V <sub>25</sub>	Yellow-orange group 20, brilliant yellow A	Present	Absent
V <sub>26</sub>	Red-purple group N74, moderate purplish pink D & Red-purple group 73, Deep purplish pink A	Present	Absent
V <sub>27</sub>	Red-group 44, vivid reddish orange B	Present	Absent
V <sub>28</sub>	Red purple group 58, strong purplish red B & Red group 53, strong red C	Present	Absent
V <sub>29</sub>	Red purple group 58, strong purplish red B & Red group 53, strong red D	Present	Absent
V <sub>30</sub>	Red group N45, moderate red A	Absent	Present

C and red-purple group 61, deep purplish pink D), V<sub>9</sub> (red-purple group N74, vivid reddish purple B), V<sub>10</sub> (purple group NN78, light reddish purple D), V<sub>11</sub> (white group NN155, white D), V<sub>12</sub> (purple group N78, strong reddish purple B and red purple group N74, moderate purplish pink D), V<sub>13</sub> (purple group N78, strong reddish purple B and pink group NN74, strong reddish purple A), V<sub>14</sub> (orange-red group 32, vivid reddish orange A and yellow -orange group 20, light yellow B), V<sub>15</sub> (yellow group 9. brilliant yellow C and grayed-red group 179, moderate red A), V<sub>16</sub> (yellow group 6, brilliant greenish yellow C), V<sub>17</sub> (yellow orange group 17, light yellow D), V<sub>18</sub> (purple group NN78, light reddish purple D), V<sub>19</sub> (red purple group N74, vivid reddish purple A), V<sub>20</sub> (yellow orange group 14, vivid yellow B), V<sub>21</sub> (greyed-orange group N163, strong orange yellow C), V<sub>22</sub> (orange group 25, strong orange B and yellow orange group 22, strong orange yellow A), V<sub>23</sub> (Yellow orange group 22, Strong orange yellow A and Orange group 26, light yellowish pink D), V<sub>24</sub> (yellow-orange group 22, strong orange yellow A and yellow orange group 14, vivid yellow B), V<sub>25</sub> (yellow-orange group 20, brilliant yellow A), V<sub>26</sub> (red-purple group N74, moderate purplish pink D and red-purple group 73, Deep purplish pink A), V<sub>27</sub> (red-group 44, vivid reddish orange B), V<sub>28</sub> (red purple group 58, strong purplish red B and red group 53, strong red C), V<sub>29</sub> (red purple group 58, strong purplish red B and Red group 53, strong red D) and V<sub>30</sub> (red group N45, moderate red A).

Flower pigmentation (the colour changes during maturity) was observed in V<sub>11</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>14</sub>, V<sub>19</sub>, V<sub>20</sub>, V<sub>21</sub>, V<sub>22</sub>, V<sub>23</sub>, V<sub>24</sub>, V<sub>25</sub>, V<sub>26</sub>, V<sub>27</sub>, V<sub>28</sub> and V<sub>29</sub>, and was not found in rest of the varieties and also observed that wherever flower pigmentation present and fragrance absent, wherever pigmentation absent and flower fragrance present. Floral fragrance was felt in V<sub>1</sub>, V<sub>2</sub>, V<sub>3</sub>, V<sub>4</sub>, V<sub>5</sub>, V<sub>6</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>9</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>15</sub>, V<sub>16</sub>, V<sub>17</sub>, V<sub>18</sub> and V<sub>30</sub> out of selected varieties (Table 20).

#### **4.1.4.4.3 Petal characters**

The qualitative petal characters of the varieties varied in all aspects and a wide range of variation was observed. The detail regarding petal shape, petal curvature, petal apex, petal margin, petal base colour, margin colour petal apex

**Table 21. Qualitative petal characters of *Ascocentrum* hybrids/varieties**

Var. No.	Petal shape	Petal curvature	Petal apex	Petal margin	Petal base colour	Petal margin colour
V <sub>1</sub>	Obovate	Incurved with straight apex	Acute	Entire	Single (Bright orange)	Single (Bright orange)
V <sub>2</sub>	Orbicular	Straight	Truncate	Undulate	Triple (White, yellow, maroon dotted)	Triple (White, yellow, maroon)
V <sub>3</sub>	Obovate	Deflexed with straight apex	Truncate	Undulate	Double (White, blue dotted)	Triple (White, violet, blue dotted)
V <sub>4</sub>	Obovate	Deflexed with straight apex	Obtuse	Undulate	Double (Yellowish orange shaded)	Single (Orangish red, scarlet)
V <sub>5</sub>	Orbicular	Incurved with incurved apex	Obtuse	Entire	Single (Pale yellow)	Single (Pale yellow)
V <sub>6</sub>	Obovate	Deflexed with incurved apex	Obtuse	Entire	Double (Orangish yellow)	Double (Orangish yellow)
V <sub>7</sub>	Obovate	Straight	Acute	Entire	Single (Purplish blue)	Single (Purplish blue)
V <sub>8</sub>	Obovate	Straight	Acute	Entire	Single (Pink)	Single (Pink)
V <sub>9</sub>	Lanceolate	Deflexed with straight apex	Acute	Entire	Single (Dark pink)	Single (Dark pink)
V <sub>10</sub>	Obovate	Straight	Acute	Entire	Single (Purplish pink)	Single (Purplish pink)
V <sub>11</sub>	Obovate	Deflexed with incurved apex	Acute	Entire	Single (White)	Single (White)
V <sub>12</sub>	Obovate	Deflexed with incurved apex	Acute	Entire	Double (pinkish white, spotted)	Double (White group, pinkish white, Shaded)
V <sub>13</sub>	Obovate	Deflexed with deflexed apex	Obtuse	Erose	Double (Spotted)	Single (pink)
V <sub>14</sub>	Orbicular	Straight	Obtuse	Entire	Double (Spotted)	Single (Orangish yellow)
V <sub>15</sub>	Orbicular	Straight	Obtuse	Entire	Double (Yellow, brown, spotted)	Single (Brown)

**Table 21. continued**

<b>Var. No.</b>	<b>Petal shape</b>	<b>Petal curvature</b>	<b>Petal apex</b>	<b>Petal margin</b>	<b>Petal base colour</b>	<b>Petal margin colour</b>
V <sub>16</sub>	Obovate	Deflexed with straight apex	Obtuse	Entire	Double (Yellow, brown, spotted)	Double (yellow)
V <sub>17</sub>	Obovate	Deflexed with straight apex	Acute	Entire	Single (Yellow)	Single (Yellow)
V <sub>18</sub>	Obovate	Deflexed with straight apex	Obtuse	Entire	Single (Pink)	Single (Pink)
V <sub>19</sub>	Orbicular	Deflexed with incurved apex	Obtuse	Undulate	Single (Dark pink)	Single (Dark pink)
V <sub>20</sub>	Orbicular	Deflexed with incurved apex	Obtuse	Entire	Double (Yellow, brown blotched)	Single (Yellow, with brown small dots)
V <sub>21</sub>	Orbicular	Deflexed with incurved apex	Obtuse	Entire	Single (Orangish yellow, with red minute small dots)	Single (Orangish yellow)
V <sub>22</sub>	Obovate	Deflexed with straight apex	Obtuse	Undulate	Double (Yellow, brown blotched)	Single (Orangish yellow )
V <sub>23</sub>	Obovate	Deflexed with deflexed apex	Obtuse	Undulate	Single (Pinkish white)	Double (Pinkish and yellowish)
V <sub>24</sub>	Obovate	Deflexed with deflexed apex	Obtuse	Undulate	Single (Orangish yellow)	Single (Yellowish, orange)
V <sub>25</sub>	Obovate	Deflexed with straight apex	Obtuse	Entire	Single (Orangish yellow)	Single (Yellowish, orange spotted)
V <sub>26</sub>	Obovate	Deflexed with straight apex	Obtuse	Entire	Double (Pinkish white with dark pink dots)	Double (Pinkish white, dark pink shades )
V <sub>27</sub>	Oblong	Deflexed with incurved apex	Obtuse	Undulate	Single (red blotched, spotted)	Single (red blotched, spotted)
V <sub>28</sub>	Obovate	Deflexed with straight apex	Acute	Entire	Single (red spotted)	Single (red spotted)
V <sub>29</sub>	Lanceolate	Deflexed with straight apex	Acute	Entire	Single (red spotted)	Single (red spotted)
V <sub>30</sub>	Orbicular	Incurved with incurved apex	Obtuse	Entire	Single (Red)	Single (Red)

**Table 22. Qualitative petal characters of *Ascocentrum* hybrids/varieties**

<b>Var. No.</b>	<b>Petal apex colour</b>	<b>Petal colour pattern</b>	<b>Remarks</b>
V <sub>1</sub>	Single (Bright orange)	Streaked	Lateral sepals comparatively bigger than petals
V <sub>2</sub>	Triple (White, yellow, maroon)	Spotted, Shaded	Lateral sepals colour is different
V <sub>3</sub>	Triple (White, violet, blue dotted)	Spotted, tessellated	Lateral sepals are tessellated
V <sub>4</sub>	Single (Orangish red, scarlet)	Uniform	Lateral sepals comparatively bigger than petals
V <sub>5</sub>	Single (Pale yellow)	Uniform, shaded at the base	Lateral sepals comparatively bigger than petals
V <sub>6</sub>	Double (Orangish yellow)	Spotted	Lateral sepals comparatively bigger than petals
V <sub>7</sub>	Single (Purplish blue)	Streaked	Lateral sepals comparatively bigger than petals
V <sub>8</sub>	Single (Pink)	Streaked	Lateral sepals comparatively bigger than petals
V <sub>9</sub>	Single (Dark pink)	Uniform	Lateral sepals comparatively bigger than petals
V <sub>10</sub>	Single (Purplish pink)	Streaked	Lateral sepals comparatively bigger than petals
V <sub>11</sub>	Single (White)	Uniform	Lateral sepals comparatively bigger than petals
V <sub>12</sub>	Single (Deep purplish pink, shaded)	Spotted, Shaded	Lateral sepals comparatively bigger than petals
V <sub>13</sub>	Single (pink)	Spotted, tessellated	Lateral sepals comparatively bigger than petals
V <sub>14</sub>	Single (Orangish, yellow)	Spotted	Lateral sepals comparatively bigger than petals
V <sub>15</sub>	Single (Brown)	Spotted	Lateral sepals comparatively bigger than petals
V <sub>16</sub>	Single (pale yellow)	Spotted, shaded at the apex	Lateral sepals comparatively bigger than petals

**Table 22. continued**

<b>Var. No.</b>	<b>Petal apex colour</b>	<b>Petal colour pattern</b>	<b>Remarks</b>
V <sub>17</sub>	Single (Dark yellow)	Uniform	Lateral sepals comparatively bigger than petals
V <sub>18</sub>	Single (Pink)	Uniform, shaded at the apex	Lateral sepals comparatively bigger than petals
V <sub>19</sub>	Single (Dark pink)	Tessellated	Lateral sepals comparatively bigger than petals
V <sub>20</sub>	Double (Yellow, with brown small dots)	Spotted	Lateral sepals comparatively bigger than petals
V <sub>21</sub>	Single (Orangish yellow)	Spotted	Lateral sepals comparatively bigger than petals
V <sub>22</sub>	Single (Orange dotted)	Spotted, shaded	Lateral sepals comparatively bigger than petals
V <sub>23</sub>	Double (Pinkish and yellowish)	Uniform, minutely spotted, tessellated	Lateral sepals comparatively bigger than petals and tessellated
V <sub>24</sub>	Single (Yellowish orange)	Uniform, minutely spotted, tessellated	Lateral sepals comparatively bigger than petals and tessellated
V <sub>25</sub>	Single (Yellowish, orange spotted)	Spotted	Lateral sepals comparatively bigger than petals
V <sub>26</sub>	Double (Pinkish white with dark pink dots)	Spotted, shaded	Lateral sepals comparatively bigger than petals
V <sub>27</sub>	Single (red blotched, spotted)	Blotched, spotted	Lateral sepals comparatively bigger than petals
V <sub>28</sub>	Single (red spotted)	Spotted	Lateral sepals comparatively bigger than petals
V <sub>29</sub>	Single (red spotted)	Spotted	Lateral sepals comparatively bigger than petals
V <sub>30</sub>	Single (Red)	Uniform	Lateral sepals comparatively bigger than petals



colour and petal colour pattern. The details on qualitative petal characters are presented in Tables 21 and 22 also explained in Appendix XXV.

Orbicular shaped petals were observed in V<sub>2</sub>, V<sub>5</sub>, V<sub>14</sub>, V<sub>15</sub>, V<sub>19</sub>, V<sub>20</sub>, V<sub>21</sub> and V<sub>30</sub>, while lanceolate type of petals were observed in V<sub>9</sub> and V<sub>29</sub>, whereas in all other varieties, obovate petals were observed. A strong variation was found with respect to petal curvature. V<sub>1</sub> was found to have incurved with straight apex petal curvature, V<sub>5</sub> and V<sub>30</sub> were found to have incurved with incurved apex petal curvature. In V<sub>6</sub>, V<sub>11</sub>, V<sub>12</sub>, V<sub>19</sub>, V<sub>20</sub>, V<sub>21</sub> and V<sub>27</sub> petal curvature was deflexed with incurved apex. V<sub>13</sub>, V<sub>23</sub> and V<sub>24</sub> were found to have petals with deflexed with deflexed apex. In rest of the varieties, petal curvature was found to be deflexed with straight apex, whereas V<sub>2</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>10</sub>, V<sub>14</sub> and V<sub>15</sub> were observed to have straight petals without any curvature (Table 21 and Appendix XXV).

There was slight variation with respect to petal apex. In V<sub>1</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>9</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>12</sub>, V<sub>17</sub>, V<sub>28</sub> and V<sub>29</sub> petal apex was found to be acute. Petal apex in V<sub>2</sub> and V<sub>3</sub> was found to be truncate. However, in rest of the varieties petal apex was found to be obtuse (Table 21 and Appendix XXV).

Undulated petal margin was found in V<sub>2</sub>, V<sub>3</sub>, V<sub>4</sub>, V<sub>19</sub>, V<sub>22</sub>, V<sub>23</sub>, V<sub>24</sub> and V<sub>27</sub>. V<sub>13</sub> was found to have erose margin. However, in rest of the varieties petal margin was found to be entire (Table 21 and Appendix XXV).

Abundant variation was found with respect to petal base, margin and apex colour characters. Three colours at the petal base were observed in V<sub>2</sub>. Double colour at the base of the petal was found in V<sub>3</sub>, V<sub>4</sub>, V<sub>6</sub>, V<sub>12</sub>, V<sub>13</sub>, V<sub>14</sub>, V<sub>15</sub>, V<sub>16</sub>, V<sub>20</sub>, V<sub>22</sub> and V<sub>26</sub>. Whereas in rest of other varieties, single colour was found at the base of petals (Table 21, 22 and Appendix XXV).

A wide range of variation was observed in the colour pattern of petals. In V<sub>3</sub> and V<sub>13</sub> spotted and the tessellated colour pattern was found to observe V<sub>19</sub> was tessellated and V<sub>5</sub> was uniform, shaded at the base, V<sub>18</sub> was uniform, shaded at the

**Table 23. Qualitative lip characters of *Ascoctrinum* hybrids/varieties**

<b>Var. No.</b>	<b>Lip shape at apical lobe region</b>	<b>Nature of lateral lip</b>	<b>Lip shape at lateral lobe region</b>	<b>Nature of lateral lip</b>	<b>Lip apex</b>	<b>Lip surface</b>
V <sub>1</sub>	Elliptic	Deflexed with deflexed apex	Obtriangular	Incurved	Truncate	Leathery, rigid
V <sub>2</sub>	Ovate	Deflexed with straight apex	Suborbicular	Strongly incurved	Bilobed	Leathery, rigid
V <sub>3</sub>	Elliptic	Deflexed with straight apex	Obtriangular	Incurved	Obtuse	Leathery, rigid
V <sub>4</sub>	Ovate	Deflexed with deflexed apex	Suborbicular	Incurved	Bilobed	Leathery, rigid
V <sub>5</sub>	Ovate	Deflexed with deflexed apex	Suborbicular	Incurved	Bilobed	Leathery, rigid
V <sub>6</sub>	Ovate	Deflexed with deflexed apex	Obtriangular	Incurved	Bilobed	Leathery, rigid
V <sub>7</sub>	Lanceolate	Straight	Ob lanceolate	Incurved	Bilobed	Leathery, rigid
V <sub>8</sub>	Lanceolate	Straight	Ob lanceolate	Incurved	Bilobed	Leathery, rigid
V <sub>9</sub>	Oblong	Deflexed with deflexed apex	Semi circular	Incurved	Obtuse	Leathery, rigid
V <sub>10</sub>	Lanceolate	Straight	Ob lanceolate	Incurved	Bilobed	Leathery, rigid
V <sub>11</sub>	Ovate	Straight	Suborbicular	Incurved	Bilobed	Glabrous
V <sub>12</sub>	Oblong	Deflexed with straight apex	Obtriangular	Incurved	Obtuse	Leathery, rigid
V <sub>13</sub>	Oblong	Deflexed with deflexed apex	Suborbicular	Incurved	Bilobed	Leathery, rigid
V <sub>14</sub>	Oblong	Deflexed with deflexed apex	Suborbicular	Incurved	Obtuse	Glabrous, rigid
V <sub>15</sub>	Oblong	Deflexed with deflexed apex	Obtriangular	Incurved	Bilobed	Glabrous, rigid

**Table 23. continued**

<b>Var. No.</b>	<b>Lip shape at apical lobe region</b>	<b>Nature of lateral lip</b>	<b>Lip shape at lateral lobe region</b>	<b>Nature of lateral lip</b>	<b>Lip apex</b>	<b>Lip surface</b>
V <sub>16</sub>	Oblong	Deflexed with deflexed apex	Suborbicular	Incurved	Bilobed	Leathery, rigid
V <sub>17</sub>	Ovate	Deflexed with deflexed apex	Ob lanceolate	Incurved	Bilobed	Leathery, rigid
V <sub>18</sub>	Oblong	Deflexed with deflexed apex	Ob triangle	Incurved	Bilobed	Leathery, rigid
V <sub>19</sub>	Ovate	Deflexed with deflexed apex	Suborbicular	Incurved	Bilobed	Leathery, rigid
V <sub>20</sub>	Ovate	Deflexed with deflexed apex	Suborbicular	Incurved	Bilobed	Glabrous, rigid,
V <sub>21</sub>	Ovate	Deflexed with deflexed apex	Suborbicular	Incurved	Bilobed	Leathery, rigid
V <sub>22</sub>	Oblong	Deflexed with straight apex	Ob triangle	Incurved	Bilobed	Leathery, rigid
V <sub>23</sub>	Oblong	Straight with deflexed apex	Ob triangle	Incurved	Bilobed	Leathery, rigid
V <sub>24</sub>	Oblong	Straight with deflexed apex	Ob triangle	Incurved	Bilobed	Leathery, rigid
V <sub>25</sub>	Oblong	Deflexed with deflexed apex	Ob triangle	Incurved	Bilobed	Leathery, rigid
V <sub>26</sub>	Oblong	Deflexed with deflexed apex	Ob triangle	Incurved	Bilobed	Leathery, rigid
V <sub>27</sub>	Lanceolate	Deflexed with straight apex	Ob lanceolate	Incurved	Bilobed	Glabrous, rigid,
V <sub>28</sub>	Lanceolate	Deflexed with straight apex	Ob lanceolate	Incurved	Obtuse	Leathery, rigid
V <sub>29</sub>	Lanceolate	Deflexed with straight apex	Ob lanceolate	Incurved	Obtuse	Leathery, rigid
V <sub>30</sub>	Lanceolate	Deflexed with straight apex	Ob lanceolate	Incurved	Obtuse	Leathery, rigid

apex, V<sub>23</sub> and V<sub>24</sub> were uniform, minutely spotted, tessellated whereas the V<sub>27</sub> was found to have blotched, spotted colour pattern (Table 22 and Appendix XXV).

#### 4.1.4.4 Lip characters

Qualitative characters with respect to lip shape, curvature, lip shape at lateral lobe region, lip colour at the base, margin, apex, and colour pattern are presented in Table 23 and 24.

Detectable variation was found with respect to shape of lip among the varieties. In V<sub>1</sub> and V<sub>3</sub> lip shape was found to be elliptic. In V<sub>7</sub>, V<sub>8</sub>, V<sub>10</sub>, V<sub>27</sub>, V<sub>28</sub>, V<sub>29</sub> and V<sub>30</sub> lip shape was found to be lanceolate. Ovate lip shape was found in V<sub>4</sub>, V<sub>5</sub>, V<sub>6</sub>, V<sub>11</sub>, V<sub>17</sub>, V<sub>19</sub>, V<sub>20</sub> and V<sub>21</sub>. However, in the rest of the varieties, lip shape was found to be oblong (Table 23 and Appendix XXV).

Lip curvature also showed wide variations among *Ascocentrum* hybrids/varieties. Straight lip was observed in V<sub>7</sub>, V<sub>8</sub>, V<sub>10</sub> and V<sub>11</sub>. Straight with deflexed apex was found in V<sub>23</sub> and V<sub>24</sub>. In V<sub>2</sub>, V<sub>3</sub>, V<sub>12</sub>, V<sub>22</sub>, V<sub>27</sub>, V<sub>28</sub>, V<sub>29</sub> and V<sub>30</sub> lip curvature was found to be deflexed with straight apex. In rest of the hybrids/varieties lip curvature was found to be deflexed with deflexed apex (Table 23 and Appendix XXV).

Lip shape at lateral lobe region was observed to have obtriangle in V<sub>1</sub>, V<sub>3</sub>, V<sub>6</sub>, V<sub>12</sub>, V<sub>15</sub>, V<sub>18</sub>, V<sub>22</sub>, V<sub>23</sub>, V<sub>24</sub>, V<sub>25</sub> and V<sub>26</sub>. In V<sub>9</sub> it was found to be semi circular while oblanceolate lip shape was observed in V<sub>7</sub>, V<sub>8</sub>, V<sub>10</sub>, V<sub>17</sub>, V<sub>27</sub>, V<sub>28</sub>, V<sub>29</sub> and V<sub>30</sub>. Suborbicular lip was found all other varieties (Table 23 and Appendix XXV).

There was not much variation observed with respect to the nature of lip. Nature of lip was strongly incurved in V<sub>2</sub> while it was normally incurved in rest of the varieties. Strong variation was not observed with respect to lip apex. Truncate lip apex was found in V<sub>1</sub> and obtuse tip apex was observed in V<sub>3</sub>, V<sub>9</sub>, V<sub>12</sub>, V<sub>14</sub>, V<sub>28</sub>, V<sub>29</sub> and V<sub>30</sub>. Whereas, in rest of the varieties bilobed lip apex was notice (Table 24 and Appendix XXV).

**Table 24. Qualitative lip characters of *Ascocentrum* hybrids/varieties**

<b>Var. No.</b>	<b>Lip base colour</b>	<b>Lip margin colour</b>	<b>Lip apex colour</b>	<b>Lip colour pattern</b>
V <sub>1</sub>	Single (Orangish yellow)	Single (strong orange)	Single (Strong orange)	Uniform
V <sub>2</sub>	Double (Yellow, maroon)	Triple (Maroon yellowish green, white)	Triple (Maroon, yellowish green, white)	Streaked, shaded
V <sub>3</sub>	Double (White, maroon)	Double (Violet, blue)	Double (Dark blue)	Shaded
V <sub>4</sub>	Double (Orangish yellow)	Single (Scarlet)	Double (Reddish orange)	Uniform
V <sub>5</sub>	Single (Pale yellow)	Double (Orangish yellow)	Double (Orangish yellow)	Shaded
V <sub>6</sub>	Single (Orange)	Single (Pale yellow)	Single (Orange)	Shaded
V <sub>7</sub>	Single (Purplish blue)	Single (Purplish blue)	Single (Purplish blue)	Uniform
V <sub>8</sub>	Single (Pink)	Single (Pink)	Single (Pink)	Uniform
V <sub>9</sub>	Double (Dark pink)	Double (Dark pink)	Double (Dark pink)	Blotched
V <sub>10</sub>	Single (Puplish Pink)	Single (Puplish Pink)	Single (Puplish Pink)	Uniform
V <sub>11</sub>	Single (White)	Single (White)	Single (White)	Uniform
V <sub>12</sub>	Single (pink)	Double (white, pink)	Single (pink)	Streaked, shaded
V <sub>13</sub>	Multiple (Maroon, yellow, white, pink)	Single (Maroon)	Single (Maroon)	Uniform, shaded
V <sub>14</sub>	Double (Orange, yellow, shaded)	Double (Orange, yellow, shaded)	Single (Orange)	Shaded, blotched
V <sub>15</sub>	Double (Orange, yellow, shaded)	Double (Orange, yellow, shaded)	Single (Brown)	Streaked, shaded, blotched

**Table 24. continued**

<b>Var. No.</b>	<b>Lip base colour</b>	<b>Lip margin colour</b>	<b>Lip apex colour</b>	<b>Lip colour pattern</b>
V <sub>16</sub>	Double (Orange, yellow, shaded, spotted)	Single (Maroon)	Single (Pale yellow)	Uniform
V <sub>17</sub>	Single (Yellow)	Single (Yellow)	Single (Yellow)	Uniform
V <sub>18</sub>	Double (Yellow, white)	Single (Pink)	Single (Pink)	Shaded
V <sub>19</sub>	Triple (Dark yellow, pink and white shaded)	Single (Pink)	Single (Pink)	Uniform, shaded
V <sub>20</sub>	Double (Yellow, brown shades)	Double (Yellow, brown shades)	Single (Brownish yellow)	Shaded
V <sub>21</sub>	Double (Orangish yellow, red, shades)	Double (Orangish yellow, red shades)	Double (Orangish yellow, red shades)	Shaded
V <sub>22</sub>	Double (Yellow, red shades)	Double (Yellow, red shades)	Single (Red)	Shaded
V <sub>23</sub>	Double (Yellow, red shades)	Double (Yellow, red shades)	Single (Red)	Streaked, shaded, spotted
V <sub>24</sub>	Double (Yellow, red shades)	Double (Yellow, red shades)	Single (Red)	Streaked, shaded, spotted
V <sub>25</sub>	Double (Yellow, red shades)	Double (Yellow dotted, red)	Single (Red)	Streaked, shaded, spotted
V <sub>26</sub>	Double (Dark pink with shades and dots)	Single (Dark pink)	Double (Dark pink dots & streaks)	Streaked, shaded, spotted
V <sub>27</sub>	Double (Yellow and red)	Double (Yellow and red)	Single (red, blotched)	Streaked, shaded, blotched
V <sub>28</sub>	Double (Yellow and red)	Double (Yellow and red)	Single (red)	Streaked, shaded, blotched
V <sub>29</sub>	Double (Yellow and red)	Double (Yellow and red)	Single (red)	Streaked, shaded, blotched
V <sub>30</sub>	Double (Yellow and red)	Double (Yellow and red)	Single (red)	Streaked, shaded, blotched

Lip surface also showed some variation among the hybrids/ varieties. Glabrous lip surface was found in V<sub>11</sub> and V<sub>14</sub> and V<sub>15</sub>. In V<sub>14</sub>, V<sub>15</sub>, V<sub>20</sub> and V<sub>27</sub> lip surface was glabrous and rigid surface while in rest of the varieties lip surface was found to be leathery and rigid (Table 24).

Profound variation was observed in lip base colour, lip margin colour and lip apex colour (Table 24). In V<sub>13</sub> multiple colours were found at the base of lip. Three colours at the lip base of V<sub>19</sub>. Single colour at the lip base was found in V<sub>1</sub>, V<sub>5</sub>, V<sub>6</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>9</sub>, V<sub>10</sub>, V<sub>11</sub> and V<sub>12</sub>. And in rest of the varieties two colours were observed at the base of lip (Table 24). Three colours were observed on the lip margin of V<sub>2</sub> were found. Double coloured lip margin was found in V<sub>3</sub>, V<sub>5</sub>, V<sub>9</sub>, V<sub>12</sub>, V<sub>14</sub>, V<sub>15</sub>, V<sub>20</sub>, V<sub>21</sub>, V<sub>22</sub>, V<sub>23</sub>, V<sub>24</sub>, V<sub>25</sub>, V<sub>27</sub>, V<sub>28</sub>, V<sub>29</sub> and V<sub>30</sub>. However single colour was only found on the lip margin of rest of the varieties. Similarly, in case of colour of lip apex. Three colours were found on the lip apex of V<sub>2</sub> and two colours were found on the lip apex of V<sub>4</sub>, V<sub>5</sub>, V<sub>9</sub>, V<sub>21</sub> and V<sub>26</sub>. While only single colour was found in the rest of the varieties.

Appreciable differences were found in lip colour pattern also it was uniform in V<sub>1</sub>, V<sub>4</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>16</sub> and V<sub>17</sub> and shaded in V<sub>3</sub>, V<sub>5</sub>, V<sub>6</sub>, V<sub>18</sub>, V<sub>20</sub>, V<sub>21</sub> and V<sub>22</sub>. In V<sub>2</sub> and V<sub>12</sub> it was found to be streaked and shaded. In V<sub>15</sub>, V<sub>27</sub>, V<sub>28</sub>, V<sub>29</sub> and V<sub>30</sub> it was found to be streaked, shaded and blotched, In V<sub>23</sub>, V<sub>24</sub>, V<sub>25</sub> and V<sub>26</sub> it was found to observe streaked, shaded and spotted. Shaded and blotched lip colour pattern was observed in V<sub>14</sub> while in V<sub>13</sub> and V<sub>19</sub>, lip colour pattern was found to be uniform and shaded (Table 24).

#### **4.1.4.4.5 Column characters**

Variation was found with respect to column characters among the varieties. Detailed about the colour, colour pattern, and length is presented in Table 25. Double coloured column with shaded colour pattern (shaded and blotched) was found in V<sub>2</sub>, V<sub>4</sub>, V<sub>18</sub> and V<sub>19</sub>, while in rest of all other varieties colour pattern is uniform in a single colour. In V<sub>3</sub>, V<sub>15</sub>, V<sub>16</sub>, V<sub>17</sub>, V<sub>18</sub>, V<sub>21</sub> and V<sub>24</sub>

long length of column was found. However, in the rest of all other varieties medium column length was observed.

**Table 25. Qualitative column and spur characters of *Ascocentrum* hybrids/varieties**

<b>Var. No.</b>	<b>Column colour</b>	<b>Column colour pattern</b>	<b>Column length</b>	<b>Spur length</b>	<b>Spur type</b>
V <sub>1</sub>	Single (yellow)	Uniform	Medium	Medium	Cylindrical, saccat at the base
V <sub>2</sub>	Double (white, maroon)	Shaded	Medium	Medium	Conical
V <sub>3</sub>	Single (white)	Uniform	Long	Long	Cylindrical
V <sub>4</sub>	Double (yellow, orange)	Shaded	Medium	Medium	Cylindrical
V <sub>5</sub>	Single (pale yellow)	Uniform	Medium	Medium	Conical
V <sub>6</sub>	Single (pale yellow)	Uniform	Medium	Medium	Cylindrical
V <sub>7</sub>	Single (white)	Uniform	Medium	Medium	Cylindrical
V <sub>8</sub>	Single (white)	Uniform	Medium	Medium	Cylindrical
V <sub>9</sub>	Single (Pink, white)	Blotched	Medium	Medium	Conical
V <sub>10</sub>	Single (white)	Uniform	Medium	Medium	Cylindrical
V <sub>11</sub>	Single (white)	Uniform	Medium	Medium	Conical
V <sub>12</sub>	Single (white)	Uniform	Medium	Short	Conical
V <sub>13</sub>	Single (white)	Uniform	Medium	Short	Conical
V <sub>14</sub>	Single (pale yellow)	Uniform	Medium	Medium	Conical
V <sub>15</sub>	Single (pale yellow)	Uniform	Long	Medium	Conical
V <sub>16</sub>	Single (pale yellow)	Uniform	Long	Medium	Conical
V <sub>17</sub>	Single (pale yellow)	Uniform	Long	Medium	Cylindrical
V <sub>18</sub>	Double (white, pink)	Shaded	Long	Short	Conical
V <sub>19</sub>	Double (white, pink)	Shaded	Medium	Medium	Cylindrical
V <sub>20</sub>	Single (pale yellow)	Uniform	Medium	Short	Conical
V <sub>21</sub>	Single (pale yellow)	Uniform	Long	Medium	Conical
V <sub>22</sub>	Single (pale yellow)	Uniform	Medium	Medium	Conical
V <sub>23</sub>	Single (pale yellow)	Uniform	Medium	Medium	Conical
V <sub>24</sub>	Single (pale yellow)	Uniform	Long	Short	Conical
V <sub>25</sub>	Single (yellowish white)	Uniform	Medium	Medium	Conical
V <sub>26</sub>	Single (pale yellow)	Uniform	Medium	Medium	Conical
V <sub>27</sub>	Single (yellowish red)	Uniform	Medium	Long	Tubular
V <sub>28</sub>	Single (yellowish red)	Uniform	Medium	Medium	Cylindrical, saccat at the base
V <sub>29</sub>	Single (yellowish red)	Uniform	Medium	Medium	Cylindrical, saccat at the base
V <sub>30</sub>	Single (yellowish red)	Uniform	Medium	Medium	Cylindric, saccat at the base



#### **4.1.4.4.6 Spur characters**

Detectable variation was observed with respect to spur length and spur type (Table 25). In V<sub>12</sub>, V<sub>13</sub>, V<sub>18</sub> and V<sub>20</sub> short spur length were observed and in V<sub>3</sub> and V<sub>27</sub> spur length observed is longest while medium spur length was observed in rest of all other varieties. The spur type was cylindrical but saccate at the base in V<sub>1</sub>, V<sub>28</sub>, V<sub>29</sub> and V<sub>30</sub>. Completely cylindrical type of spurs was noticed in V<sub>3</sub>, V<sub>4</sub>, V<sub>6</sub>, V<sub>7</sub>, V<sub>8</sub>, V<sub>17</sub> and V<sub>19</sub>. The tubular one was found in V<sub>27</sub> whereas, conical type of spurs was noticed in the rest of all other varieties.

#### **4.1.5 Visual evaluation**

Data regarding the scores obtained for the spikes and plants of thirty *Ascocentrum* hybrids/varieties are presented in Table 26 and 27. Scoring was done for spike for use as a cut flower and plant for use in the indoor display.

##### **4.1.5.1 Spike for use as a cut flower**

Scores were given according to colour and pigmentation, texture shape and pattern, size and orientation of floret, spike longevity in vase, compactness and visual appeal. Data on scoring is presented in Table 26.

The highest mean total score was obtained out of 60 in V<sub>23</sub> (54.60), whereas, least total mean score was obtained in V<sub>11</sub> (51.07). However, there was no significant variation found among the varieties with respect to total score. Floret colour and pigmentation, shape and pattern, compactness and visual appeal were found to be significantly superior among the varieties with respect to visual score for spike use as cut flower. Where, V<sub>16</sub>, V<sub>25</sub> and V<sub>20</sub> were found with high score and best varieties to use as cut flower. (Table 26).

##### **4.1.5.2 Plant for indoor display**

Scoring was done for plants according to growth and fullness, spread and orientation, spike colour and pigmentation, size and orientation of spike, spike longevity on a plant, visual appeal and general appearance. Data on scoring is presented in Table 27.

**Table 26. Visual scoring for the spikes of *Ascozentrum* hybrids/varieties**

Var. No.	Scoring for the spike for use as a cut flower (each out of 10)						Total (out of 60)
	Floret colour & pigmentation	Texture	Shape & pattern	Size & orientation of floret	Spike longevity in vase	Compactness & visual appeal	
V <sub>1</sub>	8.90	8.93	7.93	8.47	8.53	8.53	51.30
V <sub>2</sub>	8.87	8.87	8.60	8.80	8.73	8.73	52.60
V <sub>3</sub>	8.73	8.97	8.77	9.03	9.07	8.23	52.80
V <sub>4</sub>	8.87	8.80	8.37	8.80	8.73	8.40	51.97
V <sub>5</sub>	9.00	9.03	8.67	8.97	8.93	8.17	52.77
V <sub>6</sub>	8.93	9.03	8.53	8.77	8.83	8.53	52.63
V <sub>7</sub>	9.00	9.03	8.67	8.87	8.83	8.47	52.87
V <sub>8</sub>	8.77	8.80	8.57	8.77	8.73	8.43	52.07
V <sub>9</sub>	8.87	8.87	8.77	8.80	8.80	7.97	52.07
V <sub>10</sub>	8.87	8.90	8.77	8.80	8.80	8.43	52.57
V <sub>11</sub>	8.47	8.43	8.50	8.43	8.73	8.50	51.07
V <sub>12</sub>	8.87	8.83	8.83	8.93	8.50	8.67	52.63
V <sub>13</sub>	8.53	8.70	8.70	8.90	8.57	8.63	52.03
V <sub>14</sub>	9.07	8.97	8.90	8.87	8.80	8.83	53.43
V <sub>15</sub>	9.07	8.97	9.00	8.97	8.97	8.80	53.77
V <sub>16</sub>	9.33	8.63	8.77	8.60	8.43	8.77	52.53
V <sub>17</sub>	8.27	8.50	8.73	8.87	8.47	8.63	51.47
V <sub>18</sub>	8.90	8.97	8.87	8.97	8.67	8.97	53.33
V <sub>19</sub>	8.93	8.83	8.93	8.90	8.80	9.00	53.40
V <sub>20</sub>	9.37	8.87	8.87	8.90	8.83	9.40	54.23
V <sub>21</sub>	9.00	8.87	8.93	9.07	9.07	8.90	53.83
V <sub>22</sub>	8.80	8.90	8.87	8.53	8.47	8.70	52.27
V <sub>23</sub>	9.10	9.27	8.87	9.70	8.83	8.83	54.60
V <sub>24</sub>	8.83	8.97	8.87	8.83	8.73	8.60	52.83
V <sub>25</sub>	8.87	8.73	9.23	8.87	8.87	9.23	53.80
V <sub>26</sub>	8.80	8.90	8.87	9.17	9.10	8.87	53.70
V <sub>27</sub>	8.90	8.90	8.93	8.90	8.83	8.87	53.33
V <sub>28</sub>	8.57	9.20	8.73	8.53	8.57	9.03	52.63
V <sub>29</sub>	8.83	9.07	8.77	8.73	8.97	8.63	53.00
V <sub>30</sub>	8.77	8.73	8.73	8.83	8.87	8.73	52.67
<b>CD</b>	<b>0.468</b>	<b>NS</b>	<b>1.398</b>	<b>NS</b>	<b>NS</b>	<b>0.552</b>	<b>NS</b>
<b>CV</b>	<b>3.204</b>	<b>11.147</b>	<b>9.851</b>	<b>10.564</b>	<b>10.722</b>	<b>3.905</b>	<b>10.463</b>

**Table 27. Plant quality rating of *Ascozentrum* hybrids/varieties**

Var. No.	Scoring for the plant for use in the indoor display (each out of 10)						Total (out of 60)
	Growth & fullness	Spread & orientation of leaves	Spike colour & pigmentation	Size & orientation of spike	Spike longevity on plant	Visual appeal & general appearance	
V <sub>1</sub>	9.00	8.67	8.97	8.67	9.00	8.90	53.20
V <sub>2</sub>	8.70	8.67	8.77	8.57	8.80	8.77	52.27
V <sub>3</sub>	9.00	8.70	8.87	8.77	8.87	8.80	53.00
V <sub>4</sub>	8.80	8.97	8.87	8.77	8.90	8.80	53.10
V <sub>5</sub>	8.93	8.87	8.83	8.93	8.83	8.50	52.90
V <sub>6</sub>	8.87	8.53	8.70	8.90	8.70	8.57	52.27
V <sub>7</sub>	8.87	9.07	8.97	8.87	8.90	8.80	53.47
V <sub>8</sub>	8.87	9.27	9.03	8.97	9.03	8.67	53.83
V <sub>9</sub>	8.83	9.00	8.87	9.07	8.93	9.07	53.77
V <sub>10</sub>	8.77	8.90	8.90	8.90	8.93	8.83	53.23
V <sub>11</sub>	9.13	8.83	8.97	8.97	8.87	8.97	53.73
V <sub>12</sub>	8.73	8.93	8.83	8.90	8.93	8.80	53.13
V <sub>13</sub>	8.80	8.93	8.87	8.90	8.87	8.83	53.20
V <sub>14</sub>	8.85	8.93	8.03	8.47	8.40	8.77	48.50
V <sub>15</sub>	8.47	8.90	8.53	8.80	8.83	8.50	52.03
V <sub>16</sub>	8.70	8.90	8.67	8.90	8.90	8.37	52.43
V <sub>17</sub>	9.00	8.83	8.60	8.90	8.90	8.37	52.60
V <sub>18</sub>	8.80	8.83	8.57	8.93	9.03	8.40	52.57
V <sub>19</sub>	8.93	8.80	8.60	8.87	8.80	8.33	52.33
V <sub>20</sub>	8.87	8.80	8.57	8.70	8.73	8.37	52.03
V <sub>21</sub>	8.70	8.63	8.43	8.80	8.83	8.73	52.13
V <sub>22</sub>	8.97	8.93	8.17	8.83	8.83	7.90	51.63
V <sub>23</sub>	8.77	8.83	8.53	8.80	8.80	8.43	52.17
V <sub>24</sub>	8.73	8.47	8.20	8.87	8.73	8.37	51.37
V <sub>25</sub>	8.60	8.47	8.57	8.47	8.60	8.67	51.37
V <sub>26</sub>	8.87	9.07	8.97	8.97	8.87	8.67	53.40
V <sub>27</sub>	8.63	8.40	8.67	8.23	8.83	8.13	50.90
V <sub>28</sub>	8.93	9.00	8.90	8.70	8.70	8.70	52.93
V <sub>29</sub>	8.50	8.43	8.73	8.33	8.50	8.13	50.63
V <sub>30</sub>	8.87	8.73	8.70	8.70	8.73	8.60	52.33
<b>CD</b>	<b>NS</b>	<b>NS</b>	<b>NS</b>	<b>0.352</b>	<b>NS</b>	<b>0.499</b>	<b>NS</b>
<b>CV</b>	<b>11.376</b>	<b>3.375</b>	<b>11.580</b>	<b>2.476</b>	<b>3.405</b>	<b>3.514</b>	<b>11.001</b>

The highest mean total score was obtained out of 60 in V<sub>8</sub> (53.83) whereas, the least mean total score was obtained in V<sub>17</sub> (48.50). However, the significant variation was not observed among varieties with regard to the total score. Size and orientation of spikes, visual appeal and general appearance of plant were found significantly superior among the varieties with respect plant quality rating for use in indoor display. Where V<sub>9</sub> and V<sub>10</sub> were found with high rating score for plant use in indoor display.

#### 4.1.6 Incidence of pests and diseases

The list of pests and diseases, plant parts affected and number of varieties affected along with the control measure adopted are given in Table 27. Short horned grasshopper was observed on pods and different floral parts *viz.*, rachis and peduncle of spikes, petals and pollinium of florets of *Ascda*. Udomochai, *Ascda*. Yip Sum Wah × *Vanda* JVB, *Ascda*. Suksamran Sunlight, *Mok*. Rassmatozz, *Mok*. Khaw Phiak Suan × *Ascda*. Bicentennial Yellow Spot and *Mok*. Calypso Pink. NeemAzal 2ml per liter was sprayed for the control of the pest (Table 28).

*Oecophylla smaragdina* ants were found to affect the tender parts of petals, pedicel and also the pollinium. Incidence of this pest was observed on five varieties *viz.*, *Ascda*. Udomochai, *Ascda*. Suksamran Sunlight, *Mok*. Rassmatozz, *Mok*. Khaw Phiak Suan × *Ascda*. Bicentennial Yellow Spot and *Mok*. Calypso Pink. Ekalux and NeemAzal 2 ml per liter were sprayed to control the pests (Table 28).

Aphids and thrips attack was also observed on spikes of 16 varieties *viz.*, *Mok*. Calypso × *V. Dr. Anek*, *Mok*. Rassmatozz, *Mok*. Khaw Phiak Suan × *Ascda*. Bicentennial Yellow Spot, *Mok*. Khaw Phiak Suan × *Ascda*. Jiraprapra, *Mok*. Sayan × *Ascda*. Bangkuntein Gold, *Mok*. Calypso Pink, *Mok*. Calypso Jumbo, *Mok*. Chao Praya Sunset Yellow Spot, *Mok*. Chao Praya Sunset Orange, *Mok*. Sunspot, *Mok*. Omayaiy Yellow, *Mok*. Omayaiy Orange, *Mok*. Sayan × *Ascda*. Doung Porn, *Mok*. Chark Kuan Pink, *Kag*. Youthong Beauty, *Kag*. Christie

Low and *Kag*. Boon Ruby. Infected plants sprayed with oberon @ 0.8ml per liter (Table 28).

*Alternaria* leaf spot, heart rot, bacterial wilt and anthracnose were observed on leaves of 5 to 8 varieties of *Mokara* and *Ascocenda*. Infected plants were sprayed with methylobacter 2.5 ml per liter as control measure (Table 28).

**Table 28. Incidence of pests and diseases throughout study period**

Sl. No.	Pest and disease	Affected plant part	No of varieties affected	Control measure
1	Short horned grasshopper	Pods, Spikes, (rachis and peduncle) florets	6	NeemAzal 2 ml/lit
2	Ants ( <i>Oecophylla</i> )	Florets at bud stage, pedicel, petals and pollinia	5	Ekalux 2 ml/lit
3	Aphid	Spikes and florets	16	Oberon 0.8 ml/lit
4	Thrips	Spikes and florets	8	Oberon 0.8 ml/lit
5	Leaf spot	Leaves	4	Methyalobacter 2.5 ml/lit as a disease resistant, tebuconazole 1ml/lit, phylotene 3.5 g/lit
6	Heart rot	Leaves	2	
7	Bacterial wilt	Leaves	3	
8.	Anthracnose	Leaves	2	

## 4.2 COMPATIBILITY STUDIES

Data pertaining to compatibility studies among eleven selected varieties with respect to their anthesis time, stigma receptivity (days), pollen studies, post pollination changes, self and cross compatibilities are presented in Table 28 to 32.

### 4.2.1 Anthesis

Anthesis was observed to be slow during the day time but faster during morning and evening hours. In selected eleven *Ascocentrum* hybrids, flowers opened during morning and during evening, flower opening started from morning 5.30 am-11.30 am and afternoon 3.00 pm-6.00 pm (Table 28).

However, in *Vasco*. Aroonsri Beauty anthesis observed comparatively faster (5.30 am-6.15 am and 4.00 am-4.30 pm) composed to rest of the varieties. In *Ascda*. Udomochai anthesis time recorded from morning 8.30 am-10 00 am and afternoon 3.30 to 4.30 pm. In hybrid *Ascda*. Kultana × *V. Bitzs Heartthrob* anthesis was observed in 9.30 am-11.00 am and 4.00 pm-5.30 pm. During 6.30 am-8.30 am and 5.30 am-6.00 pm, flower opening was observed in *Ascda*. Sirichi Fragrance.

In *Vasco*. Pine River Blue flower opening was observed 9.00 - 11 00 am and 4.00 - 5.30 pm while in *Vasco*. Pine River Pink anthesis was observed during 8.30 to 10 30 am and 4.30 - 5.30 pm. During 8.30 - 10.00 am and 3.00 - 4.30 pm flower opening was observed in *Vasco*. Pine Rivers Fuchsia Delight. During 7.30 - 9.30 am and 3.30 to 4.30 pm anthesis time was recorded in *Mok*. Calypso Pink. In *Mok*. Chao Praya Sunset Yellow Spot complete flower opening was observed in 10.00 to 11.30 am and 4.00 to 5.30 pm. In *Mok*. Sayan × *Ascda*. Doung Porn anthesis time was recorded during 6.30 to 8 00 am and 4.30 to 5.50 pm. In *Kag*. Youthong Beauty complete opening of floret was observed in the morning from 7.30 - 9 00 am and in the evening 5.30 to 6.00 pm.

#### **4.2.2 Stigma receptivity**

Details of stigma receptivity are presented in Table 29. Stigma receptivity ranged from 1-14 days after anthesis.

The longest duration of stigma receptivity was found in *Ascda*. Kultana × *V. Bitzs Heartthrob* from 2<sup>nd</sup> to 14<sup>th</sup> day followed by *Mok*. Chao Praya Sunset Yellow Spot (4-14 days), *Vasco*. Pine River Pink (2-12 days) and *Vasco*. Pine River Blue (2-10 days). The stigma was remained receptive for minimum duration in *Vasco*. Aroonsri Beauty (1-2 days) and *Ascda*. Sirichi Fragrance (2-4 days). However, the stigma found to be receptive on the first day of anthesis in *Ascda*. Udomochai. Maximum stigma receptivity was observed during 1<sup>st</sup> - 6<sup>th</sup> day, in *Vasco*. Pine Rivers Fuchsia Delight (1-8 days) and in *Vasco*. Aroonsri Beauty (1-2 days).

The stigma was remained receptive from the second day of anthesis in *Ascda*. Sirichi Fragrance (2-4 days), *Mok*. Calypso Pink (2-8 days), *Mok*. Sayan × *Ascda*. Doung Porn from 2-8 days, *Kag*. Youthong Beauty (2-6 days) and *Ascda*. Kultana × *Vanda* Bitzs Heartthrob from (2-14 days), *Vasco*. Pine River Pink (2-12 days) and *Vasco*. Pine River Blue (2-10 days). The stigma remained receptive from the fourth day of anthesis in *Mok*. Chao Praya Sunset Yellow Spot (4-14 days) and *Vasco*. Pine River Blue (4-10).

**Table 29. Anthesis time and stigma receptivity period in the parents**

Sl. No.	Hybrid/ variety name.	Anthesis time	Maximum stigma receptivity (days)
1	<i>Ascda</i> . Udomochai	8.30 to 10 00 am and 3.30 to 4.30 pm	1-6
2	<i>Ascda</i> . Kultana × <i>V</i> . Bitzs Heartthrob	9.30 to 11.00 am and 4.00 to 5.30 pm	2-14
3	<i>Ascda</i> . Sirichi Fragrance	6.30 to 8.30 am and 5.30 to 6.00 pm	2-4
4	<i>Vasco</i> . Pine River Blue	9.00 to 11 00 am and 4.00 to 5.30 pm	4-10
5	<i>Vasco</i> . Pine River Pink	8.30 to 10 30 am and 4.30 to 5.30 pm	2-12
6	<i>Vasco</i> . Aroonsri Beauty	5.30 to 6.15 am and 4.00 to 4.30 pm	1-2
7	<i>Vasco</i> . Pine Rivers Fuchsia Delight	8.30 to 10.00 am and 3.00 to 4.30 pm	1-8
8	<i>Mok</i> . Calypso Pink	7.30 to 9.30 am and 3.30 to 4.30 pm	2-8
9	<i>Mok</i> . Chao Praya Sunset Yellow Spot	10.00 to 11.30 am and 4.00 to 5.30 pm	4-14
10	<i>Mok</i> . Sayan × <i>Ascda</i> . Doung Porn	6.30 to 8 00 am and 4.30 to 5.50 pm	2-8
11	<i>Kag</i> . Youthong Beauty	7.30 to 9 00 am and 5.30 to 6.00 pm	2-6

### 4.2.3 Pollen studies

Pollen studies were done in eleven selected varieties with respect to pollen production per pollinia, pollen viability and pollen germination data are presented in Table 30.

#### 4.2.3.1 Pollen production per pollinium

Pollen production per pollinium/pollinia (Table 30) was noted to be the highest in *Ascda. Kultana* × *V. Bitzs Heartthrob* (198611.1) followed by *Mok. Sayan* × *Ascda. Doung Porn* (197222.2), *Ascda. Sirichi Fragrance* (190277.8) and *Vasco. Aroonsri Beauty* (90740.70). However, the minimum pollen production per pollinium was noticed in *Kag. Youthong Beauty* (40740.7) followed by *Ascda. Udomochai* (49074.1), *Mok. Chao Praya Sunset Yellow Spot* (57407.4), *Mok. Calypso Pink* (59259.3), *Vasco. Pine Rivers Fuchsia Delight* (67592.6), *Vasco. Pine River Pink* (75694.4) and *Vasco. Pine River Blue* (77083.3) (Plate 9).

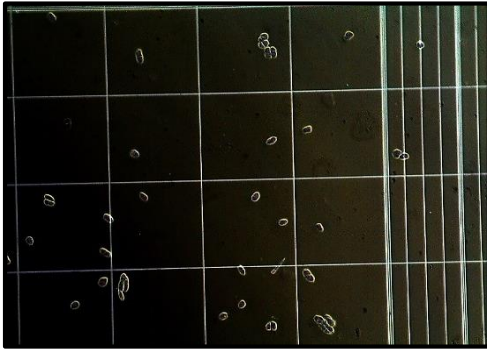
#### 4.2.3.2 Pollen viability/ fertility

Percentage of pollen viability (Table 30) was observed to be highest in (100%) *Ascda. Sirichi Fragrance* in *Mok. Sayan* × *Ascda. Doung Porn* which are closely followed by *Ascda. Udomochai* (98.38 %), *Vasco. Pine River Pink* (98.14 %) and *Vasco. Aroonsri Beauty* (97.00 %). The lowest percentage of fertile pollen was observed in *Kag. Youthong Beauty* (72.4 %) followed by *Mok. Chao Praya Sunset Yellow Spot* (79.33 %), *Ascda. Kultana* × *V. Bitzs Heartthrob* (83.92 %), *Vasco. Pine Rivers Fuchsia Delight* (84.25 %), *Mok. Calypso Pink* (87.54 %) and 92.33 percent in *Vasco. Pine River Blue* (Plate 9 and 10).

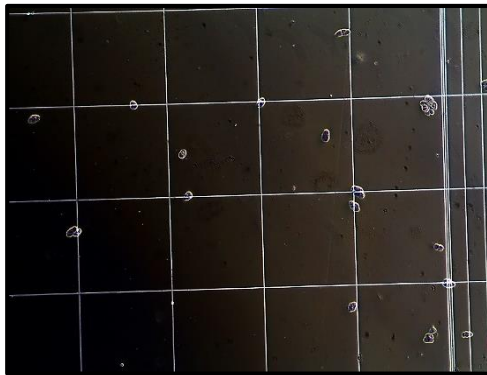
#### 4.2.3.3 Pollen germination

Pollen germination percentage was observed to be highest in *Ascda. Sirichi Fragrance* (86.33 %) followed by *Mok. Sayan* × *Ascda. Doung Porn* (77.47 %) while below 60 per cent of pollen germination was observed in the rest of the varieties. In *Kag. Youthong Beauty* had the lowest pollen germination (21.50 %) was observed followed by *Mok. Chao Praya Sunset Yellow Spot* was (25.57 %),





(A) Pollen count- *Ascda. Kultana* × *V. Bitz's Heartthrob & Mok. Sayan* × *Ascda. Doung Porn*

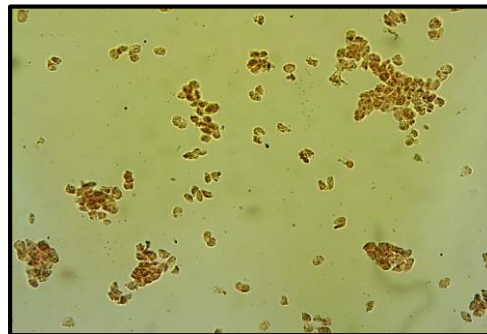


(B) Pollen count- *Ascda. Sirichi Fragrance*

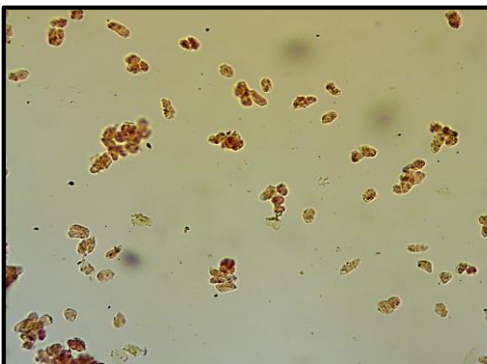


(C)  
Sterile  
pollen

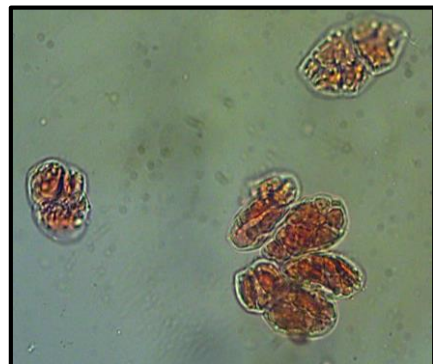
(C) Viable pollens



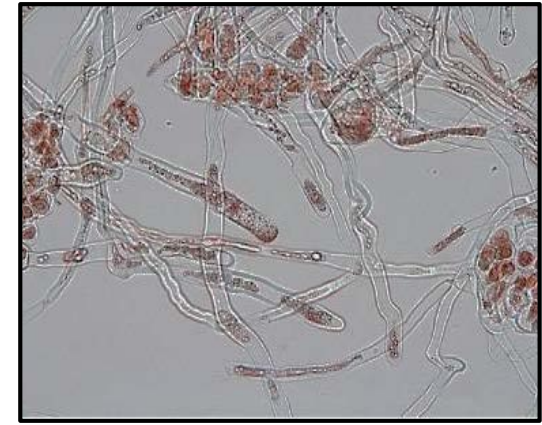
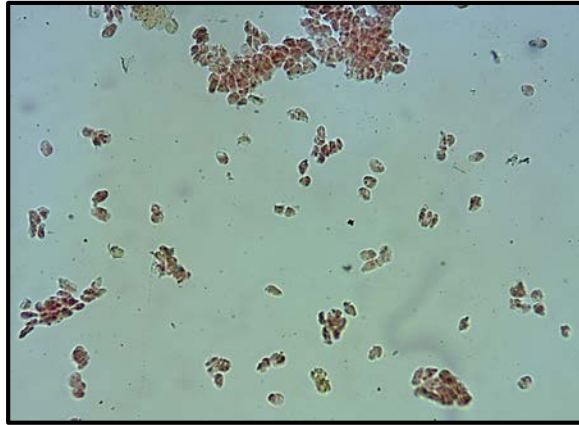
(D) Pollen viability of *Ascda. Sirichi Fragrance* under 10x and 40x lenses



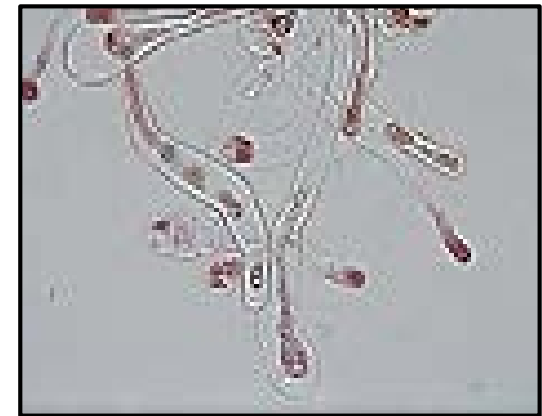
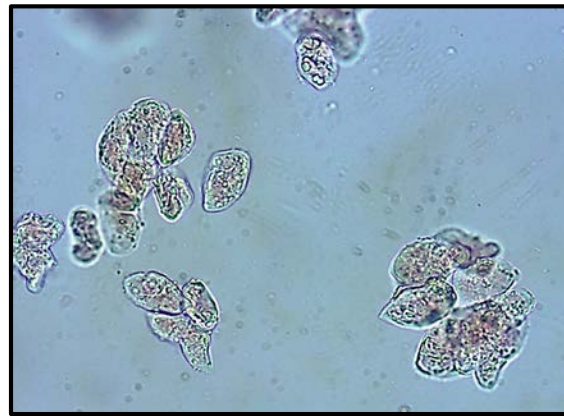
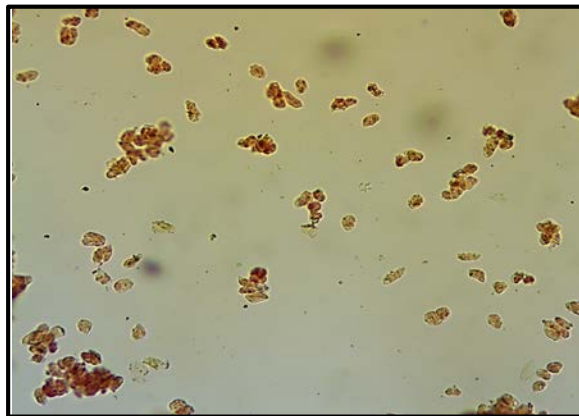
(E) Pollen viability of *Mok. Sayan* × *Ascda. Doung Porn* under 10x and 40x lenses



**Plate 9. Micro graphs of pollen count using haemocytometer and viability under low and high power microscope**



(A) Pollen viability of *Ascda*. Sirichi Fragrance under 10x and 40x lenses and germination under 10x lens



(B) Pollen viability of *Mok. Sayan* × *Ascda*. Doung Porn under 10x and 40x lenses and germination under 10x lens

**Plate 10. Pollen viability and germination in selected male parents under low and high power microscope**

*Vasco*. Pine River Blue (33.26 %), *Vasco*. Pine Rivers Fuchsia Delight (40.20 %), *Ascda*. Kultana × *V. Bitzs* Heartthrob (40.54 %), *Ascda*. Udomochai (42.33 %), *Vasco*. Aroonsri Beauty (51.33 %) and *Vasco*. Pine River Pink (52.67 %) and the same presented in Table 30 (Plate 10).

#### 4.2.4 Self compatibility

Self compatibility studies were done in the selected eleven parents and details regarding post pollination changes after self pollination (Plate 11 and 12) and self compatibility studies among the parents are presented in Table 31.

**Table 30. Pollen studies in eleven selected hybrids/varieties**

Sl. No.	Hybrid/ variety name.	Pollen production per pollinia (No.)	Pollen fertility %	Pollen germination %
1	<i>Ascda</i> . Udomochai	49074.1	98.38	42.33
2	<i>Ascda</i> . Kultana × <i>V. Bitzs</i> Heartthrob	198611.1	83.92	40.54
3	<i>Ascda</i> . Sirichi Fragrance	190277.8	100.00	86.33
4	<i>Vasco</i> . Pine River Blue	77083.3	92.33	33.26
5	<i>Vasco</i> . Pine River Pink	75694.4	98.14	52.67
6	<i>Vasco</i> . Aroonsri Beauty	90740.7	97.00	51.33
7	<i>Vasco</i> . Pine Rivers Fuchsia Delight	67592.6	84.25	40.20
8	<i>Mok</i> . Calypso Pink	59259.3	87.54	44.43
9	<i>Mok</i> . Chao Praya Sunset Yellow Spot	57407.4	79.33	25.57
10	<i>Mok</i> . Sayan × <i>Ascda</i> . Doung Porn	197222.2	100.00	77.47
11	<i>Kag</i> . Youthong Beauty	40740.7	72.4	21.50

The varieties *Ascda*. Udomochai, *Ascda*. Kultana × *V. Bitzs* Heartthrob, *Ascda*. Sirichi Fragrance, *Vasco*. Pine River Blue and *Vasco* and *Mok*. Sayan × *Ascda*. Doung Porn. Pine River Pink are found to be self-compatible, whereas therest of the varieties *viz.*, *Vasco*. Aroonsri Beauty, *Vasco*. Pine Rivers Fuchsia



(A) *Vasco. Aroonsri Beauty*



(B) *Kag. Youthong Beauty*



(C) *Ascda. Udomochai*



(D) *Mok. Chao Praya Sunset Yellow Spot*



(E) *Vasco. Pine Rivers Fuchsia Delight*



(F) *Vasco. Pine River Blue*

**Plate 11. Enlargement of pedicel after pollination in *Ascocentrum* hybrids**

Delight, *Mok.* Calypso Pink, *Mok.* Chao Praya Sunset Yellow Spot and *Kag.* Youthong Beauty were found to be self incompatible.

#### **4.2.5 Post pollination changes after selfing**

The data pertaining to post pollination changes after the self pollination (Plate 11 to 14), with respect to flower fall, enlargement of pedicel, pod development, percentage of pod set, days taken for maturity, percentage of *in vitro* seed germination and days for planting out are presented in Table 31.

##### **4.2.5.1 Flower fall**

Flower fall was found within a week in *Vasco.* Aroonsri Beauty, *Mok.* Calypso Pink and *Kag.* Youthong Beauty and was not found in rest of the varieties, they remained attached to spike even after a week and more (Table 31 and Plate 12).

##### **4.2.5.2 Enlargement of the pedicel**

Enlargement of the pedicel was found in *Ascda.* Udomochai, *Ascda.* Kultana × *V.* Bitzs Heartthrob, *Ascda.* Sirichi Fragrance, *Vasco.* Pine River Blue, *Vasco.* Pine River Pink, *Vasco.* Pine Rivers Fuchsia Delight, *Mok.* Chao Praya Sunset Yellow Spot and *Mok.* Sayan × *Ascda.* Doung Porn. Enlargement of pedicel was not found to be noticed in *Mok.* Calypso Pink, *Vasco.* Aroonsri Beauty and *Kag.* Youthong Beauty (Table 31 and Plate 11).

##### **4.2.5.3 Pod development (pod set)**

Pod set was found in *Ascda.* Udomochai, *Ascda.* Kultana × *V.* Bitzs Heartthrob, *Ascda.* Sirichi Fragrance, *Vasco.* Pine River Blue, *Vasco.* Pine River Pink and *Mok.* Sayan × *Ascda.* Doung Porn while the pod development was not observed in *Vasco.* Pine Rivers Fuchsia Delight and *Mok.* Chao Praya Sunset Yellow Spot (Table 31, Plate 11 and 12).

#### **4.2.5.4 Percentage of pod set**

The highest percentage of pod set (Table 31) was found in *Ascda. Kultana* × *Vanda* Bitzs Heartthrob (100 %) followed by *Ascda. Sirichi* Fragrance (86.48 %) and *Ascda. Udomochai* (66.87 %) and *Mok. Sayan* × *Ascda. Doung Porn* (64.00%), whereas the lowest percentage of pod set was found in *Vasco. Pine River Pink* (50.00%) and *Vasco. Pine River Blue* (33.30 %).

#### **4.2.5.5 Days taken for maturity of pod**

Minimum number of days was taken for maturity of pods (Table 31) in *Ascda. Udomochai* (124 days) followed by *Vasco. Pine River Blue* took (126.2 days), *Vasco. Pine River Pink* (139.4 days) and the maximum number of days was taken for maturity of pods was found in *Ascda. Kultana* × *V. Bitzs Heartthrob* (148 days). However, in *Ascda. Sirichi* Fragrance and *Mok. Sayan* × *Ascda. Doung Porn* pods were not matured fully, they shrivelled and dried on plant, even after the healthy pod development (dried after 2 to 3.5 months).

#### **4.2.5.6 Seed germination (%)**

Seeds were extracted from the matured pod and were cultured under *in vitro* condition. Highest percent of seed germination was found in *Vasco. Pine River Pink* (87.26 %) followed by *Ascda. Kultana* × *V. Bitzs Heartthrob* (84.4 %). However, protocorms initiations showed declined growth after seed germination in *Ascda. Udomochai* (Table 31 and Plate 14).

#### **4.2.5.7 Days for planting out**

Minimum number of days was taken for planting out in *Vasco. Pine River Pink* (268.3 days) and maximum number of days was taken for planting out in *Ascda. Kultana* × *V. Bitzs Heartthrob* (312 days) (Table 31).

### **4.2.6 Cross compatibility studies**

Cross compatibility studies were conducted using *Ascda. Sirichi* Fragrance and *Mok. Sayan* × *Ascda. Doung Porn* as male parents. The details on post

**Table 31. Self-compatibility and post pollination changes in the selected parents**

Sl. No.	Selfed parents (♀)	Self-compatibility	Post pollination changes								Remarks
			Flower fall (within a week)	Enlargement of pedicel	Pod Set	Pod set (%)	Days taken for maturity	Seed germination (%)	Days for planting out		
1	<i>Asca</i> . Udomochai	S	Not found	Found	Found	66.87	124	50.26		Protocorms initiation showed declined growth	
2	<i>Asca</i> . Kultana × <i>V. Bitzs</i> Heartthrob	S	Not found	Found	Found	100.0	148	84.4	312		
3	<i>Asca</i> . Sirichi Fragrance	S	Not found	Found	Found	86.48	Not matured fully	-	-	Pods were dried on the plants after two and half months	
4	<i>Vasco</i> . Pine River Blue	S	Not found	Found	Found	33.30	126.2	Not found	-	Seed failed to germinate	
5	<i>Vasco</i> . Pine River Pink	S	Not found	Found	Found	50.00	139.4	87.26	268.3		
6	<i>Vasco</i> . Aroonsri Beauty	Sx	Found	-	-	-	-	-	-	Flower fall was observed within a week	

Table 31. continued

Sl. No.	Selfed parents	Self-compatibility	Post pollination changes							Remarks
			Flower fall (within a week)	Enlargement of pedicel	Pod set	Pod set (%)	Days taken for maturity	Seed germination (%)	Days for planting out	
7	<i>Vasco</i> . Pine Rivers Fuchsia Delight	Sx	Not found	Found	Not Found	-	-	-	-	Pedicel was shrivelled after swelling before pod set
8	<i>Mok</i> . Calypso Pink × <i>Mok</i> . Calypso Pink	Sx	Found	-	Found	-	-	-	-	Flower fall was found within a week
9	<i>Mok</i> . Chao Praya Sunset Yellow Spot	Sx	Not found	Found	Not found	-	-	-	-	Pedicel shrivelled & fallen after swelling
10	<i>Mok</i> . Sayan × <i>Ascada</i> . Doung Porn	S	Not found	Found	Found	64.00	Not matured fully	-	-	Pods were shrivelled & fallen after one and half month of pod setting
11	<i>Kag</i> . Youthong Beauty	Sx	Found	-	-	-	-	-	-	Flower fall was found before swelling of the pedicel

S- Self compatible  
Sx-Self incompatible



pollination (Plate 11-13) and about cross compatibility studies among the parents are presented in Table 32 and 33.

Combinations of *Kag. Youthong Beauty* × *Ascda. Sirichi Fragrance* and *Vasco. Aroonsri Beauty* × *Ascda. Sirichi Fragrance* were found to be cross incompatible, when *Ascda. Sirichi Fragrance* was used as a male parent and the rest of the combinations were attempted were found to be cross compatible with *Ascda. Sirichi Fragrance*.

However, when *Mok. Sayan* × *Ascda. Doung Porn* was used as a male parent all varieties were found to be cross compatible except cross combinations of *Vasco. Pine River Blue* × *Mok. Sayan* × *Ascda. Doung Porn*, *Vasco. Aroonsri Beauty* × *Mok. Sayan* × *Ascda. Doung Porn* and *Kag. Youthong Beauty* × *Mok. Sayan* × *Ascda. Doung Porn*.

#### **4.2.7 Post pollination changes after crossing**

The data pertaining to post pollination changes after the cross pollination (Plate 11-14) with respect to flower fall, enlargement of pedicel, pod development, percentage of pod set, days taken for maturity, percentage of seed germination *in vitro* and days for planting out are given in Table 32 and 33.

##### **4.2.7.1 Enlargement of the pedicel**

Enlargement of the pedicel was not found in the cross *Vasco. Aroonsri Beauty* × *Ascda. Sirichi Fragrance* while in the rest of the varieties pedicel was found to be swollen and enlarged when *Ascda. Sirichi Fragrance* was used as male parent (Table 32 and Plate 12).

However, when *Mok. Sayan* × *Ascda. Doung Porn* used as a male parent, enlargement of pedicel was found in all the selected crosses except for *Vasco. Pine River Blue* × *Mok. Sayan* × *Ascda. Doung Porn* and *Vasco. Aroonsri Beauty* × *Mok. Sayan* × *Ascda. Doung Porn* (Table 33 and Plate 12).



(A) Flower fall



(B) Pods of *Vasco*. Pine River Pink turned pinkish



(C) Pods turned yellow and shrivelled in *Ascda*. Sirichi Fragrance



(D) Swelling of pod in *Mok*. Sayan  
× *Ascda*. Doung Porn



(E) Developed pods shrivelled later in *Vasco*. Aroonsri Beauty



(F) *Mok*. Chao Praya Sunset Yellow Spot



(G) *Ascda*. Kultana × *V.* Bitz's Heartthrob

**Plate 12. Post pollination changes in different hybrids/varieties**

#### **4.2.7.2 Pod development (pod set)**

In the crosses between *Kag. Youthong Beauty* × *Ascda. Sirichi Fragrance*, *Mok. Chao Praya Sunset Yellow Spot* × *Ascda. Sirichi Fragrance* and *Vasco. Aroonsri Beauty* × *Ascda. Sirichi Fragrance* pod setting was not found. Whereas in the rest of the parents, pod setting was found properly (Table 32, Plate 12 and 13).

Pod setting and proper pod development were seen in all the parents crossed with *Mok. Sayan* × *Ascda. Doung Porn* after the enlargement of their pedicels but in the cross between *Kag. Youthong Beauty* × *Mok. Syam Ascda. Doung Porn* pod setting was not found to be developed after the enlargement of pedicel (Table 33, Plate 12 and 13).

#### **4.2.7.3 Percentage of pod set**

Percentage of pod set was found to be highest in cross combinations between *Mok. Calypso Pink* × *Ascda. Sirichi Fragrance* (89.3 %) followed by *Ascda. Kultana* × *V. Bitzs Heartthrob* × *Ascda. Sirichi Fragrance* (80 %), *Vasco. Pine River Pink* × *Ascda. Sirichi Fragrance* (77.28 %), *Ascda. Udomochai* × *Ascda. Sirichi Fragrance* (73.2 %) and *Mok. Sayan* × *Ascda. Doung Porn* × *Ascda. Sirichi Fragrance* (71.33 %) and found to be lowest in crosses with *Vasco. Pine River Blue* × *Ascda. Sirichi Fragrance* (54 %) followed by *Vasco. Pine Rivers Fuchsia Delight* × *Ascda. Sirichi Fragrance* (60.2 %) (Table 32).

In the case of *Mok. Sayan* × *Ascda. Doung Porn* crosses with another male parent pod set percentage was found to be highest in *Ascda. Sirichi Fragrance* × *Mok. Syam Ascda. Doung Porn* (87.9 %) followed by *Ascda. Kultana* × *V. Bitzs Heartthrob* × *Mok. Sayan* × *Ascda. Doung Porn* (86.4 %) and *Mok. Chao Praya Sunset Yellow Spot* × *Mok. Sayan* × *Ascda. Doung Porn* (79.75 %). Whereas, the least percentage of pod set was found in *Mok. Calypso Pink* × *Mok. Sayan* × *Ascda. Doung Porn* (50 %) followed by *Ascda. Udomochai* × *Mok. Sayan* × *Ascda. Doung Porn* (67.33 %) and *Vasco. Pine River Pink* × *Mok. Sayan* × *Ascda. Doung Porn* (67.80 %) (Table 33).



(A) *Vasco*. Pine River Pink



(B) *Mok*. Chao Praya Sunset Yellow Spot



(C) *Vasco*. Pine River Blue



(D) *Asda*. Sirichi Fragrance

**Plate 13. Successfully matured pods in different hybrids**

#### **4.2.7.4 Days taken for maturity**

Minimum number of days was taken for the maturity of pods in *Ascda*. Udomochai × *Ascda*. Sirichi Fragrance (134 days) followed by *Vasco*. Pine River Blue × *Ascda*. Sirichi Fragrance (137 days) and the maximum number of days taken for the maturity of pods in *Ascda*. Kultana × *V. Bitz's Heartthrob* × *Ascda*. Sirichi Fragrance (161 days) and *Vasco*. Pine River Pink × *Ascda*. Sirichi Fragrance (141 days). However, in *Vasco*. Pine Rivers Fuchsia Delight × *Ascda*. Sirichi Fragrance, *Mok*. Sayan × *Ascda*. Doung Porn × *Ascda*. Sirichi Fragrance, and *Mok*. Calypso Pink × *Ascda*. Sirichi Fragrance pods are not found to be matured fully they were shrivelled and dried after 1 to 3 months of pod setting (Table 32 and Plate 14).

In the cross combinations with *Mok*. Sayan × *Ascda*. Doung Porn as male parent found that cross between *Ascda*. Sirichi Fragrance × *Mok*. Sayan × *Ascda*. Doung Porn took 128.8 days for pod maturity while in the rest of the varieties, pods were not found to be matured properly, before the full growth pods started shrivelling and drying up (within 1 to 3 months of pod setting (Table 33).

#### **4.2.7.5 Seed germination (%)**

Highest percentage of seed germination was found to be in *Ascda*. Kultana × *V. Bitz's Heartthrob* × *Ascda*. Sirichi Fragrance (86.33%) followed by *Ascda*. Udomochai × *Ascda*. Sirichi Fragrance (74.33%). The seeds of crosses between the crosses of *Vasco*. Pine River Blue × *Ascda*. Sirichi Fragrance and *Vasco*. Pine River Pink × *Ascda*. Sirichi Fragrance failed to germinate (Table 32 and Plate 14).

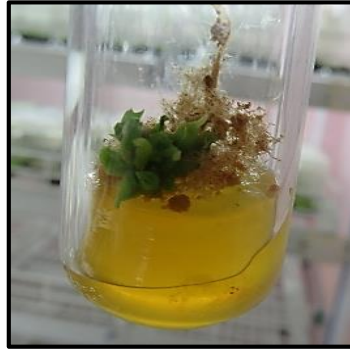
In cross combination of *Ascda*. Sirichi Fragrance × *Mok*. Sayan × *Ascda*. Doung Porn 62.33 per cent successful *in vitro* seed germination was found (Table 33 and Plate 14).



(A) Harvested pod



(B) Seed germination



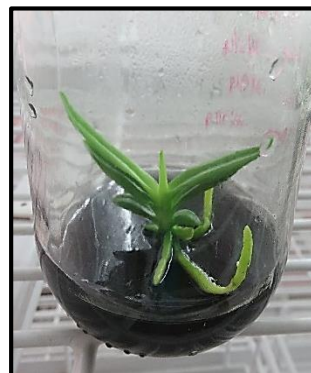
(C) Protocorms showing declined growth



(D) Leaf and root initiation



(E) Multiplication stage



(F) Fully grown seedlings

**Plate 14. Different stages of germination to seedling formation**

**Table 32. Cross compatibility and post pollination changes by using *Ascda*. Sirichi Fragrance as the male parent**

Sl. No.	Crosses made between parents (♂×♀)	Cross compatibility	Post pollination changes							Remarks
			Flower fall (within a week)	Enlargement of pedicel	Pod Set	Pod set (%)	Days taken for maturity	Seed germination (%)	Days for planting out	
1	<i>Ascda</i> . Udomochai × <i>Ascda</i> . Sirichi Fragrance	C	Not found	Found	Found	73.20	134	74.33	246	
2	<i>Ascda</i> . Kultana × V. Bitzs Heartthrob × <i>Ascda</i> . Sirichi Fragrance	C	Not found	Found	Found	80.00	161	86.33	-	Protocorms showed declined growth and dried
3	<i>Vasco</i> . Pine River Blue × <i>Ascda</i> . Sirichi Fragrance	C	Not found	Found	Found	54.00	137	Not found	-	Seed failed to germinate
4	<i>Vasco</i> . Pine River Pink × <i>Ascda</i> . Sirichi Fragrance	C	Not found	Found	Found	77.28	141	Not found		Seed failed to germinate
5	<i>Vasco</i> . Aroonsri Beauty × <i>Ascda</i> . Sirichi Fragrance	Cx	Found	Found	Not found	-	-	-	-	Pedicel shrivelled & fallen after swelling
6	<i>Vasco</i> . Pine Rivers Fuchsia Delight × <i>Ascda</i> . Sirichi Fragrance	C	Not found	Found	Found	60.20	Not matured fully	-	-	Pod set was observed which fallen after one month

**Table 32. continued**

Sl. No.	Crosses made between parents (♂ × ♀)	Cross compatibility	Post pollination changes							
			Flower fall (within a week)	Enlargement of pedicel	Pod set	Pod set (%)	Days taken for maturity	Seed germination (%)	Days for planting out	Remarks
7	<i>Mok. Calypso Pink</i> × <i>Ascda. Sirichi</i> Fragrance	C	Not found	Found	Found	89.30	Not matured fully	-	-	Pod set was observed which fallen after one month
8	<i>Mok. Chao Praya Sunset Yellow Spot</i> × <i>Ascda. Sirichi</i> Fragrance	C	Not found	Found	Not Found	-	-	-	-	Pedicel shrivelled & fallen after swelling
9	<i>Mok. Sayan</i> × <i>Ascda. Doung Porn</i> × <i>Ascda. Sirichi</i> Fragrance	C	Not found	Found	Found	71.33	Not matured fully	-	-	Pod set was observed which fallen after one month
10	<i>Kag. Youthong Beauty</i> × <i>Ascda. Sirichi</i> Fragrance	Cx	Not found	Found	Not Found	-	-	-	-	Pedicel shrivelled & fallen after swelling

**C- Cross compatible**  
**Cx-Cross incompatible**



**Table 33. Cross compatibility and post pollination changes by using *Mok. Sayan* × *Ascida. Doung Porn* as the male parent**

Sl. No.	Crosses made between parents (♂×♀)	Cross compatibility	Post pollination changes							Remarks
			Flower fall (within a week)	Enlargement of pedicel	Pod set	Pod set (%)	Days taken for maturity	Seed germination (%)	Days for planting out	
1	<i>Ascida. Udomochai</i> × <i>Mok. Sayan</i> × <i>Ascida. Doung Porn</i>	C	Not found	Found	Found	67.33	Not matured	-	-	Pods were shrivelled and dried in two weeks
2	<i>Ascida. Kultana</i> × <i>V. Bitz</i> Heartthrob × <i>Mok. Sayan</i> × <i>Ascida. Doung Porn</i>	C	Not found	Found	Found	86.4	Not matured properly	-	-	Pods were shrivelled after two months
3	<i>Ascida. Sirichi</i> Fragrance × <i>Mok. Sayan</i> × <i>Ascida. Doung Porn</i>	C	Not found	Found	Found	87.9	128.8	-	62.33	Seeds germinate but protocorms showed declined growth
4	<i>Vasco. Pine River Blue</i> × <i>Mok. Sayan</i> × <i>Ascida. Doung Porn</i>	Cx	Found	-	-	-	-	-	-	Flower fall was found within a week
5	<i>Vasco. Pine River Pink</i> × <i>Mok. Sayan</i> × <i>Ascida. Doung Porn</i>	C	Not found	Found	Found	67.8	Not matured fully	-	-	Pods were dried and fallen after a month
6	<i>Vasco. Aroonsri Beauty</i> × <i>Mok. Sayan</i> × <i>Ascida. Doung Porn</i>	Cx	Found	-	-	-	-	-	-	Flower fall was found within a week

Table 33. continued

Sl. No.	Crosses made between parents (♂×♀)	Cross compatibility	Post pollination changes							Remarks
			Flower fall (within a week)	Enlargement of pedicel	Pod set	Pod set (%)	Days taken for maturity	Seed germination (%)	Days for planting out	
7	<i>Vasco</i> . Pine Rivers Fuchsia Delight × <i>Mok</i> . Sayan × <i>Ascda</i> . Doung Porn	C	Not found	Found	Found	-	-	-	-	Pod were dried in and shrivelled after three months of pod setting
8	<i>Mok</i> . Calypso Pink × <i>Mok</i> . Sayan × <i>Ascda</i> . Doung Porn	C	Not found	Found	Found	50	-	-	-	Pod were dried in and shrivelled after two months of pod setting.
9	<i>Mok</i> . Chao Praya Sunset Yellow Spot × <i>Mok</i> . Sayan × <i>Ascda</i> . Doung Porn	C	Not found	Found	Found	79.75	Not matured properly	-	-	Pod were dried in and fallen after a month of pod setting.
10	<i>Kag</i> . Youthong Beauty × <i>Mok</i> . Sayan × <i>Ascda</i> . Doung Porn	Cx	Not found	Found	Not found	-	-	-	-	Pedicel shrivelled and fallen after swelling in two weeks before pod set

C- Cross compatible  
Cx-Cross incompatible

#### 4.2.7.6 Days for planting out

The cross between *Ascda. Udomochai* × *Ascda. Sirichi* Fragrance was taken 246 days for deflasking/planting out after inoculation while in *Ascda. Kultana* × *V. Bitz's Heartthrob* × *Ascda. Sirichi* Fragrance protocorms initiation showed declined growth after *in vitro* seed germination (Table 32).

In *Ascda. Sirichi* Fragrance × *Mok. Sayan* × *Ascda. Doung Porn* protocorms initiation showed declined growth after the *in vitro* seed germination (Table 33).

#### 4.2.8 Relationship between Self and cross compatibility studies

The relationship matrix of self and cross compatibility studies between selected eleven parents is presented in Table 34.

The varieties *Ascda. Udomochai*, *Ascda. Kultana* × *Vanda Bitz's Heartthrob* and *Vasco. Pine River Pink* were found to be self compatible and cross compatible with both male parents.

The male parents *Ascda. Sirichi* Fragrance and *Mok. Sayan* × *Ascda. Doung Porn* were also found to be self compatible and cross compatible with each other (Table 34).

The variety *Vasco. Pine River Blue* was also found to be self compatible and cross compatible with *Ascda. Sirichi* Fragrance but was found cross incompatible with *Mok. Sayan* × *Ascda. Doung Porn* male parent.

Varieties *Kag. Youthong Beauty* and *Vasco. Aroonsri Beauty* was found to be self incompatible and cross incompatible with both of the male parents. However, rest of the varieties *viz.*, *Vasco. Pine Rivers Fuchsia Delight*, *Mok. Calypso Pink* and *Mok. Chao Praya Sunset Yellow Spot* are found to be self incompatible but cross compatible with both parents (Table 34).

**Table 34. Relationship matrix of compatibility studies**

Sl. No.	Hybrid/ variety used as female parent (♀)	Self-compatibility	Cross compatibility by male parents	
			<i>Ascda.</i> Sirichi Fragrance (♂)	<i>Mok.</i> Sayan × <i>Ascda.</i> Doung Porn (♂)
1	<i>Ascda.</i> Udomochai	S	C	C
2	<i>Ascda.</i> Kultana × <i>V.</i> Bitzs Heartthrob	S	C	C
3	<i>Ascda.</i> Sirichi Fragrance	S	-	C
4	<i>Vasco.</i> Pine River Blue	S	C	C <sub>X</sub>
5	<i>Vasco.</i> Pine River Pink	S	C	C
6	<i>Vasco.</i> Aroonsri Beauty	S <sub>X</sub>	C <sub>X</sub>	C <sub>X</sub>
7	<i>Vasco.</i> Pine Rivers Fuchsia Delight	S <sub>X</sub>	C	C
8	<i>Mok.</i> Calypso Pink	S <sub>X</sub>	C	C
9	<i>Mok.</i> Chao Praya Sunset Yellow Spot	S <sub>X</sub>	C	C
10	<i>Mok.</i> Sayan × <i>Ascda.</i> Doung Porn	S	C	-
11	<i>Kag.</i> Youthong Beauty	S <sub>X</sub>	C <sub>X</sub>	C <sub>X</sub>

**S- Self-compatible****S<sub>X</sub>-Self-incompatible****C- Cross compatible****C<sub>X</sub>-Cross incompatible**

### 4.3 MOLECULAR CHARACTERISATION

Twenty intergeneric hybrids of *Ascocentrum* were selected for molecular characterisation. It was done in was done using microsatellite markers (SSR) and ISSR. In addition to morphological characterisation. Microsatellite markers are fast, reliable and reproducible system for molecular genotyping.

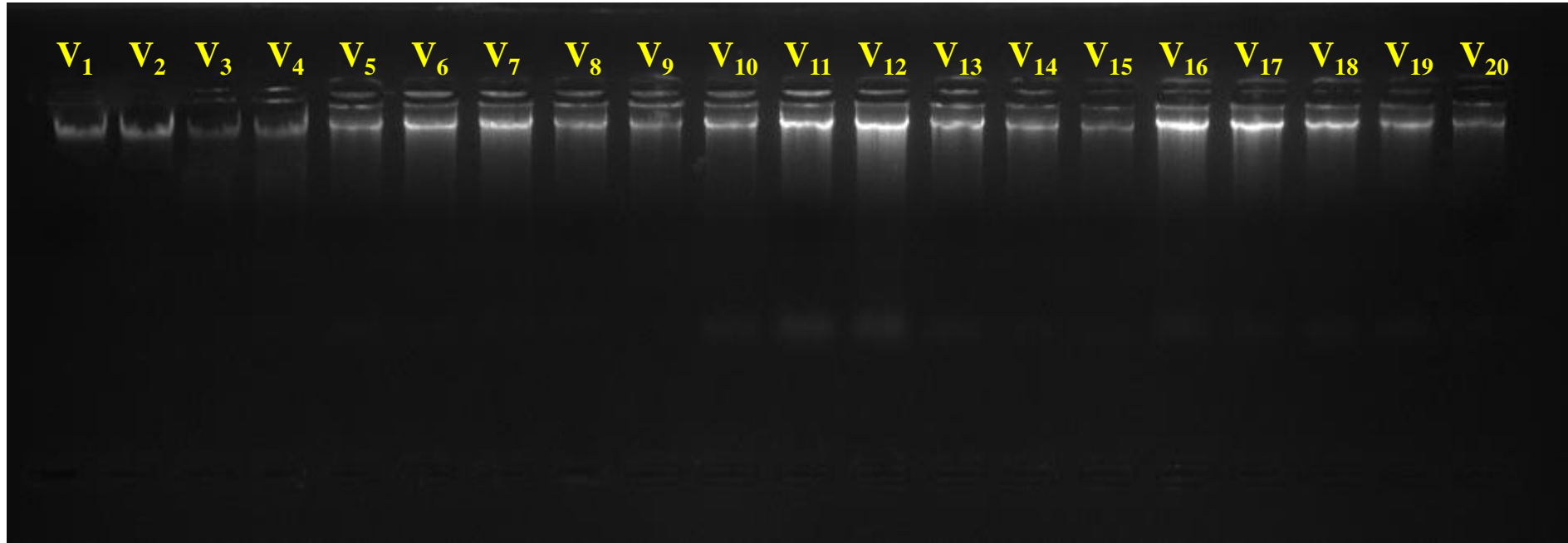
#### 4.3.1 Quality and quantity of DNA isolated

The isolation of high quality DNA is important for all molecular analyses, because contaminants (proteins, polyphenols, *etc.*) can interfere with the

end result. Genomic DNA was isolated from young tender leaves of each variety using protocol suggested by Lim *et al.* (1998). The isolated DNA from all samples (Plate 15) were checked for its quality and quantity using Nanodrop spectrophotometer (Jenway- Genova Nano). The extracted DNA was confirmed to be of good quality through Agarose gel electrophoresis as well as computation of the OD value (ratio of absorbance at 260 nm and 280 nm) and quantity ( $\mu\text{g/ml}$ ) given by Nanodrop (Table 35). The quality and the quantity of isolated DNA from all hybrids were well within the accepted level to carry out further SSR and ISSR analysis.

**Table 35. Analysis of DNA determined by Nanodrop spectrophotometer in different varieties**

Sr. No.	Var. No.	Hybrid/ variety name	OD value (260/280)	Quantity ( $\mu\text{g}/\mu\text{l}$ )
1	V <sub>1</sub>	<i>Kag.</i> Christie Low	2.00	337.32
2	V <sub>2</sub>	<i>Mok.</i> Omayaiy Yellow	2.09	949.01
3	V <sub>3</sub>	<i>Mok.</i> Sunspot	1.98	509.09
4	V <sub>4</sub>	<i>Vasco.</i> Pine River Pink	2.01	1058.00
5	V <sub>5</sub>	<i>Ascda.</i> Sirichi Fragrance	1.83	105.65
6	V <sub>6</sub>	<i>Ascda.</i> Udomochai	1.90	454.37
7	V <sub>7</sub>	<i>Vasco.</i> Blue Bay White	1.87	285.96
8	V <sub>8</sub>	<i>Vasco.</i> Pine Rivers Fuchsia Delight	1.99	260.00
9	V <sub>9</sub>	<i>Ascda.</i> Kultana $\times$ <i>Vanda</i> Bitzs Heartthrob	1.86	355.75
10	V <sub>10</sub>	<i>Kag.</i> Samrong	1.91	331.00
11	V <sub>11</sub>	<i>Mok.</i> Sayan $\times$ <i>Ascda.</i> Bangkuntein Gold	1.97	105.56
12	V <sub>12</sub>	<i>Mok.</i> Khaw Phiak Suan $\times$ <i>Ascda.</i> Jiraprapra	1.91	210.66
13	V <sub>13</sub>	<i>Ascda.</i> Suksamran Sunlight	1.98	220.62
14	V <sub>14</sub>	<i>Ascda.</i> Yip Sum Wah $\times$ <i>V.</i> JVB	1.96	325.54
15	V <sub>15</sub>	<i>Vasco.</i> Aroonsri Beauty	1.92	338.41
16	V <sub>16</sub>	<i>Mok.</i> Walter Oumae Pink	1.90	190.06
17	V <sub>17</sub>	<i>Mok.</i> Rassmatozz	1.82	212.10
18	V <sub>18</sub>	<i>Mok.</i> Calypso $\times$ <i>V.</i> Dr. Anek	1.91	201.00
19	V <sub>19</sub>	<i>Mok.</i> Khaw Phiak Suan $\times$ <i>Ascda.</i> Bicentennial Yellow Spot	1.91	238.92
20	V <sub>20</sub>	<i>Mok.</i> Chao Praya Sunset Yellow Spot	1.99	195.01



**Plate 15. Agarose gel electrophoresis of isolated DNA with selected 20 intergeneric hybrids**

### **4.3.2 Genotyping with molecular markers**

Assessment of genetic diversity is a prerequisite for genetic improvement of any agricultural/horticultural crops. Knowledge of genetic diversity within the available germplasm is a key for the successful breeding programme in crop improvement. The working solution of DNA (50 µg/µl) was prepared by diluting DNA with ultra-pure type 1 water before subjecting it to PCR reaction. The DNA sample of 20 hybrids/varieties were used for SSR and ISSR analysis.

A total of 21 SSR and 29 ISSR primers were used to assess the genetic diversity and estimate genetic polymorphism in 20 hybrids. Out of 21 SSR primers used, 10 primers produced polymorphic patterns in at least two hybrids (Table 36 and Plate 16-20).

Out of 29 ISSRs primer used, only 20 showed amplification in all the hybrids with polymorphic bands (Table 39 and Plate 21-30). Data of these polymorphic primers were then used to study the molecular divergence among all hybrids and the analysis of this data revealed a high level of diversity among all genotypes.

#### **4.3.2.1 Genotyping with SSR markers**

Results of this study confirmed the existence of a high genetic diversity among 20 hybrids/varieties. Out of 21 SSR markers used in the study (Table 6), only 10 produced polymorphic bands (Plate 16-20). The number of amplicons detected varied from 2 to 7. The highest number of alleles was found in the FJ539054 (7), FJ539061 (5) and JN375718 (5). Primers DQ501383, DQ501384, DQ501387 and FJ539057 had amplified 4 amplicons. Primers DQ494847 (3) observed to have less number of amplicons. One unique band each was produced by JN375713, FJ539050 V<sub>10</sub> and V<sub>15</sub>, respectively (Table 36).

To identify informative markers for cultivar identification, Polymorphic Information Content (PIC) value was calculated. The high PIC of primers indicates the highly informative nature of the SSR primers and the diversity of the

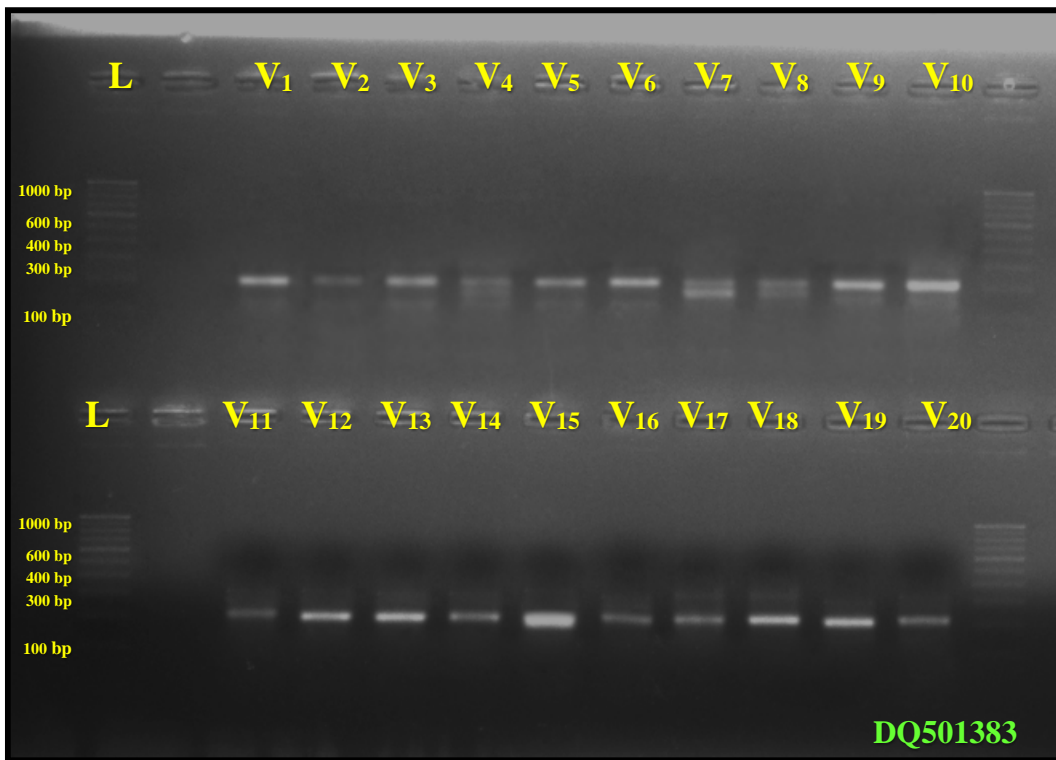
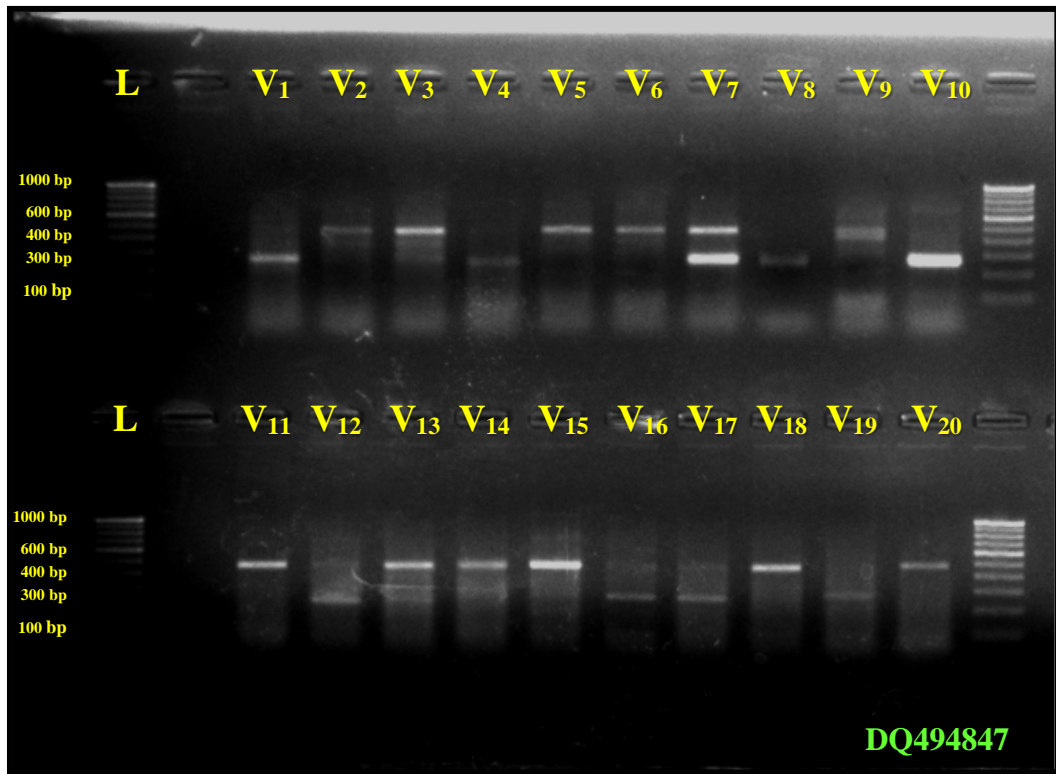


Plate 16. Amplification pattern generated by DQ494847 and DQ501383 with 20 hybrids of *Ascocentrum*



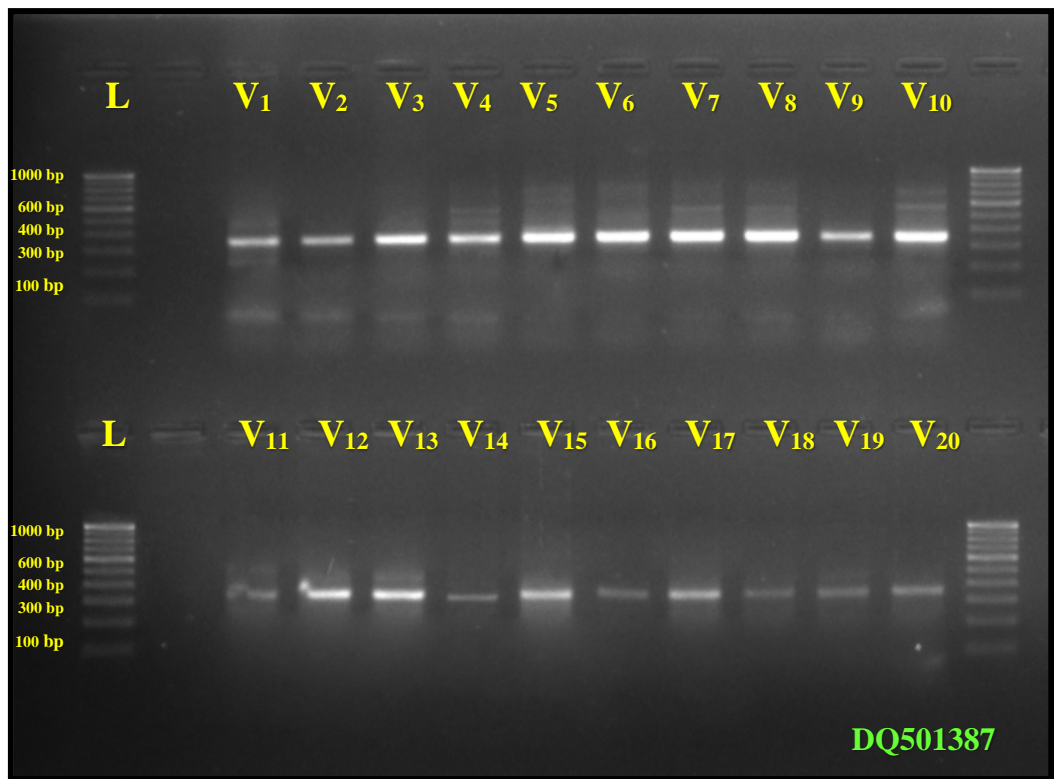
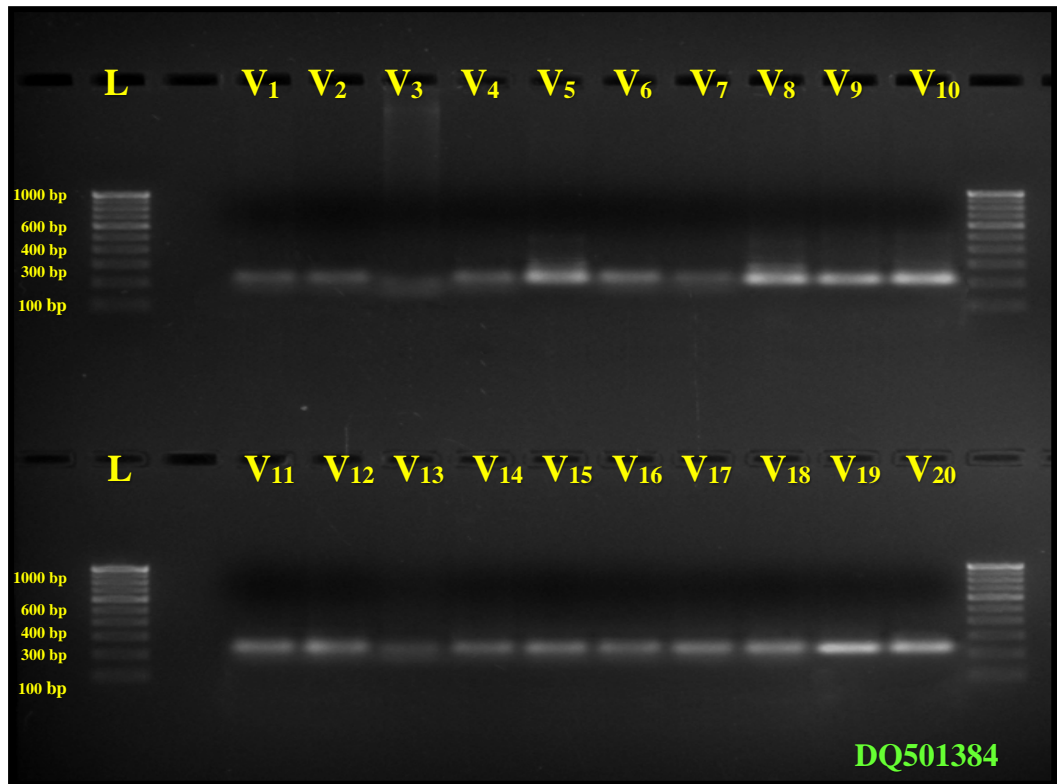
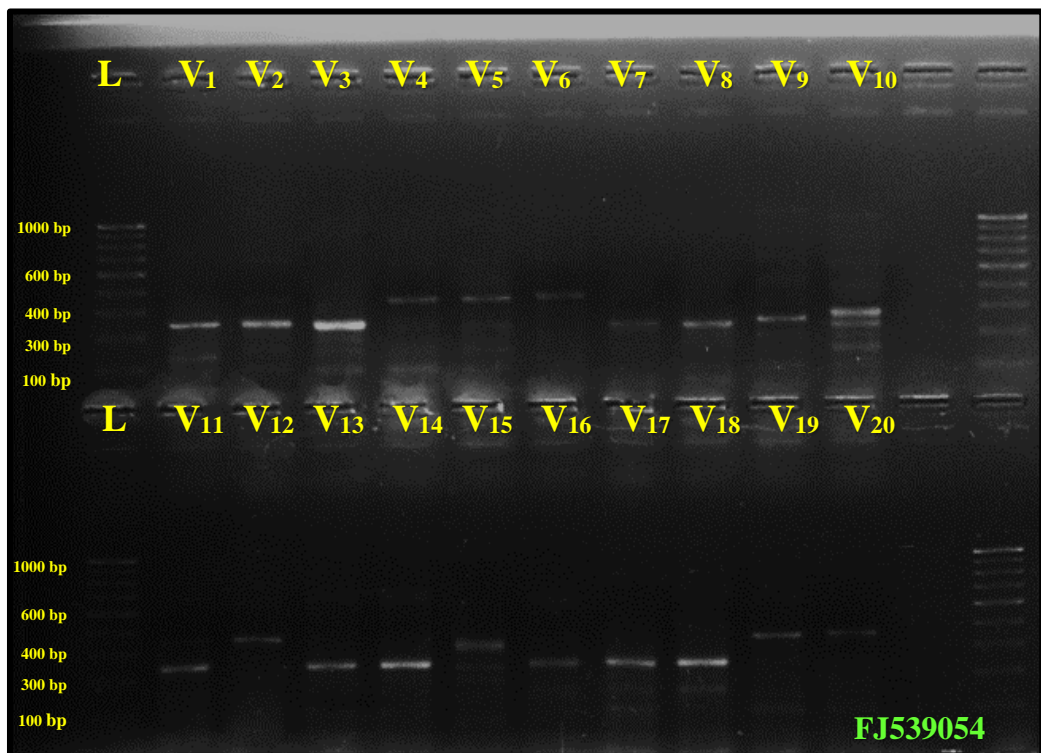
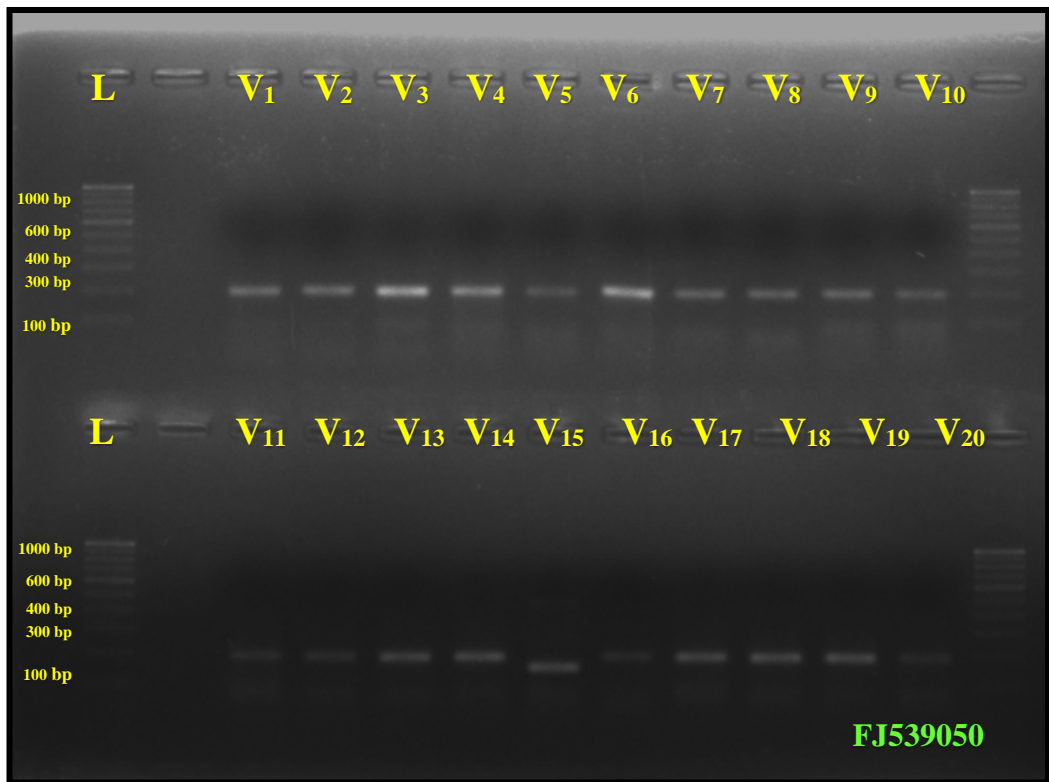


Plate 17. Amplification pattern generated by DQ501384 and DQ501387 with 20 hybrids of *Ascocentrum*



**Plate 18. Amplification pattern generated by FJ539050 and FJ539054 with 20 hybrids of *Ascocentrum***

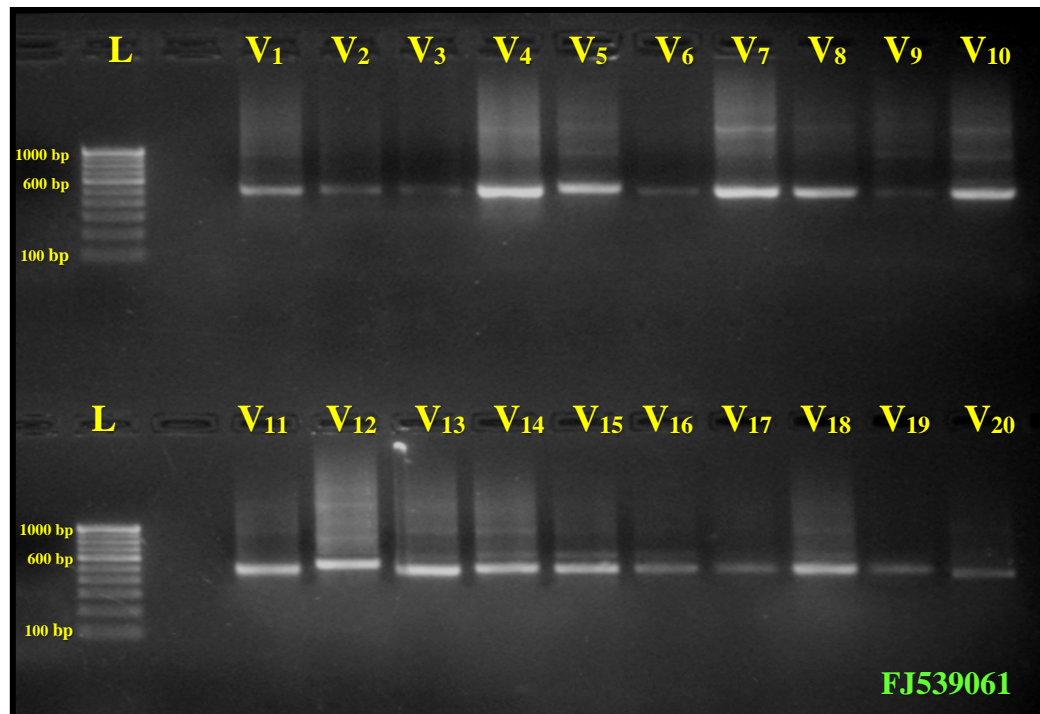
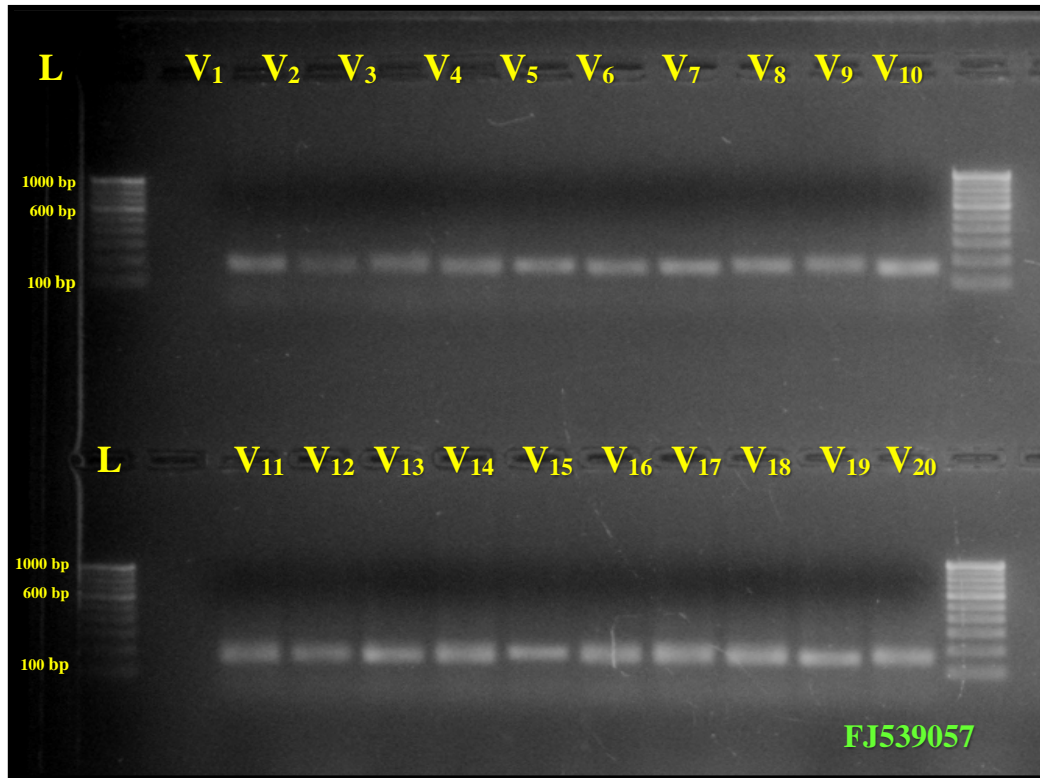


Plate 19. Amplification pattern generated by FJ539057 and FJ539061 with 20 hybrids of *Ascocentrum*

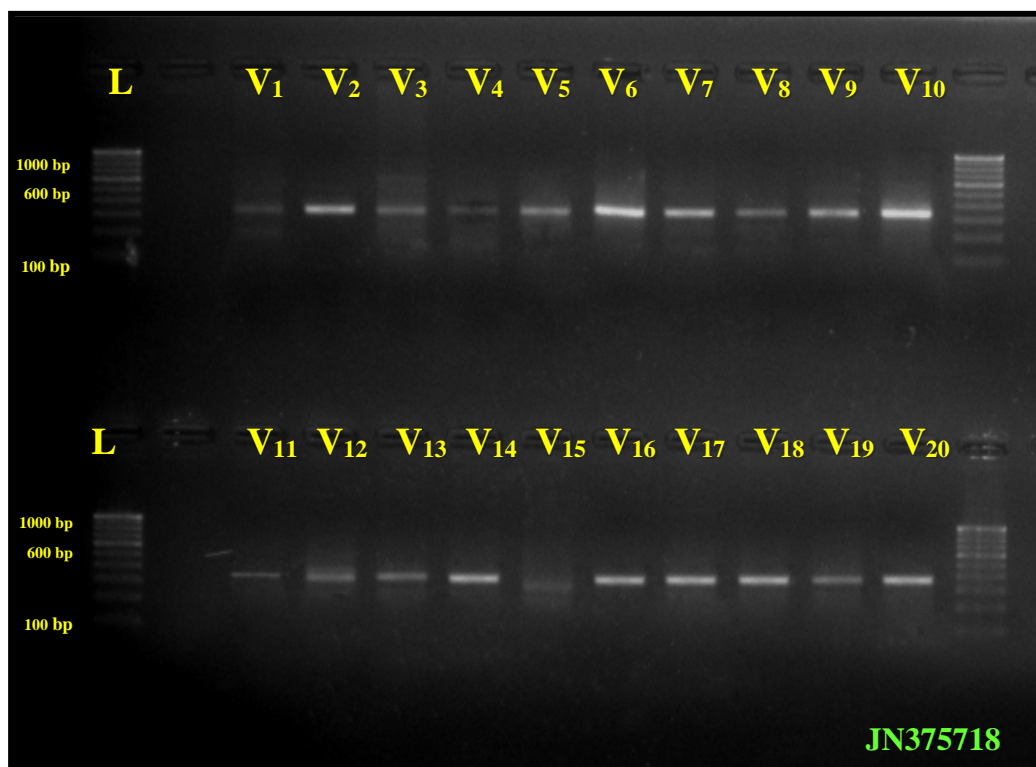
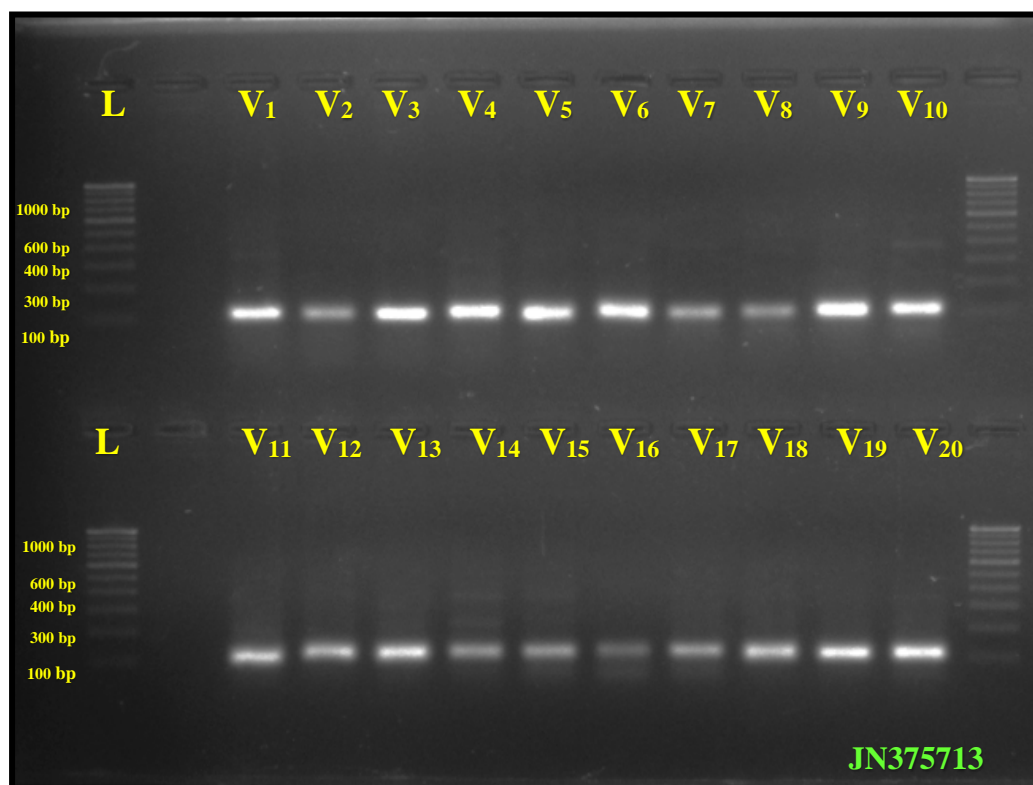


Plate 20. Amplification pattern generated by JN375713 and JN375718 with 20 hybrids of *Ascocentrum*

used populations. The PIC value ranged from 0.095 to 0.800. Primers FJ539050 and JN375713 observed to have lowest PIC value (0.095), whereas, FJ539054 recorded the highest PIC value (0.800). Primers DQ501387 (0.525), JN375718 (0.570), DQ501383 (0.591), DQ494847 (0.612), DQ501384 (0.665), FJ539061 (0.669) and FJ539057 (0.705) also produced good amount of polymorphism.

**Table 36. Particulars of polymorphic SSR markers**

Sl. No.	Primer	AT (°C)	No. of amplicons	Amplicon size (bp)		PIC
				Min.	Max.	
1	DQ494847	48.3	3	256.43	500	0.612
2	DQ501383	57.8	4	135.51	229.2	0.591
3	DQ501384	55.35	4	183.23	223.89	0.665
4	DQ501387	59.15	4	309.75	376.18	0.525
5	FJ539050	56.05	2	187.82	200	0.095
6	FJ539054	60.2	7	343.04	508.15	0.800
7	FJ539057	60.65	4	132.49	192.47	0.705
8	FJ539061	60.45	5	389.72	585.36	0.669
9	JN375713	58.9	2	116.63	133.8	0.095
10	JN375718	57.4	5	262.59	370.44	0.570

Based on PIC values, SSR primers FJ539054 and FJ539057 can be used to distinguish the hybrids/varieties, whereas, JN375713 and FJ539050 can help in establishing the uniqueness of varieties V<sub>10</sub> and V<sub>15</sub>, respectively (Table 36).

#### **4.3.2.2 Cluster analysis and dendrogram construction using SSR data**

Using Jaccard's similarity coefficient for SSR primers, genetic similarity was calculated for all 20 hybrids/varieties of orchids (Table 37). The highest Jaccard's similarity value (0.82) was observed between V<sub>1</sub> and V<sub>4</sub>, whereas, the least Jaccard's similarity value (0.05) was observed in between V<sub>10</sub> and V<sub>13</sub>, V<sub>14</sub>, V<sub>15</sub>, which indicates that these hybrids/varieties are dissimilar to each other.

**Table 37. Pair wise similarity between varieties based on SSR data**

Var. No.	V <sub>1</sub>	V <sub>2</sub>	V <sub>3</sub>	V <sub>4</sub>	V <sub>5</sub>	V <sub>6</sub>	V <sub>7</sub>	V <sub>8</sub>	V <sub>9</sub>	V <sub>10</sub>	V <sub>11</sub>	V <sub>12</sub>	V <sub>13</sub>	V <sub>14</sub>	V <sub>15</sub>	V <sub>16</sub>	V <sub>17</sub>	V <sub>18</sub>	V <sub>19</sub>	V <sub>20</sub>	
V <sub>1</sub>	1.00																				
V <sub>2</sub>	0.67	1.00																			
V <sub>3</sub>	0.54	0.54	1.00																		
V <sub>4</sub>	0.82	0.54	0.43	1.00																	
V <sub>5</sub>	0.54	0.54	0.54	0.67	1.00																
V <sub>6</sub>	0.33	0.54	0.33	0.33	0.54	1.00															
V <sub>7</sub>	0.47	0.38	0.38	0.38	0.38	0.29	1.00														
V <sub>8</sub>	0.50	0.31	0.40	0.40	0.24	0.17	0.44	1.00													
V <sub>9</sub>	0.25	0.18	0.25	0.25	0.25	0.18	0.29	0.40	1.00												
V <sub>10</sub>	0.25	0.18	0.11	0.25	0.11	0.11	0.16	0.40	0.25	1.00											
V <sub>11</sub>	0.33	0.25	0.43	0.25	0.25	0.11	0.29	0.31	0.33	0.05	1.00										
V <sub>12</sub>	0.33	0.18	0.33	0.33	0.25	0.18	0.29	0.31	0.25	0.11	0.43	1.00									
V <sub>13</sub>	0.25	0.43	0.43	0.25	0.33	0.33	0.22	0.17	0.18	0.05	0.33	0.43	1.00								
V <sub>14</sub>	0.24	0.24	0.31	0.24	0.31	0.24	0.35	0.16	0.31	0.05	0.24	0.31	0.50	1.00							
V <sub>15</sub>	0.31	0.31	0.40	0.40	0.50	0.24	0.28	0.16	0.31	0.05	0.24	0.24	0.31	0.47	1.00						
V <sub>16</sub>	0.18	0.11	0.18	0.18	0.11	0.11	0.22	0.40	0.33	0.25	0.25	0.43	0.43	0.31	0.11	1.00					
V <sub>17</sub>	0.33	0.25	0.33	0.33	0.25	0.18	0.22	0.24	0.18	0.11	0.33	0.54	0.67	0.40	0.24	0.67	1.00				
V <sub>18</sub>	0.33	0.43	0.43	0.33	0.33	0.25	0.16	0.24	0.11	0.11	0.25	0.33	0.67	0.40	0.24	0.43	0.67	1.00			
V <sub>19</sub>	0.33	0.25	0.25	0.33	0.33	0.25	0.16	0.17	0.11	0.18	0.18	0.33	0.33	0.24	0.17	0.33	0.54	0.43	1.00		
V <sub>20</sub>	0.25	0.25	0.25	0.25	0.33	0.25	0.16	0.24	0.33	0.25	0.25	0.18	0.18	0.11	0.17	0.25	0.25	0.25	0.54	1.00	

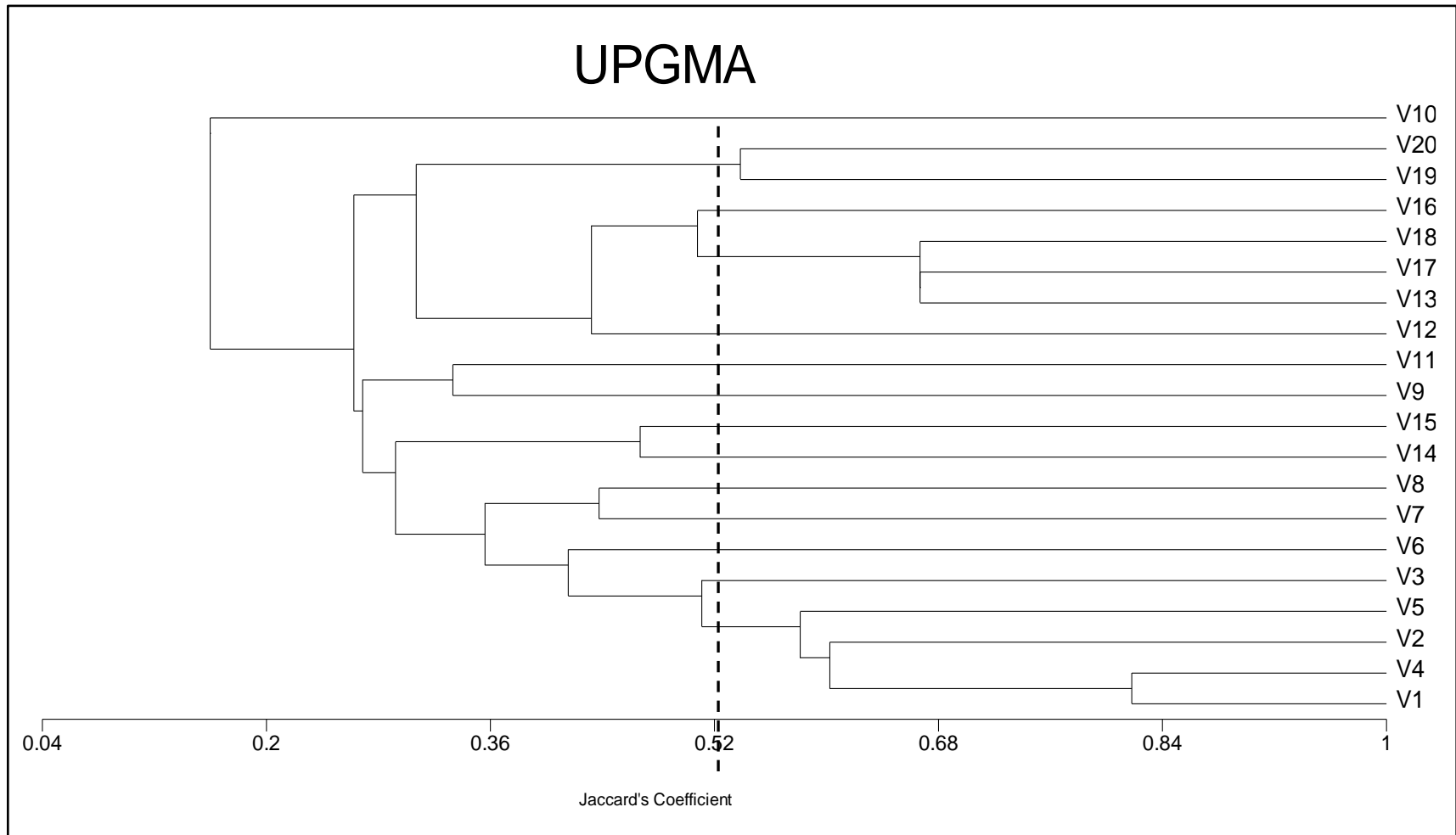
**Table 38. Clustering based on SSR scoring**

Sl. No.	Cluster	No. of members	Members of cluster
1	Cluster I	5	V <sub>1</sub> , V <sub>2</sub> , V <sub>3</sub> , V <sub>4</sub> , V <sub>5</sub>
2	Cluster II	1	V <sub>6</sub>
3	Cluster III	1	V <sub>7</sub>
4	Cluster IV	1	V <sub>8</sub>
5	Cluster V	1	V <sub>9</sub>
6	Cluster VI	1	V <sub>11</sub>
7	Cluster VII	1	V <sub>12</sub>
8	Cluster VIII	3	V <sub>13</sub> , V <sub>17</sub> , V <sub>18</sub>
9	Cluster IX	1	V <sub>14</sub>
10	Cluster X	1	V <sub>15</sub>
11	Cluster XI	1	V <sub>16</sub>
12	Cluster XII	2	V <sub>19</sub> , V <sub>20</sub>
13	Cluster XIII	1	V <sub>10</sub>

The UPGMA clustering algorithm grouped the varieties into 2 main clusters according to dendrogram. The variety V<sub>10</sub> clustered separately from all other members, whereas, other members formed in another cluster. At 50 per cent level of similarity, the hybrids/varieties clustered into 13 cluster (Table 38 and Fig. 16) Cluster I was the biggest cluster with five members (V<sub>1</sub>, V<sub>2</sub>, V<sub>3</sub>, V<sub>4</sub>, V<sub>5</sub>), followed by Cluster VIII with three members (V<sub>13</sub>, V<sub>17</sub>, V<sub>18</sub>) and Cluster XII with two varieties (V<sub>19</sub>, V<sub>20</sub>). Whereas, all remaining members grouped each into separate cluster.

#### ***4.3.2.3 Genotyping with ISSR markers***

To characterise and measure the extent of variation between the 20 intergeneric hybrids, DNA of each orchid hybrids were subjected to PCR amplification using 29 ISSR primers mentioned earlier in Table 7. Out of 29 ISSRs primer used, only 20 showed amplification in all the hybrids with polymorphic



**Fig. 16. Dendrogram showing clustering of 20 hybrids/varieties based on SSR profile**



bands (Table 39 and Plate 21-30). The details of primers and amplification is indicated.

The total number of amplicons detected by an individual primer ranged from 11 to 18. ISSR primer (GACAC)<sub>4</sub> generated 11 amplicons with 20 hybrids/varieties, whereas ISSR 901 amplified 31 amplicons. Four ISSR primers *viz.*, DAT (25), MAO (29), UBC808 (30) and UBC858 (29) observed with more than 25 amplicons. Six ISSR primers *viz.*, 17899A (18), ACTG(4) (18), GOOFY (17), UBC809 (16), UBC810 (17) and UBC841 (15) amplified less than 25 amplicons. The amplicon size ranged from 126 bp to 2487 bp. Primer ISSR 7 observed with the least size of amplicon (126 bp-1780 bp). Whereas, ISSR primer OMAR amplified amplicon of size in between 155 bp- 2487 bp (Table 39).

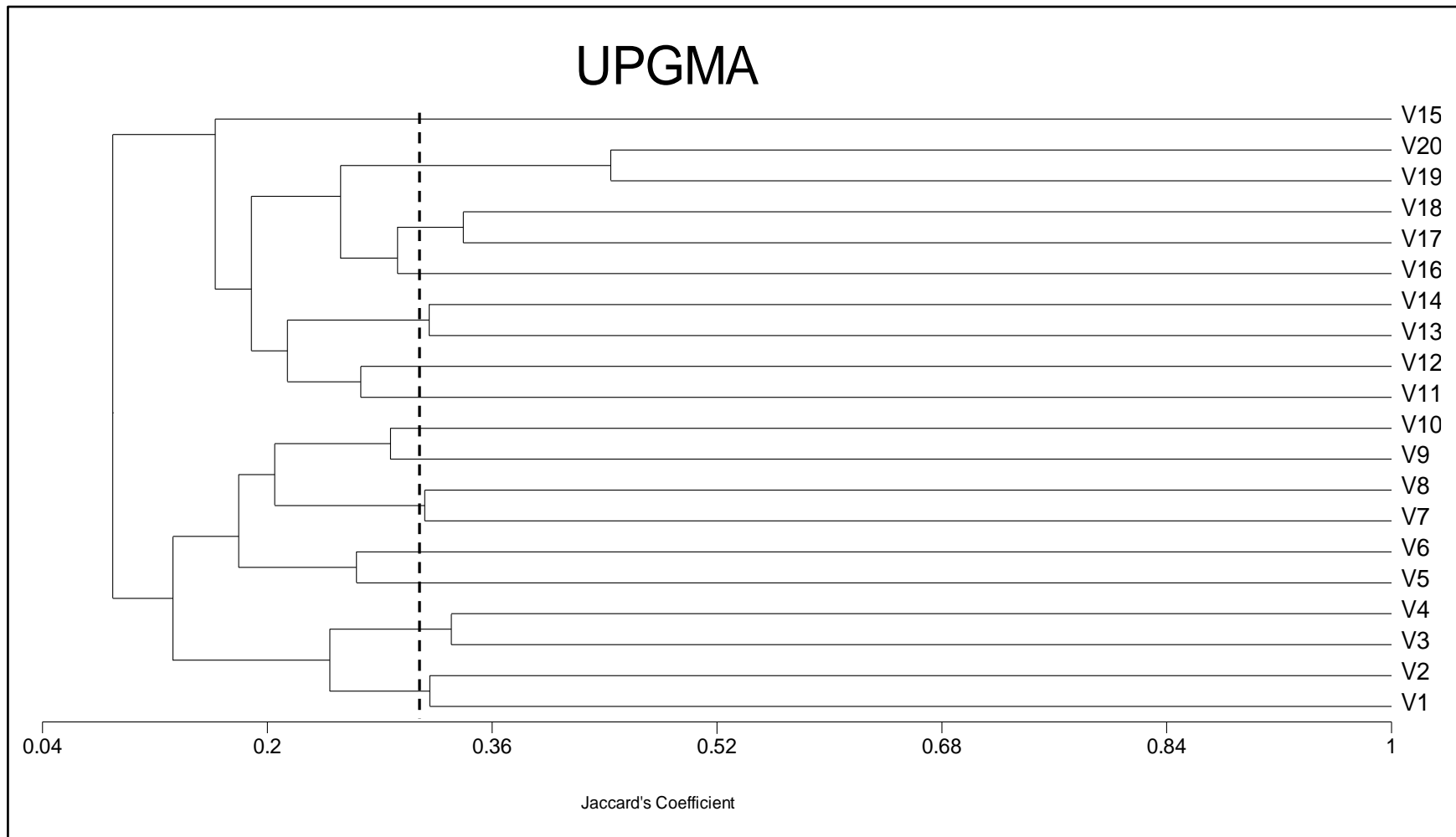
ISSR primer (GACAC)<sub>4</sub> observed with low PIC value, 0.594, whereas UBC810 observed with the highest PIC value, 0.926. It was also observed that 17 out of 20 ISSR markers recorded PIC value more than 0.8 (Table 39). This indicated high discriminatory and differentiation power of these markers for the population of orchids used in this study.

#### ***4.3.2.4 Cluster analysis and dendrogram construction using ISSR data***

Cluster analysis revealed the presence of high genetic variation among all varieties studied. The dendrogram based on Jaccard's similarity coefficients was constructed using UPGMA after analysis of banding patterns generated by 20 polymorphic primers across the 20 hybrids/varieties of orchids (Fig. 17).

The highest Jaccard's similarity value (0.44) was observed between V<sub>19</sub> and V<sub>20</sub>. Whereas, the least Jaccard's similarity value (0.03) was observed in between V<sub>7</sub> and V<sub>19</sub>, which indicates that these hybrids/varieties are dissimilar to each other (Table 40.)

A UPGMA-based dendrogram separated the 20 hybrids of orchids into two main clusters each with 10 members. It was also observed that the ISSR primers



**Fig. 17. Dendrogram showing clustering of 20 hybrids/varieties based on ISSR profile**

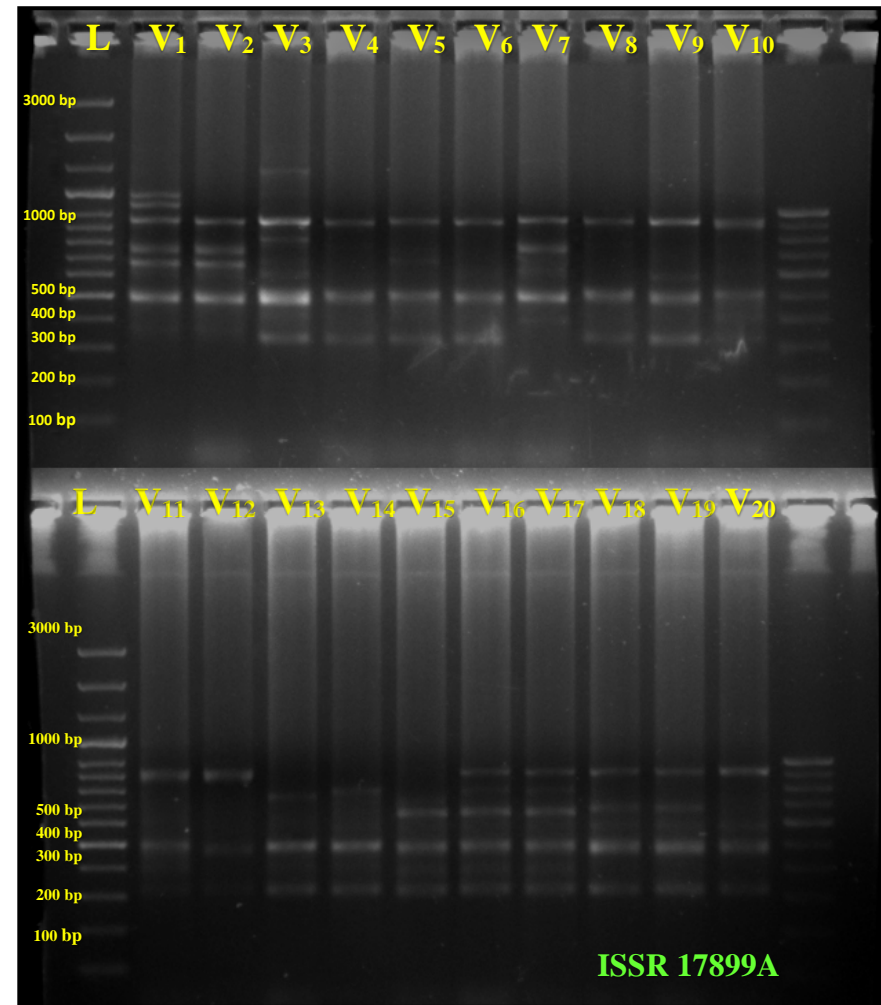
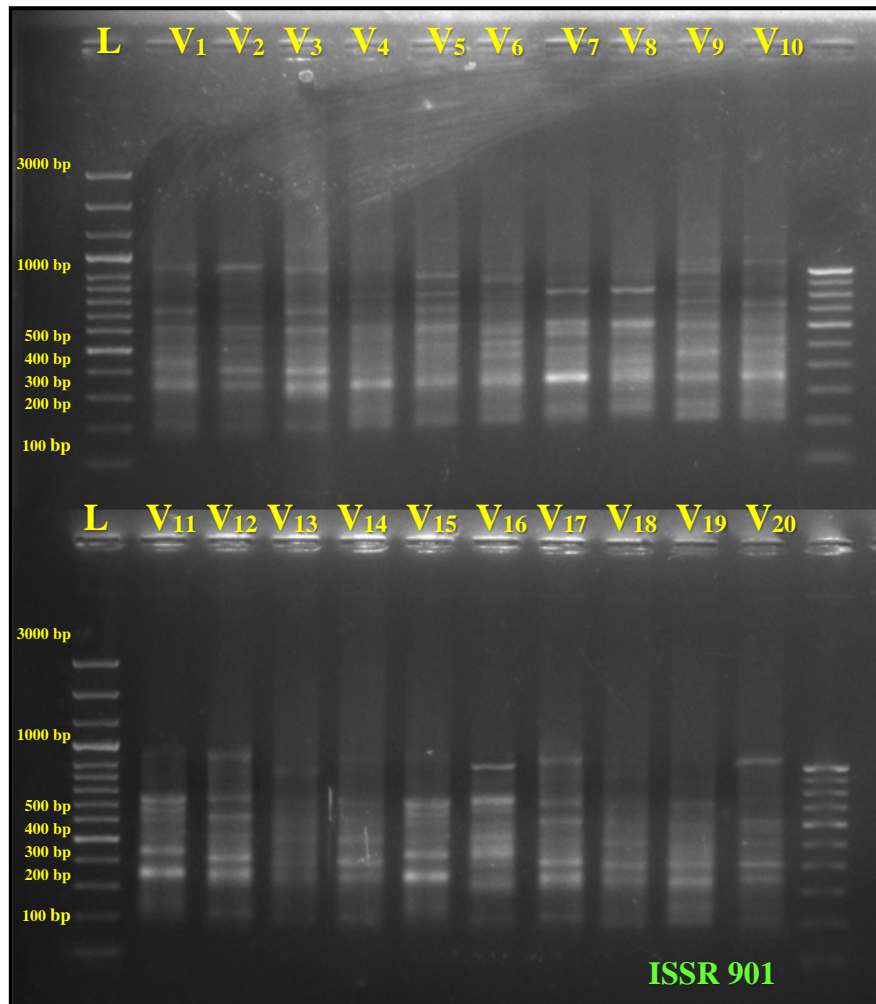
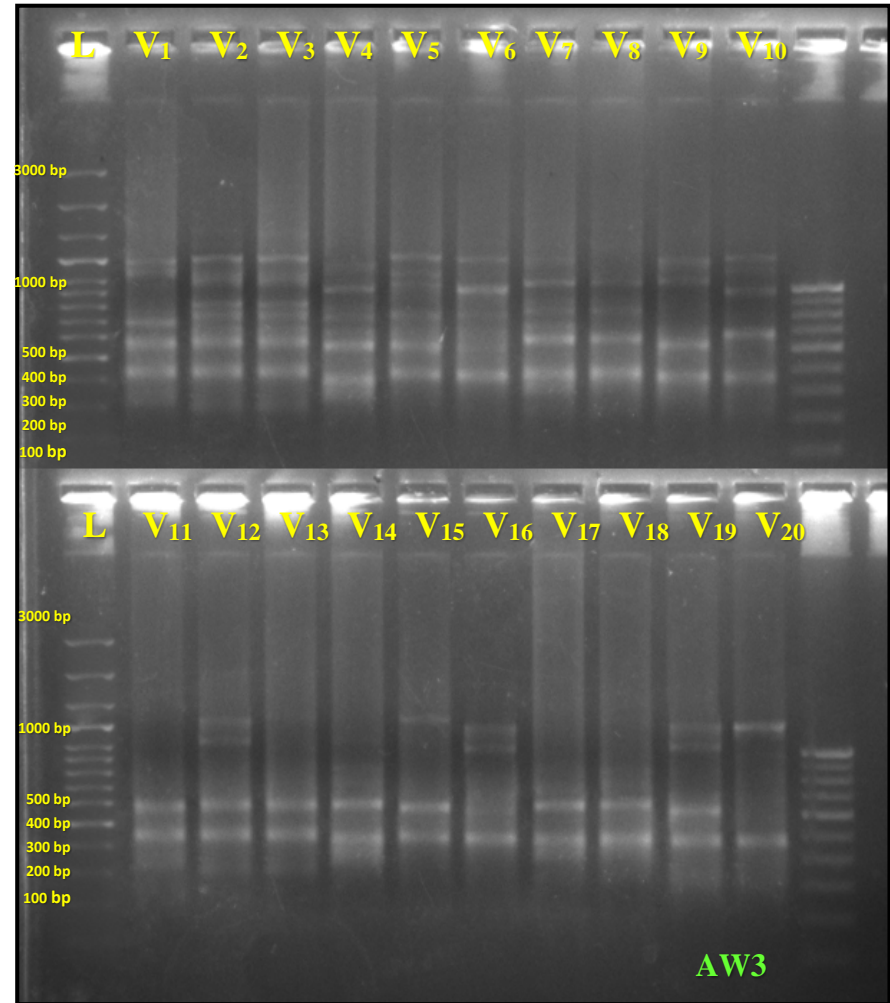
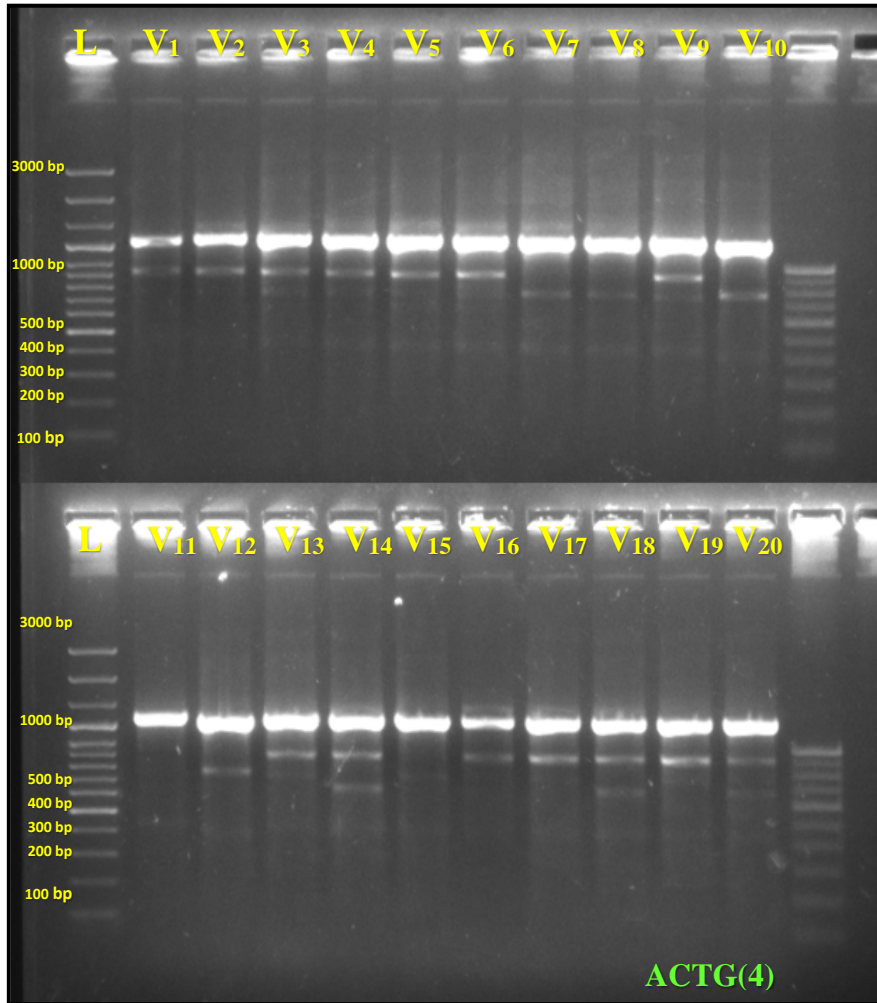


Plate 21. Amplification pattern generated by ISSR 901 and ISSR 17899A with 20 hybrids of *Ascocentrum*



**Plate 22. Amplification pattern generated by ACTG(4) and AW3 with 20 hybrids of *Ascocentrum***

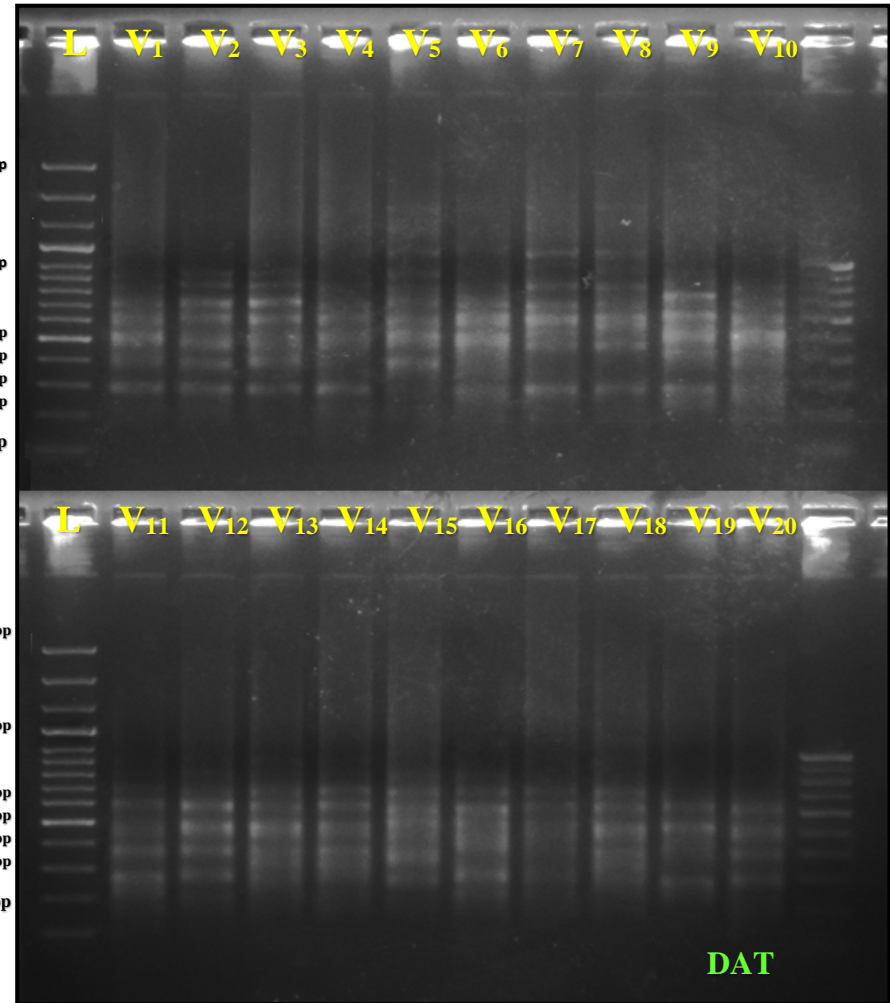
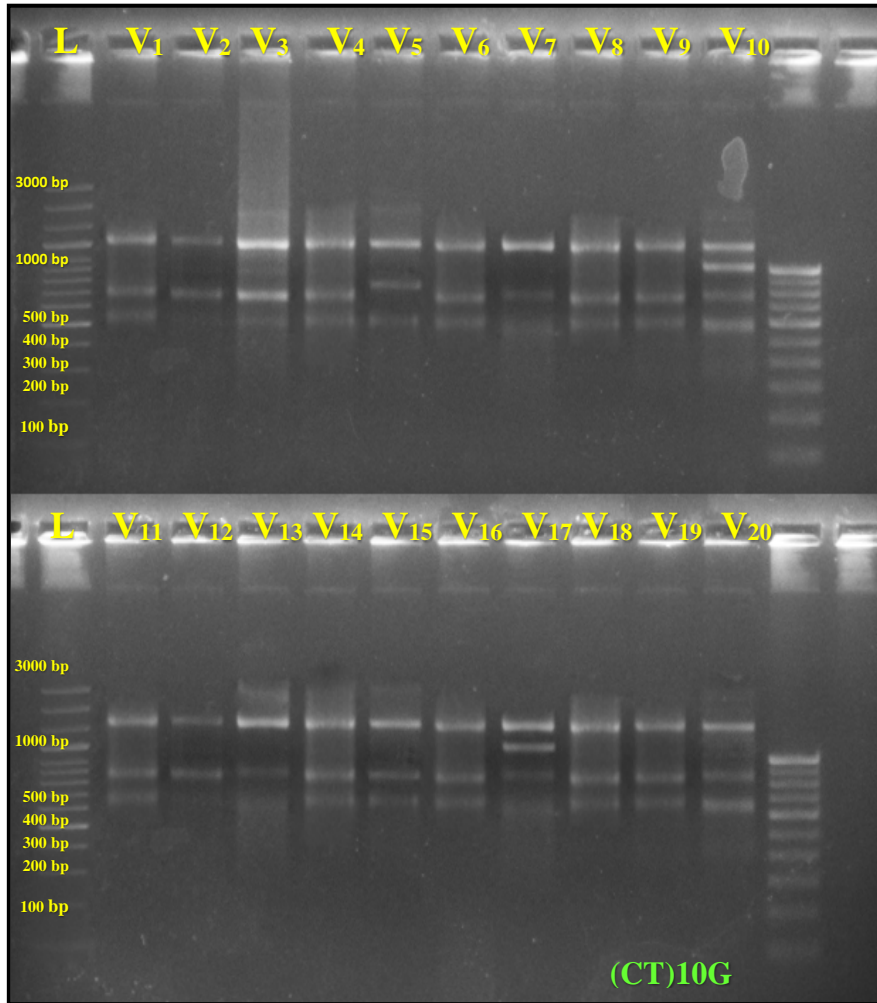


Plate 23. Amplification pattern generated by (CT)10G and DAT with 20 hybrids of *Ascocentrum*

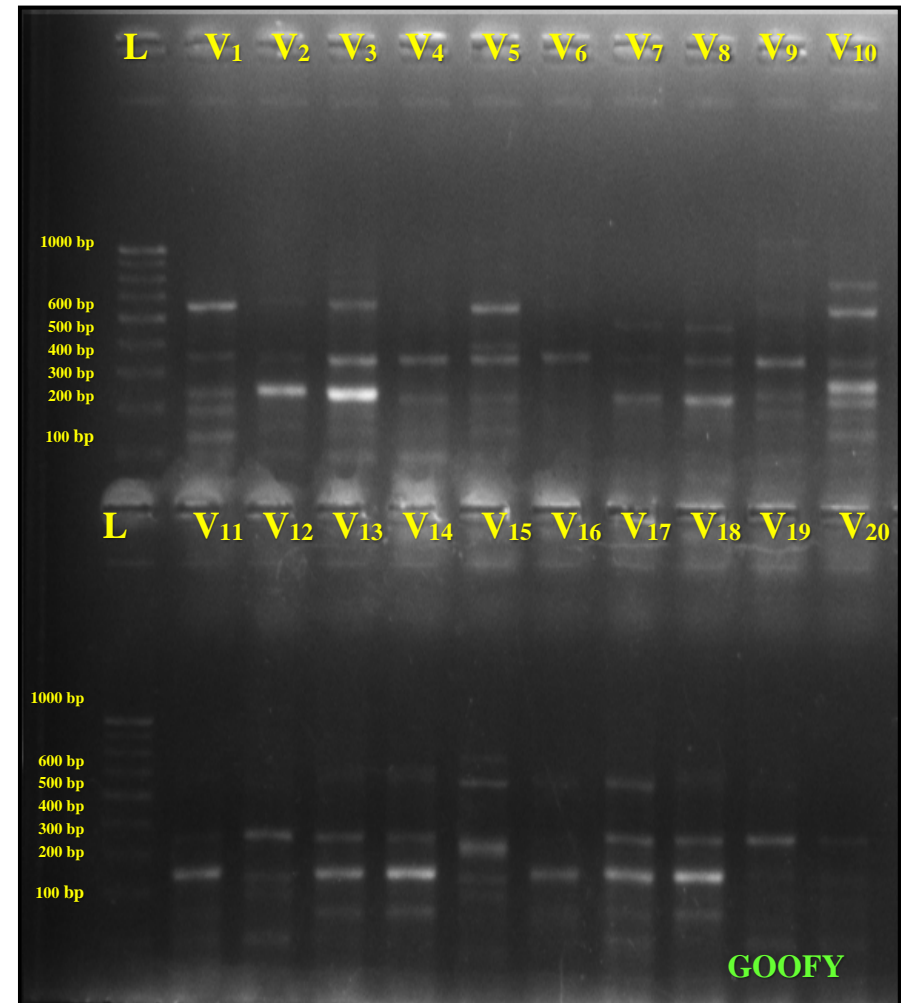
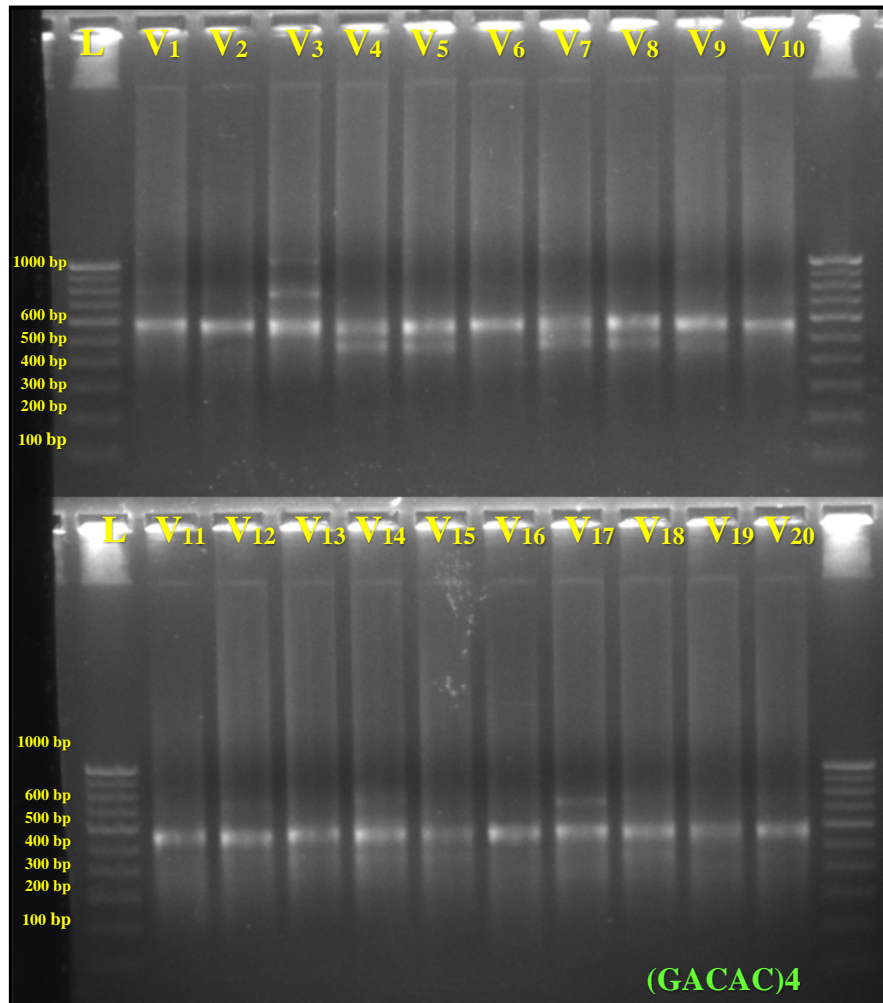


Plate 24. Amplification pattern generated by (GACAC)4 and GOOFY with 20 hybrids of *Ascozentrum*

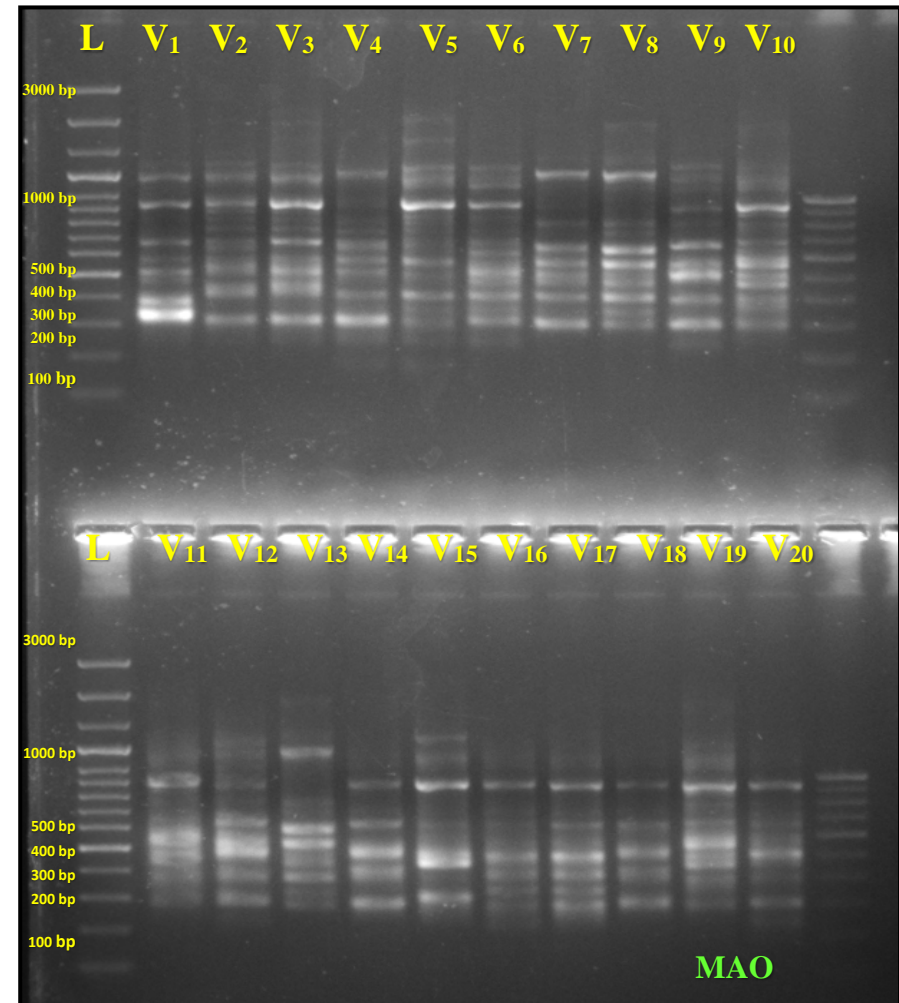
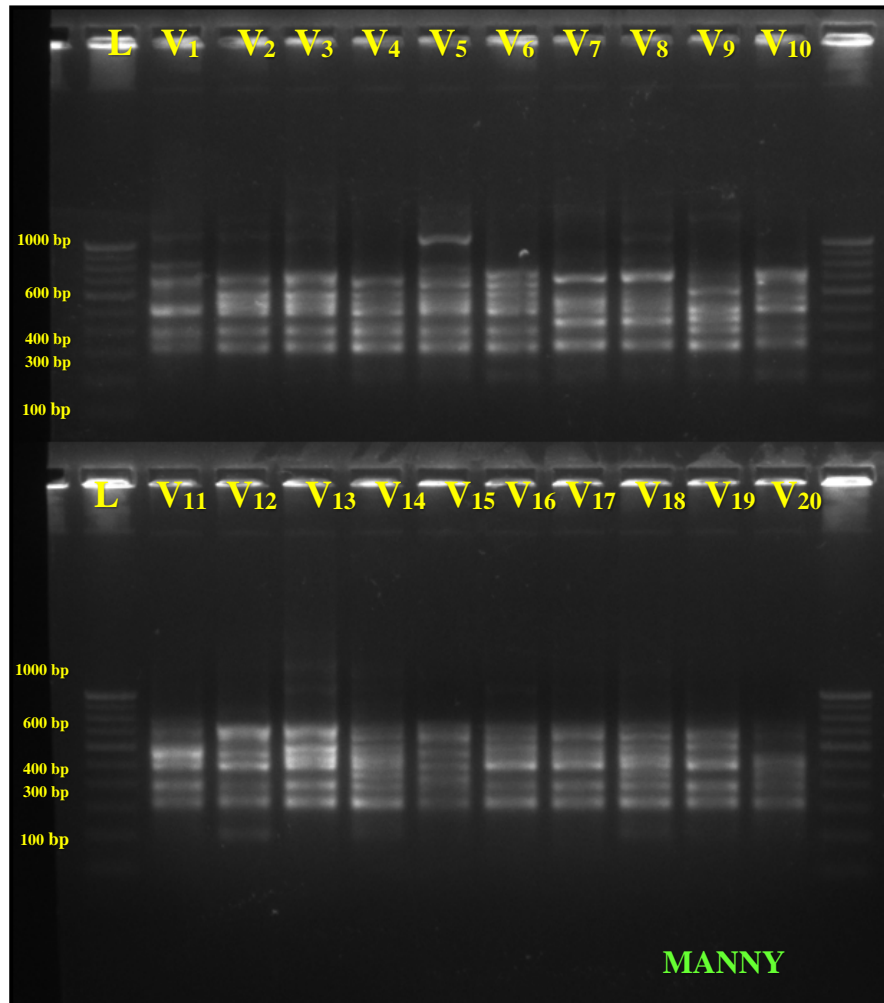


Plate 25. Amplification pattern generated by MANNY and MAO with 20 hybrids of *Ascozentrum*

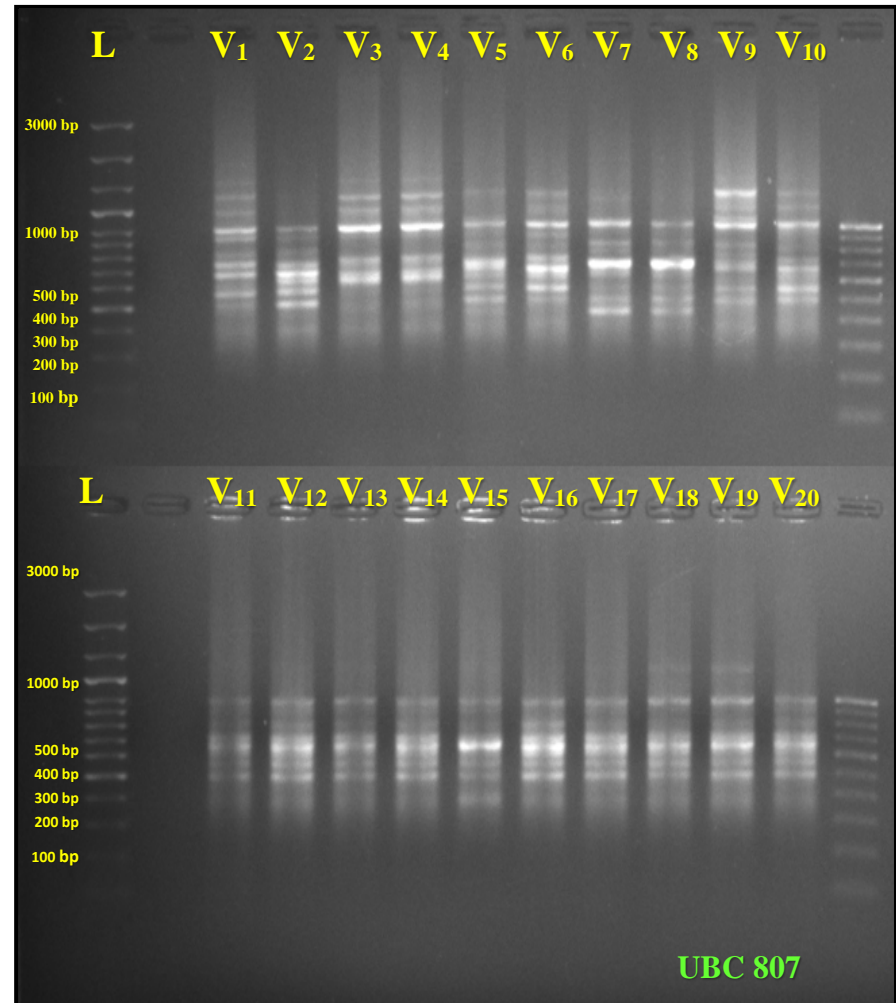
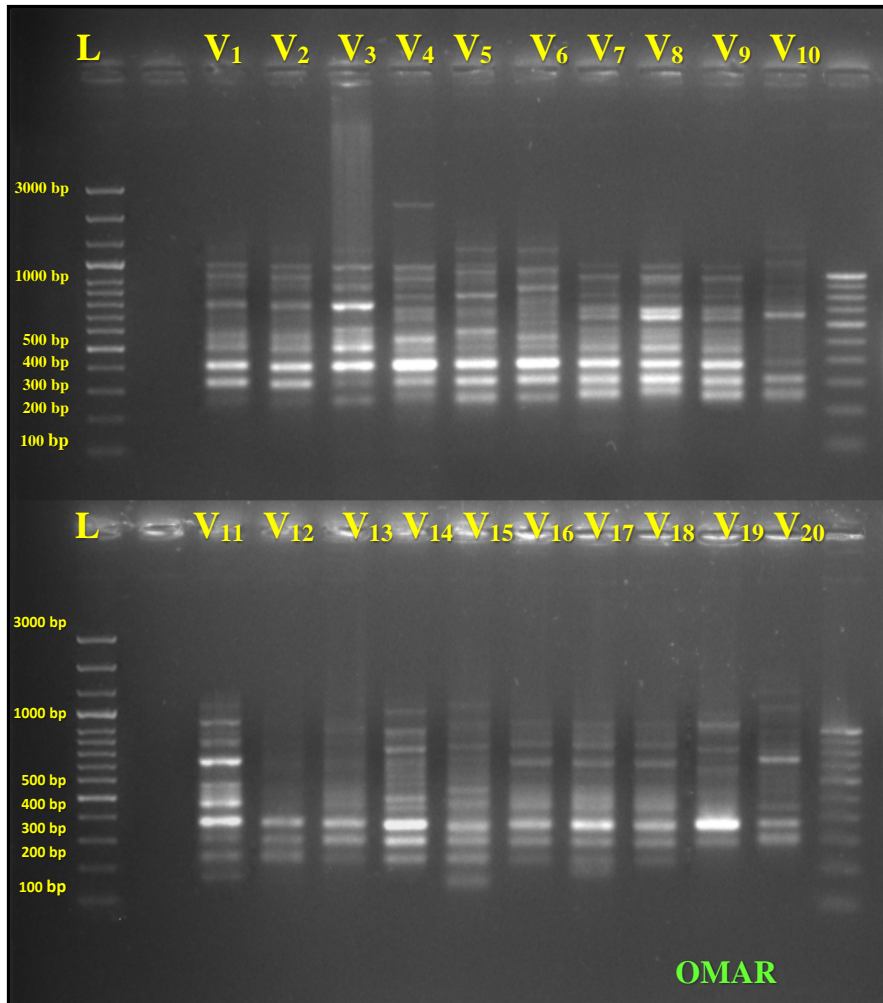


Plate 26. Amplification pattern generated by OMAR and UBC 807 with 20 hybrids of *Ascocentrum*



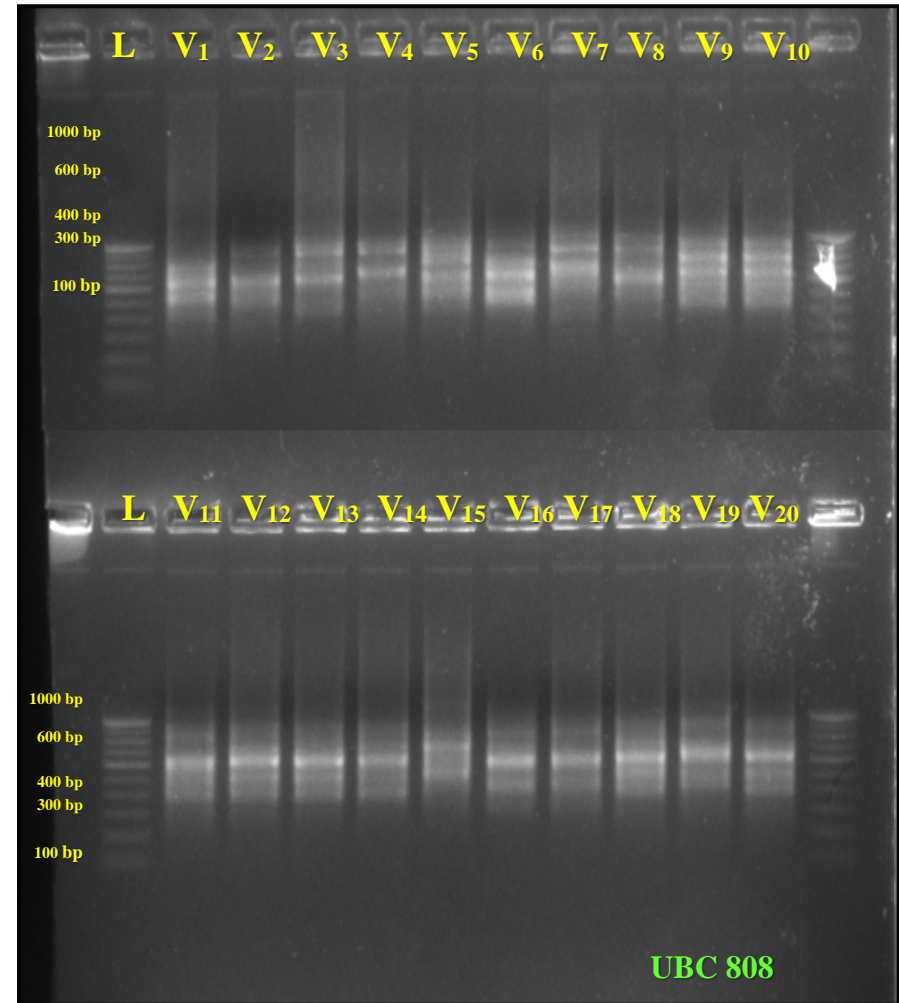
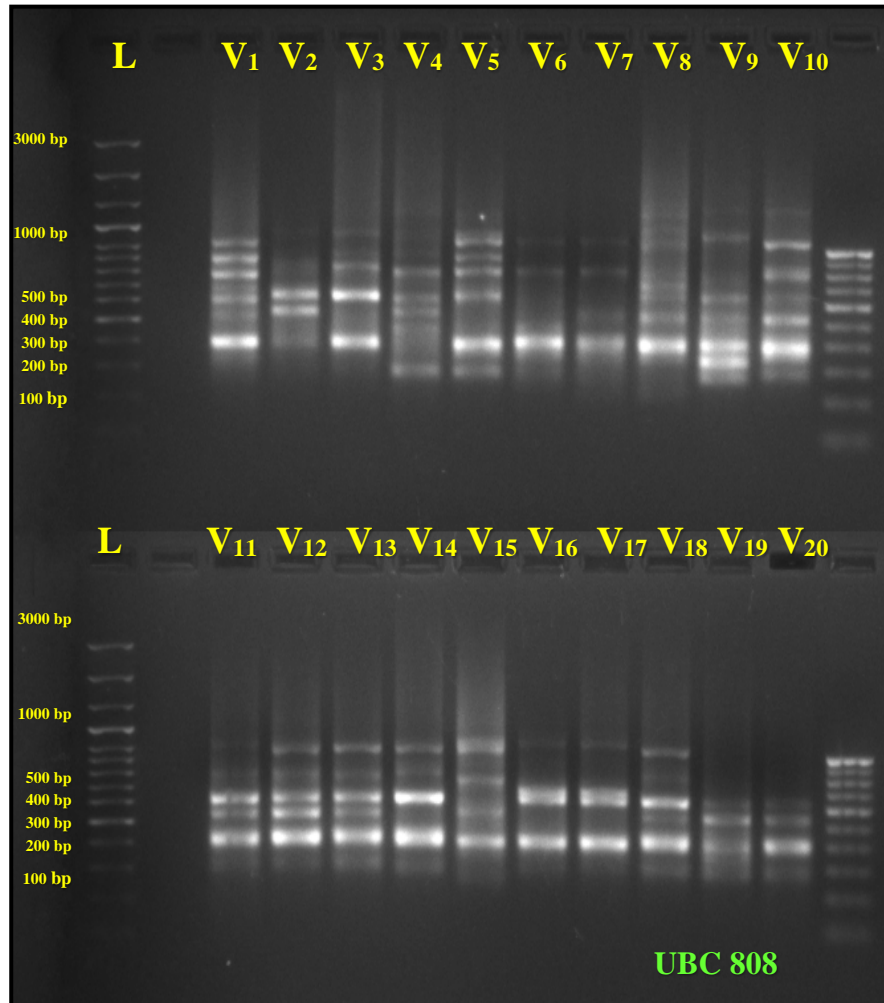


Plate 27. Amplification pattern generated by UBC 808 and UBC 809 with 20 hybrids of *Ascocentrum*

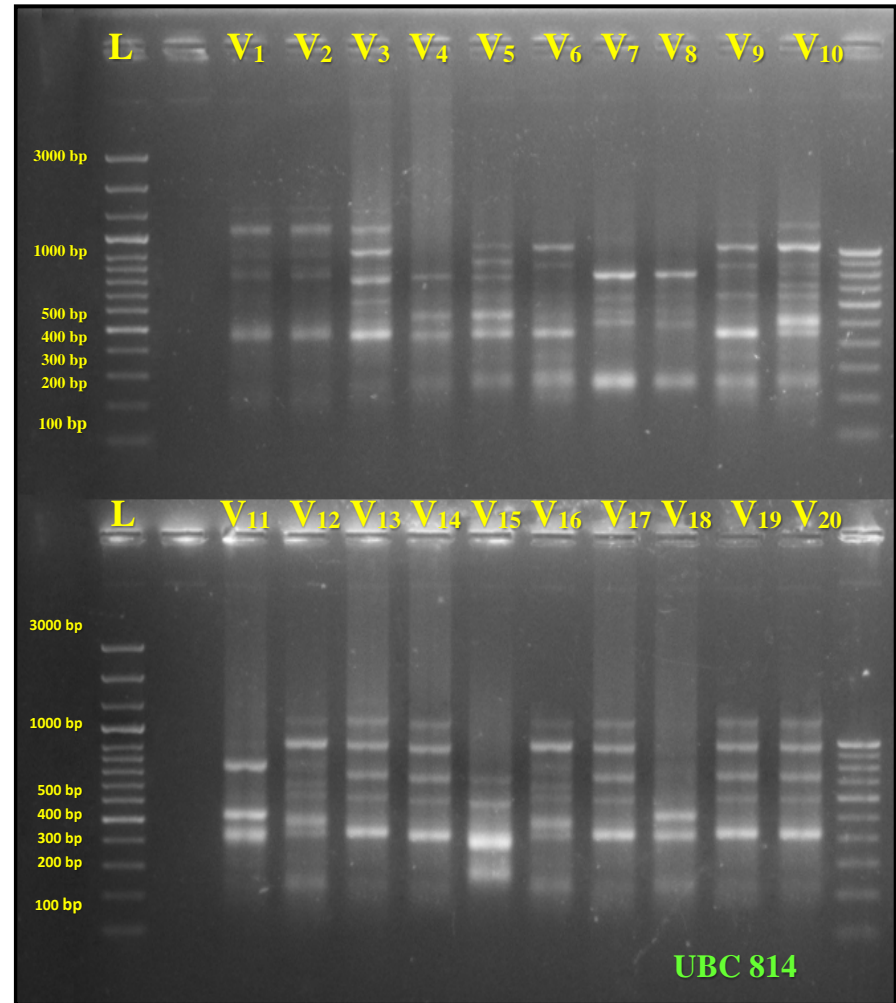
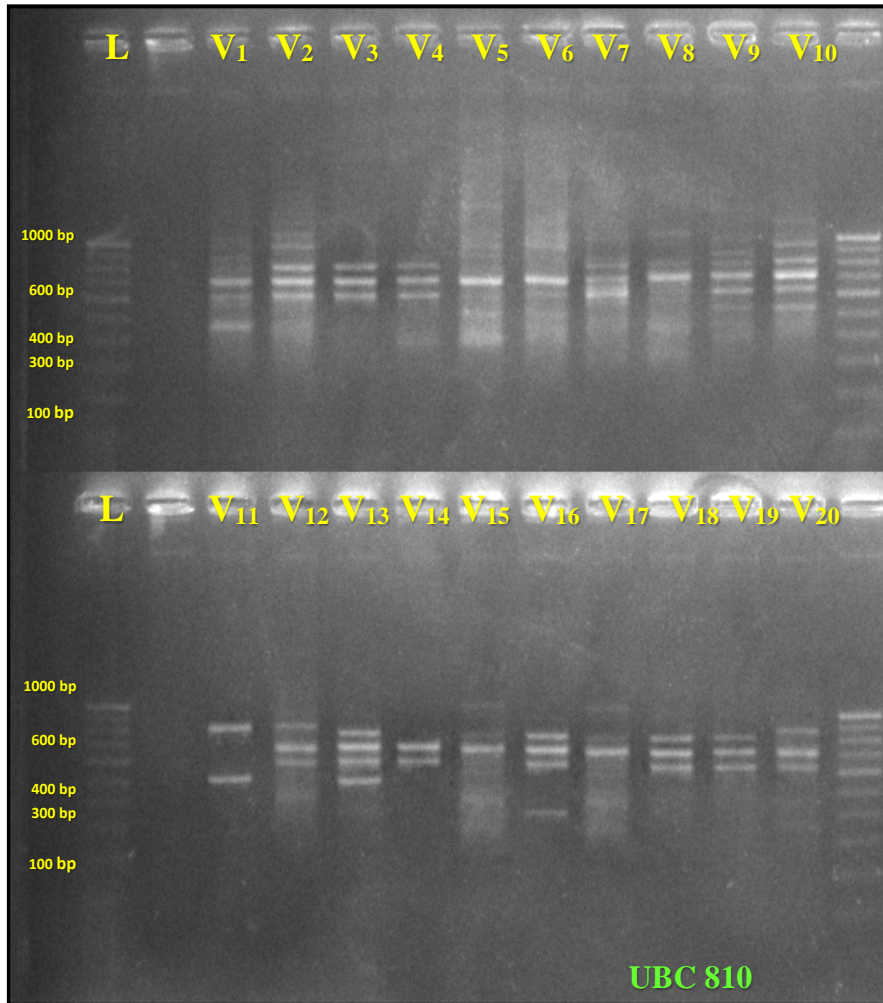


Plate 28. Amplification pattern generated by UBC 810 and UBC 814 with 20 hybrids of *Ascocentrum*

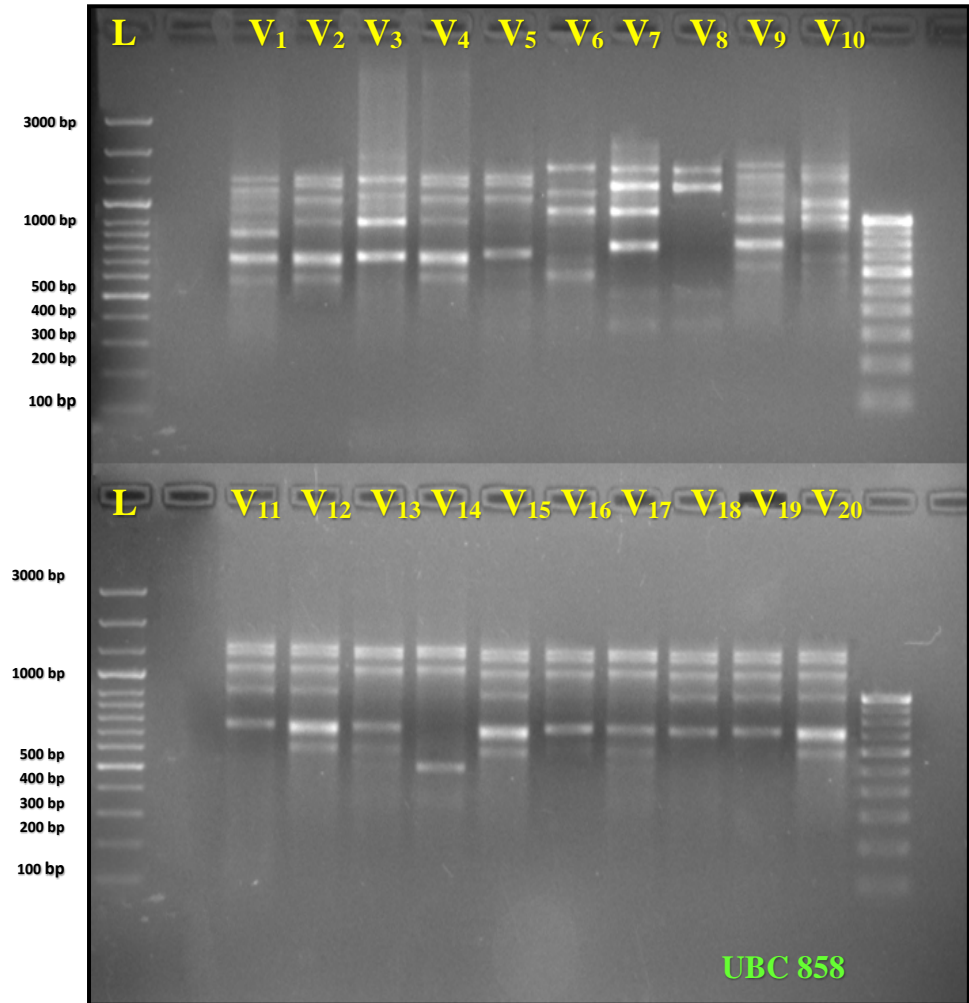
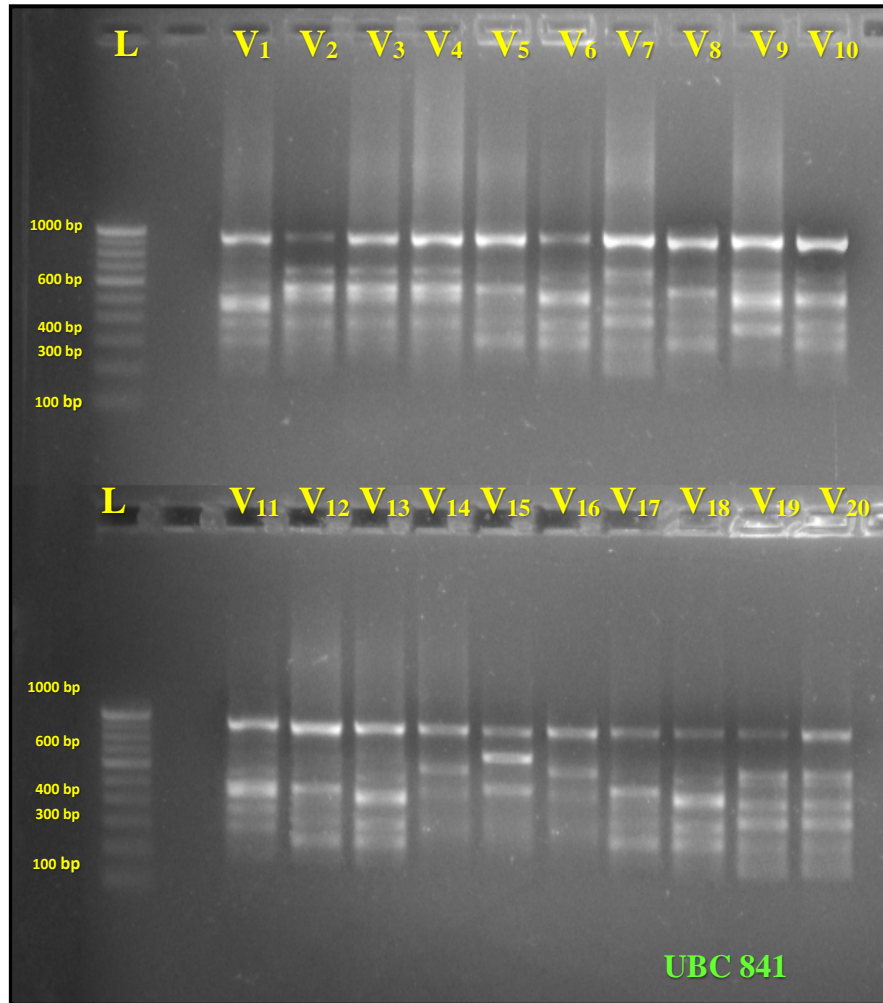


Plate 29. Amplification pattern generated by UBC 841 and UBC 858 with 20 hybrids of *Ascocentrum*

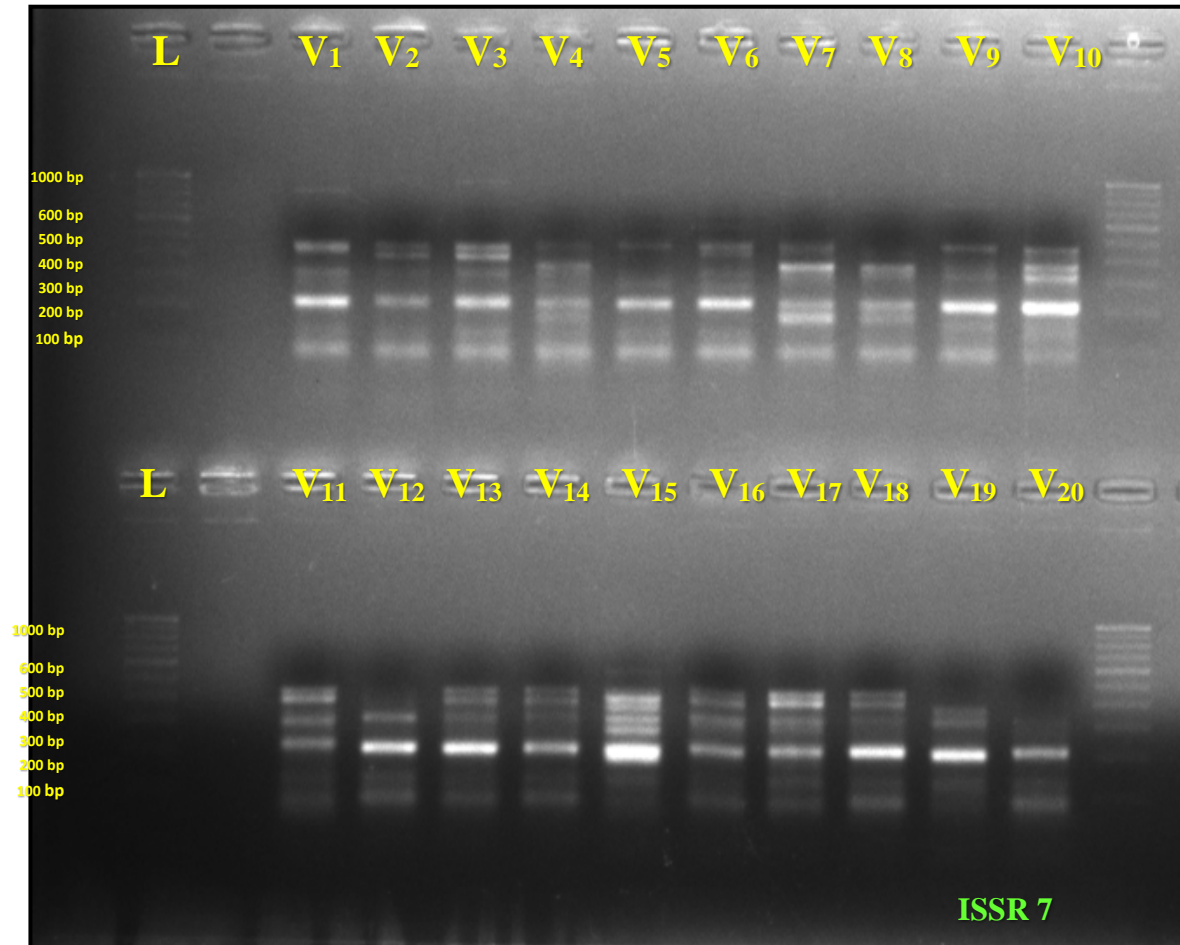


Plate 30. Amplification pattern generated by ISSR 7 with 20 hybrids of *Ascocentrum*

**Table 39. Particulars of polymorphic ISSR markers**

Sl. No.	Primer name	AT (°C)	No. of amplicons	Amplicon size (bp)		PIC value
				Min.	Max.	
1	7	54	20	126	1780	0.888
2	901	54	31	191	1163	0.930
3	17899A	48	18	314	1457	0.838
4	ACTG(4)	52	18	372	1346	0.770
5	AW3	54	21	388	1296	0.838
6	(CT)10G	60	22	500	1676	0.917
7	DAT	54	25	253	1154	0.912
8	(GACAC)4	52	11	453	1000	0.594
9	GOOFY	56	17	194	964	0.803
10	HB 9	52	16	219	900	0.864
11	Manny	52	21	200	1075	0.875
12	OMAR	54.3	24	155	2487	0.893
13	MAO	45	29	273	1367	0.896
14	UBC807	53	23	265	1181	0.652
15	UBC808	54	30	393	1477	0.926
16	UBC809	53.5	16	446	1040	0.872
17	UBC810	50.4	17	336	970	0.926
18	UBC814	52	24	216	1400	0.969
19	UBC841	53	15	206	958	0.928
20	UBC858	54	29	491	1787	0.925

showed very high level for diversity, and at 50 per cent level of similarity all 20 *Ascocentrum* hybrids grouped into a separate cluster. Hence, the clustering was done with minimum of 30 per cent similarity. At this similarity, all 20 hybrids into 14 different clusters. Six clusters *viz.*, Cluster I, Cluster II, Cluster V, Cluster X, Cluster XII and Cluster XIII observed with two members each, whereas all remaining cluster was observed with only one member each (Table 41.).

**Table 40. Pair wise similarity between varieties based on ISSR data**

Var. No.	V <sub>1</sub>	V <sub>2</sub>	V <sub>3</sub>	V <sub>4</sub>	V <sub>5</sub>	V <sub>6</sub>	V <sub>7</sub>	V <sub>8</sub>	V <sub>9</sub>	V <sub>10</sub>	V <sub>11</sub>	V <sub>12</sub>	V <sub>13</sub>	V <sub>14</sub>	V <sub>15</sub>	V <sub>16</sub>	V <sub>17</sub>	V <sub>18</sub>	V <sub>19</sub>	V <sub>20</sub>
V <sub>1</sub>	1.00																			
V <sub>2</sub>	0.32	1.00																		
V <sub>3</sub>	0.26	0.32	1.00																	
V <sub>4</sub>	0.20	0.20	0.33	1.00																
V <sub>5</sub>	0.17	0.19	0.16	0.23	1.00															
V <sub>6</sub>	0.18	0.13	0.14	0.17	0.26	1.00														
V <sub>7</sub>	0.12	0.10	0.14	0.14	0.17	0.21	1.00													
V <sub>8</sub>	0.11	0.09	0.10	0.17	0.15	0.17	0.31	1.00												
V <sub>9</sub>	0.11	0.13	0.09	0.13	0.18	0.18	0.24	0.24	1.00											
V <sub>10</sub>	0.07	0.09	0.10	0.14	0.18	0.20	0.16	0.18	0.29	1.00										
V <sub>11</sub>	0.12	0.12	0.09	0.07	0.13	0.15	0.07	0.09	0.10	0.07	1.00									
V <sub>12</sub>	0.12	0.10	0.08	0.08	0.11	0.12	0.12	0.09	0.09	0.11	0.27	1.00								
V <sub>13</sub>	0.10	0.08	0.07	0.08	0.09	0.08	0.06	0.05	0.09	0.07	0.22	0.23	1.00							
V <sub>14</sub>	0.11	0.09	0.08	0.12	0.11	0.11	0.09	0.07	0.09	0.11	0.19	0.22	0.32	1.00						
V <sub>15</sub>	0.11	0.08	0.12	0.12	0.09	0.07	0.08	0.07	0.08	0.08	0.18	0.16	0.16	0.16	1.00					
V <sub>16</sub>	0.08	0.10	0.08	0.11	0.11	0.11	0.06	0.06	0.08	0.08	0.17	0.19	0.15	0.24	0.18	1.00				
V <sub>17</sub>	0.10	0.09	0.11	0.10	0.09	0.10	0.08	0.07	0.08	0.08	0.20	0.23	0.25	0.29	0.19	0.26	1.00			
V <sub>18</sub>	0.08	0.06	0.07	0.08	0.08	0.08	0.05	0.07	0.06	0.11	0.14	0.27	0.24	0.20	0.18	0.33	0.34	1.00		
V <sub>19</sub>	0.16	0.14	0.13	0.15	0.06	0.07	0.06	0.09	0.11	0.09	0.13	0.13	0.18	0.17	0.16	0.20	0.22	0.30	1.00	
V <sub>20</sub>	0.10	0.09	0.09	0.10	0.05	0.08	0.03	0.06	0.11	0.12	0.13	0.13	0.16	0.16	0.12	0.25	0.23	0.31	0.44	1.00

**Table 41 Clustering based on ISSR scoring**

Sl. No.	Cluster	No. of members	Members of cluster
1	Cluster I	2	V <sub>1</sub> , V <sub>2</sub>
2	Cluster II	2	V <sub>3</sub> , V <sub>4</sub>
3	Cluster III	1	V <sub>5</sub>
4	Cluster IV	1	V <sub>6</sub>
5	Cluster V	2	V <sub>7</sub> , V <sub>8</sub>
6	Cluster VI	1	V <sub>9</sub>
7	Cluster VII	1	V <sub>10</sub>
8	Cluster VIII	1	V <sub>11</sub>
9	Cluster IX	1	V <sub>12</sub>
10	Cluster X	2	V <sub>13</sub> , V <sub>14</sub>
11	Cluster XI	1	V <sub>16</sub>
12	Cluster XII	2	V <sub>17</sub> , V <sub>18</sub>
13	Cluster XIII	2	V <sub>19</sub> , V <sub>20</sub>
14	Cluster XIV	1	V <sub>15</sub>

#### 4.3.2.5 Comparison between clustering pattern made by SSR and ISSR primers

The primers SSR and ISSR were efficient to cluster all the hybrids/varieties of *Ascocentrum*. For SSR the similarity was considered at or above 50 per cent, whereas multiple amplicons were produced by ISSR primers with all varieties/ hybrids, hence the similarity level was considered at or above 30 per cent. It was clearly evident from the similarity matrices and dendograms of SSR and ISSR primers (Table 37, 40, Fig. 16 and 17) that both types of primers produced almost similar kind of clustering for all 20 hybrids/varieties of orchids.

In SSR clustering, hybrids *viz.*, V<sub>1</sub>, V<sub>2</sub>, V<sub>3</sub>, V<sub>4</sub> and V<sub>5</sub> grouped into same cluster, whereas in ISSR clustering hybrids V<sub>1</sub>, V<sub>2</sub> clustered in one cluster and V<sub>3</sub>, V<sub>4</sub> clustered in another cluster. In clustering of both types of primers *i.e.* SSR and ISSR, hybrids/varieties *viz.*, V<sub>6</sub>, V<sub>9</sub>, V<sub>10</sub>, V<sub>11</sub>, V<sub>12</sub>, V<sub>16</sub> and V<sub>15</sub> grouped each into separate cluster, whereas V<sub>17</sub>, V<sub>18</sub> and V<sub>19</sub>, V<sub>20</sub> clustered into same cluster in both clustering.



# Discussion





## 5. DISCUSSION

Flamboyant, beautiful and intriguing, orchids have evolved to become the largest family of flowering plants in the world. Orchids are grown for their astonishing beauty and variety of flowers. They are also valued for their incredible diversity, beautiful appearance, brilliant colours and prolonged vase life. Orchids are important for their horticultural and floricultural appeal. Orchids account for a large share of global floriculture trade both as cut flowers and as potted plants for indoor display. Nowadays many people have turned up to growing orchids, because of the availability of a large number of hybrids and varieties worldwide. *Ascocentrum* is a monopodial orchid with beautiful long lasting flowers and there are many intergeneric hybrids with *Ascocentrum* as one of the parents.

In the present study, thirty intergeneric *Ascocentrum* hybrids/varieties were evaluated for their morpho molecular characters and the results obtained are discussed in this chapter.

### 5.1 MORPHOLOGICAL CHARACTERISATION

Orchids are known for their high magnitude of diversity and response to the environment. In India, the West Coast, especially Kerala, is one of the rare locations all over the world where orchids come up well, without the use of much sophisticated conditions (Rajeevan *et al*, 2009a). According to Abraham and Vatsala (1981a), the genetic plasticity inherent in Orchidaceae permitted an intermingling of genomes, not only at the species level but also at the generic level. The passport data of orchids collected and evaluated at various co-ordinating centres under AICRP on Floriculture in India were documented by Bhattacharjee *et al*. (2002). Rajeevan *et al*. (2002) described the genera of orchids and varieties/ hybrids which could be commercially grown under the conditions prevailing in India. Family Orchidaceae is known for its morphological and ecological diverse adaptation.

### 5.1.1 Quantitative plant characters

The quantitative plant characters evaluated under the study were plant height, plant spread, shoot girth, shoot diameter and internodal length; leaf characters were number of leaves, leaf length, leaf breadth, leaf area; and root characters were number of roots, length of roots and girth of roots.

*Ascocenda* is a bigeneric hybrid, whereas *Mokara*, *Vascostylis* and *Kagawara* are trigeneric hybrids and are the intermediate climbing orchids. *Ascocenda* are compact monopodial plants that can easily grow indoors, growth pattern is intermediate climbing pattern unlike vandaceous orchids and *Ascocentrum*; whereas, *Mokara*, *Vascostylis* and *Kagawara*, have growth pattern from tall climbing to intermediate climbing unlike their parents (Kaveriamma, 2007; Kaveriamma *et al.*, 2010; Sebastian, 2015 and Deepa, 2017).

In the present study maximum plant height was recorded in *Mok.* Omayaiy Yellow (144.11 cm) followed by *Mok.* Sayan × *Ascda.* Doung Porn (130.55 cm) and were found to be significantly superior to all other varieties throughout study period and these varieties also had comparatively good internodal length (4.63 cm). Plant height was minimum in *Vasco* Aroonsri Beauty (22.21 cm) which also had the least internodal length (2.10 cm). Plant height is influenced by growing conditions as well as plant genetic constitution. The variation in height in the present study might be due to the differences in internodal length. Since the varieties also had better internodal length. Abraham and Vatsala (1981) reported that an interaction was observed between plant genetic constitution and environmental factors which directly reflected on their growth, development and productivity. Geetha *et al.* (2009) made an attempt to study the variation through diversity analysis in 27 genotypes of *Arachnis*, belonging to monogeneric, bigeneric and trigeneric origin under warm humid tropical condition and reported significant differences with respect to vegetative and floral characters and the genotypes were grouped into four clusters.

There are also similar findings from other research workers. Forty monopodial orchids belonging to mono generic (15), bigeneric (15) and trigeneric (10) origin were evaluated under field conditions and distinguishable differences were noticed in both vegetative and floral characters (Kaveriamma *et al.* (2008); Kaveriamma *et al.*, 2010). Detailed morphological characterization of varieties belonging to monopodial orchids like *Mokara*, *Vanda*, *Phalaenopsis* and sympodial orchids like *Oncidium*, *Dendrobium* and *Cattleya* also have already been done (PPV & FRA, 2012; PPV & FRA, 2012a and De *et al.*, 2018).

In the present study plant spread (59.23 cm), leaf length (38.74 cm) and leaf area (68.63 cm) recorded were maximum in *Kag. Youthong Beauty*, whereas *Vasco. Aroonsri Beauty* had minimum plant spread (24.43 cm), leaf length (15.02 cm), leaf area (37.00 cm<sup>2</sup>) and also leaf breadth (1.04 cm) throughout the study period. Maximum leaf breadth was observed in *Mok. Omayaiy Yellow* (3.94 cm). The minimum value was found to be not more than 1.04 cm, might be due to its terete shaped leaves. Present findings are in line with the reports of Abraham and Vatsala (1981); Kaveriamma *et al.* (2008) and Geetha *et al.* (2009).

The more the spread, the spacing between the plants should also be more. Area occupied by the plant is indicated by plant spread which also determines the plant density in the growing environment. Leaf characteristics such as length, orientation and arrangement have direct influence in determining the plant spread, especially in the case of *Kagawara*, since it is non branching. The long and arching nature of leaves also has role in the spacing between plants.

According to Deepa (2017) leaf quality attributes both quantitative and qualitative also have an important role in the selection of plants for ornamental traits. It was also noticed that leaf length largely contributed to the leaf area in comparison to leaf width. Leaf area, number of leaves, leaf production interval along with leaf sheath characters could be directly correlated to the photosynthetic efficiency of plant

(Bose *et al.*, 1999). Such leaf characters of different orchids were described by Bhattacharjee *et al.* (2002); Kaveriamma (2007); Sebastian (2015); Deepa (2017) and Rahi (2017) in their studies.

Maximum number of leaves was found in *Mok. Sayan* × *Ascda. Doung Porn* (74.80) which was on par with *Mok. Omayaiy Yellow* (72.80), whereas the minimum number of leaves was found in *Mok. Khaw Phiak Suan* × *Ascda. Jiraprapa* (16.60). This might be due to the varietal differences. This is in confirmation with Kaveriamma (2007) who reported that the leaf production interval defined leaf yield which was co related with internodal length and plant height in most of intermediate to tall climbing orchids. Angle of orientation of leaves benefits the plant by proper interception of light. Leaf quality attributes both quantitative and qualitative also have important role in the selection of plants for ornamental traits.

Maximum shoot girth and shoot diameter were recorded in *Mok. Sayan* × *Ascda. Doung Porn* and the minimum in *Vasco. Aroonsri Beauty* which was on par with *Mok. Sayan* × *Ascda. Bangkuntein Gold*. Shoot girth and shoot diameter have profound influence on the plant size and strength and it could be inferred that the varieties with better shoot girth and diameter would have good anchorage and could accommodate more number of leaves and enhanced growth.

According to Sebastian (2015), the girth of shoot gave an indication of the strength of the stem and the shoot girth recorded in different varieties varied considerably. In their study maximum shoot girth was found in *Vanda Taweesuksa* × *V. Kultana Gold* × *V. Green Gold* and the minimum in *V. Varuvathe Pink*. The present results are in line with these findings.

Maximum number of roots was observed in *Ascda. Yip Sum Wah* × *V. JVB* (28.80) which was significantly superior to all other hybrids/varieties. The aerial roots arising from the base of the stem or seen along the stem help the plant to absorb

nutrients and moisture from the growing environment. The more number of roots would help for enhanced absorption and in turn more growth and good flowering. In different vandaceous orchids, roots were long and hung freely thus they indicated the need to cling on a support (Bose *et al*, 1999).

Goh (1983) reported that production pattern of roots was not driven by the genetic constitution but possibly by the physiological and environmental factors, as observed in *Aranda* orchids.

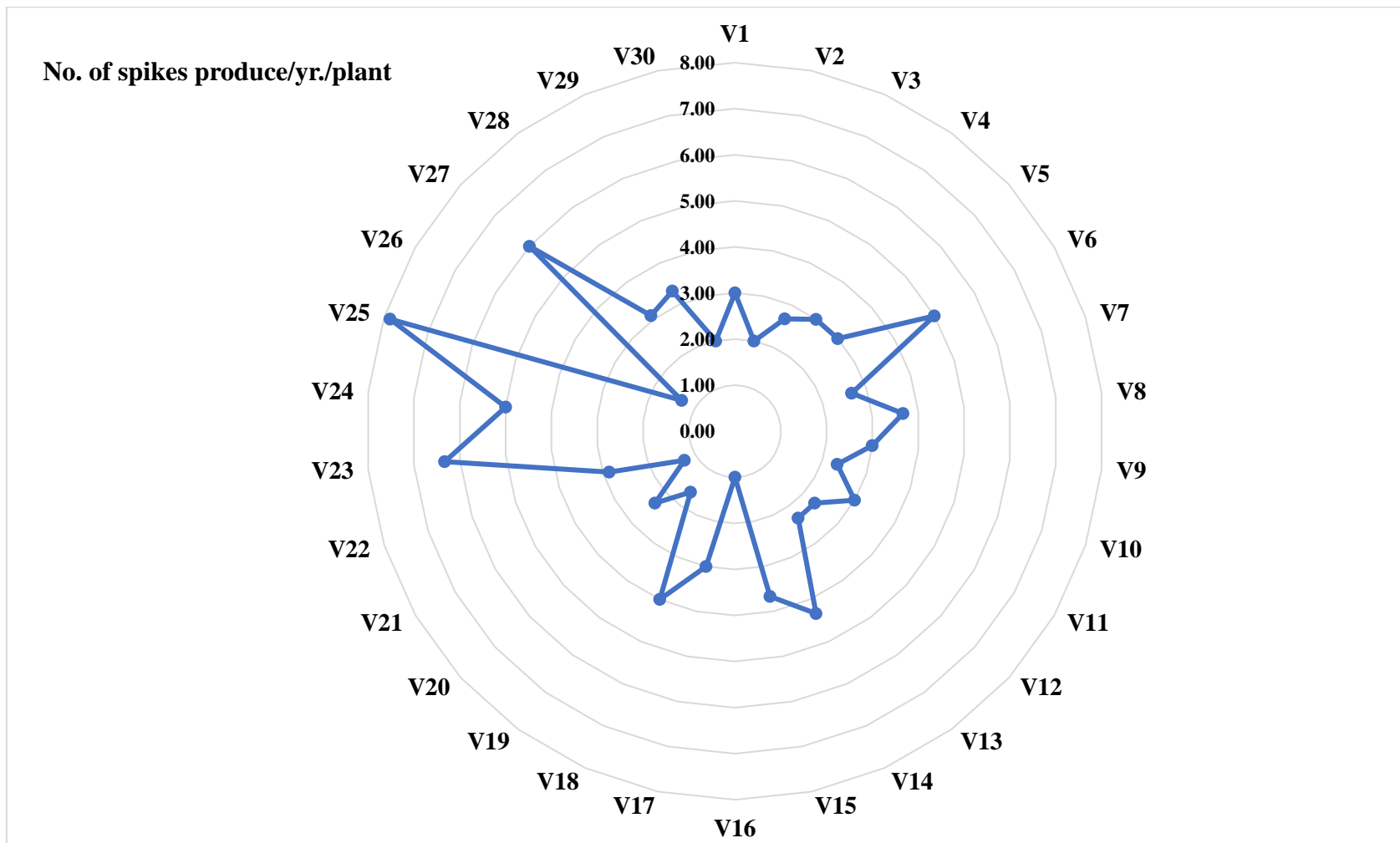
Brian and Wilma (2014) reported wide variation with regard to the root characteristics. Besides, the absorbent outer cover of roots formed by a layer of dead cells called velamen helps for the absorption of water and nutrients through the entire length.

### **5.1.2 Quantitative floral characters**

The quantitative floral characters evaluated under this study were spike characters like number of spikes produced per year, length of spike, length of rachis, length of the floral stalk, girth of a spike at the base, floret characters like number of florets per spike, internodal length, pedicel length (individual flower stalk length), length of floret, breadth of floret, length of labellum, width of labellum, length of column and spur length (Fig. 18 to Fig. 21).

Natural flowering occurs when environmental conditions become favorable for the reproduction of plants, since the plant starts to respond to the photoperiod and temperature (Lopez and Runkle, 2004).

*Mok. Sayan* × *Ascda. Doung Porn* was observed to produce the maximum number of spikes per plant per year which was on par with *Mok. Omayaiy Yellow* and *Kag. Youthong Beauty* (Fig.18). Eventhough least was observed in *Mok. Khaw Phiak Suan* × *Ascda. Jiraprapa*, it was statistically on par with the other 26 varieties.



**Fig. 18.** Number spikes produced per plant per year (yield of spikes) of intergeneric hybrids of *Ascocentrum*

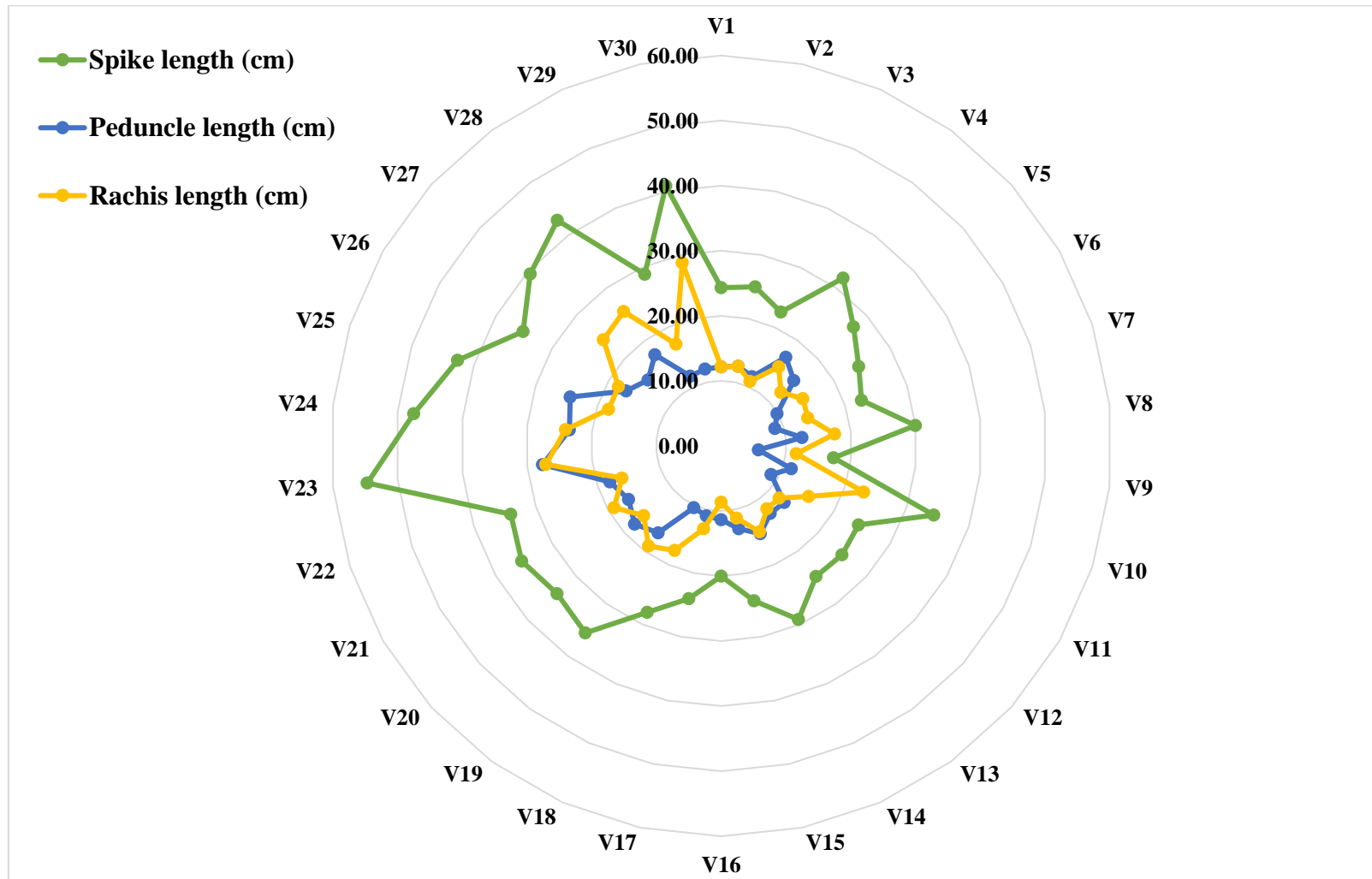
Orchid with higher yield of spikes could be considered as the best variety for commercial exploitation.

Appreciable variations were noted in spike length. Significantly higher spike length was noted in *Mok. Omayaiy Yellow* which was on par with *Mok. Omayaiy Orange*, whereas least was noted in *Vasco. Aroonsri Beauty* which was on par with 11 different varieties. Spike length is a desirable character for cut flowers; the more the spike length, the more price it would fetch. In the present study *Mok. Omayaiy Yellow* and *Mok. Omayaiy Orange* could be considered with high cut flower value (Fig. 19). Orchids with good spike length find a place in the cut flower trade which is not the criterion for pot plants.

Rachis length decides the flower bearing area. From the ornamental point of view, rachis length has to be considered together with the number and size of florets. In the present study, *Kag. Samrong* had maximum rachis length which was on par with 12 other varieties. It was minimum in *Mok. Khaw Phiak Suan × Ascda. Jiraprapa* which was on par with 10 different varieties (Fig. 19).

Flower stalk/peduncle length is given prime importance in cut flowers and those with more length will have more value and more demand. *Mok. Omayaiy Yellow* was observed to have the longest flower stalk which was on par with *Mok. Omayaiy Orange* and *Mok. Sayan × Ascda. Doung Porn* which means it would enhance the cut flower value (Fig. 19).

Spikes with good girth would be sturdier, hence it is more preferred to use as cut flower. Spike girth was maximum in *Mok. Chao Praya Sunset Orange* which was on par with five other hybrids. Spike girth was the minimum in *Vasco. Blue Bay White* which was on par with *Vasco. Aroonsri Beauty, Ascda. Sirichi Fragrance* and *Vasco. Pine Rivers Fuchsia Delight*. Spike girth influences the space occupied by each floret and also has helps in the compact arrangement of florets on the flower



**Fig. 19.** Spike, peduncle and rachis length of intergeneric hybrids of *Ascocentrum*



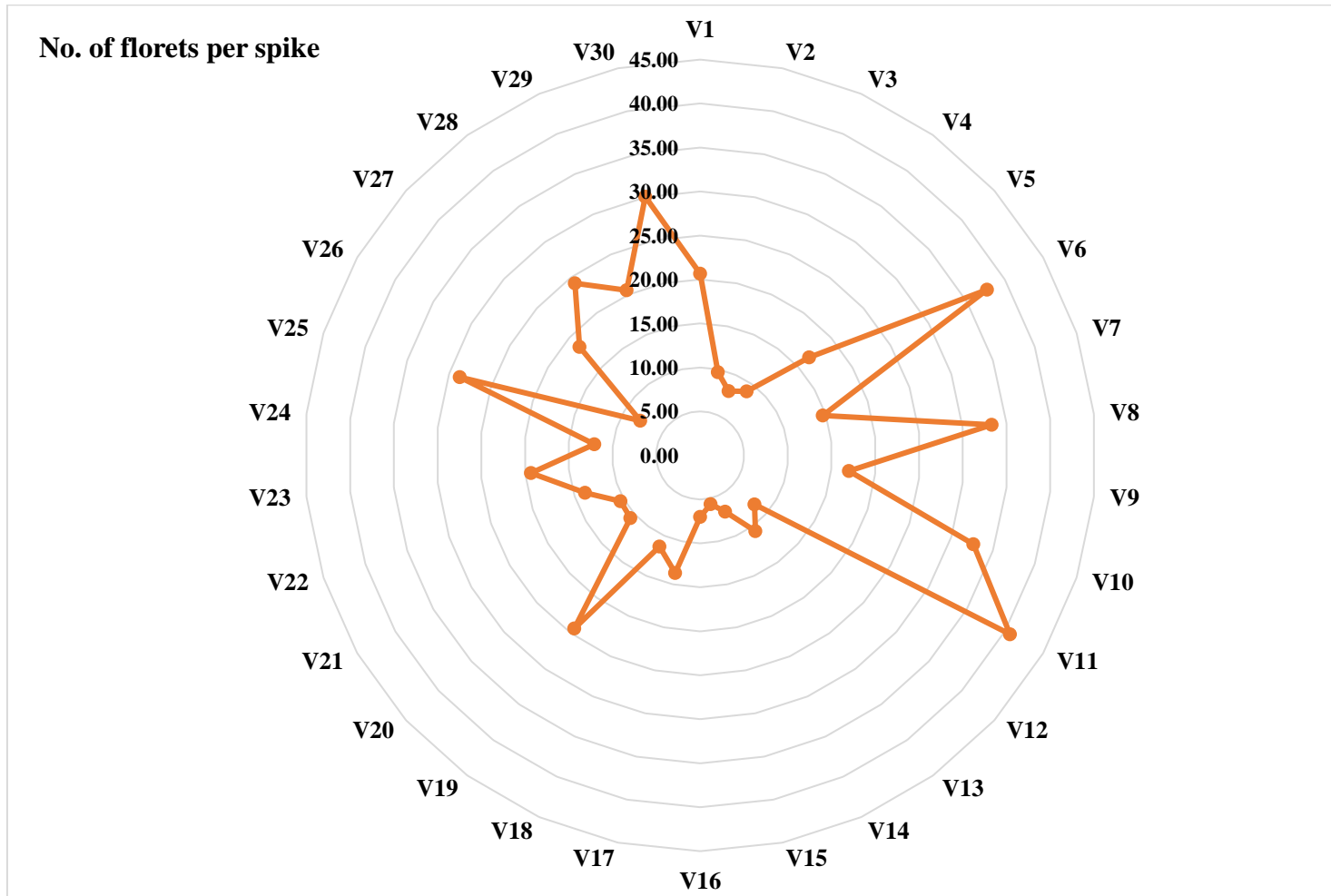
stalk. Thus it has much value in cut flower trade by giving good appeal and increased aesthetic value.

More number of florets per spike is a preferred character for use as a cut flower, which also gives beautiful appearance to cut flower as well as for indoor display of plants. During the period of study, *Vasco*. Blue Bay White produced maximum number of florets/spike which was on par with *Ascda*. Sirichi Fragrance (Fig. 20). This character may also helps in the compactness of the spike which is important in the ornamental point of view and enhances the commercial value.

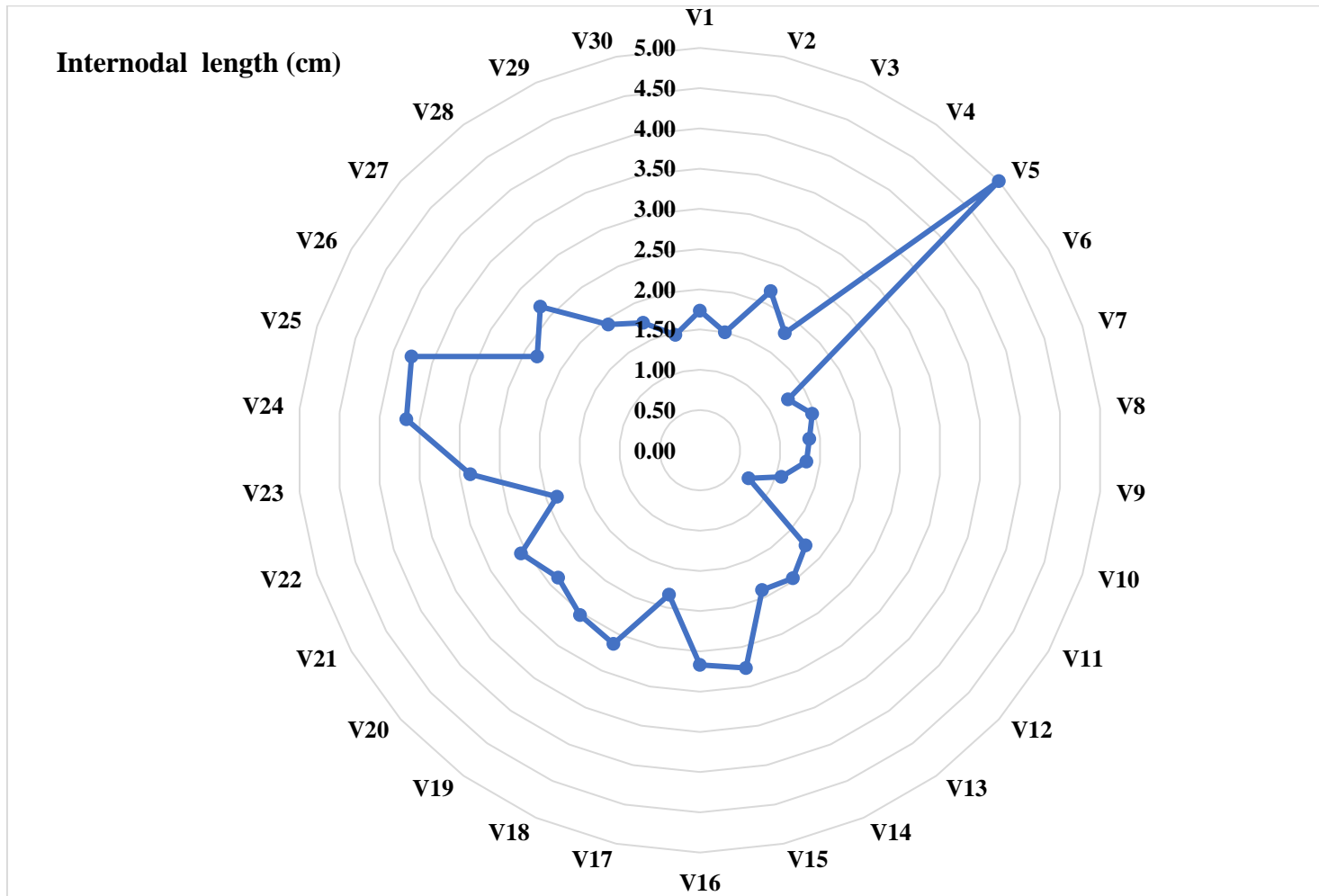
Increase in the internodal length between florets, is not a desirable character for cut flowers because of scattered appearance. Spikes with minimum internodal distance have good appeal leading to compactness of the spike and better quality. In the present study the minimum internodal length was observed in *Vasco*. Blue Bay White which was on par with *Vasco*. Pine Rivers Fuchsia Delight, which could be considered better for cut flower purpose (Fig. 21).

*Ascozentrum* hybrids showed detectable variations with regards to the pedicel/floret stalk length. Florets with good pedicel length are suitable for floral corsages. During the period of study, maximum pedicel length was recorded in *Ascda*. Kultana × *V. Bitzs Heartthrob* followed by *Mok*. Omayaiy Orange, *Mok*. Omayaiy Yellow and *Mok*. Sayan × *Ascda*. Doung Porn. Pedicel length was lowest in *Vasco*. Blue Bay White which was on par with *Vasco*. Pine River Blue, *Vasco*. Pine Rivers Fuchsia Delight, *Vasco*. Pine River Pink and *Ascda*. Sirichi Fragrance. In the present study a positive relation of flower size with pedicel length was also noticed and those varieties with good flower size had higher values for pedicel length. This, being a desirable trait for cut flowers, can be exploited.

Length and width of floret together determines the flower size. Floret size adds much to the ornamental value of cut flowers. Among all the *Ascozentrum*



**Fig. 20.** Number of florets per spike of intergeneric hybrids of *Ascozentrum*



**Fig. 21.** Internodal length of spikes of intergeneric hybrids of *Ascocentrum*

hybrids, *Ascda*. Kultana × *V. Bitzs Heartthrob* recorded the maximum floret size followed by *Ascda*. Suksamran Sunlight Yellow whereas it was minimum in *Vasco* Blue Bay White. *Vasco* Blue Bay White had densely arranged small flowers with the least internodal length which were very attractive and could be absolutely fitted as a pot plant for indoor display. Large sized showy flowers always attract attention and good floret size would be an added advantage for cut flowers and also can be used to make garlands and corsages.

Loksha and Vasudeva (1994) analyzed 746 Indian orchids and reported that those with large showy flowers were the most vulnerable for commercial exploitation and the most likely to be endangered species.

Lip or labellum is the most attractive part of an orchid flower giving it different shapes and names. It varies in colour, size and shape among varieties. The lip was oriented upward in the bud, but, as it later developed, the pedicel or ovary twisted so that the lip usually got oriented downward by the time the flower opened, a process called floral resupination (Anon. 2019b), Slight variations were observed in quantitative characters like length and width of labellum, in the hybrids/varieties studied.

Cluster analysis with 14 different floral characters revealed 12 clusters apt 75 per cent similarity. It was observed that cluster 2 and cluster 5 were least similar with each other, whereas, the highest inter cluster distance was observed in cluster 6 and cluster 10. Cluster 6 which included *Ascda*. Sirichi Fragrance and *Vasco* Blue Bay White was found to have the lowest internodal length with the highest value for number of florets per spike, also observed to have lower flower length and flower width. Cluster 10 was found to have the high mean values for spike length, flower length and flower width. Geetha *et al.* (2009) made an attempt to study the variation through diversity analysis in 27 genotypes of *Arachnis*, belonging to monogeneric, bigeneric and trigeneric origin under warm humid tropical condition and reported

significant differences with respect to vegetative and floral characters studied and the genotypes were grouped into four clusters to understand the inter cluster distance.

### 5.1.3 Phenological characters

Phenological plant characters recorded were interval of leaf production, leaf longevity (days), days taken from spike emergence to opening of first floret, 75 per cent florets and complete opening of all florets; longevity of spike on plant and life of individual floret on the spike. Generally orchids were grouped into two types, monopodial and sympodial depending upon their growth habit. Monopodial orchids such as *Ascocentrum*, *Arachnis*, *Renanthera*, *Ryncostylis*, *Vanda*, *Phalaenopsis*, etc. have a main stem which continues to grow year after year. Sympodials like *Dendrobium*, *Cymbidium*, *Cattleya*, etc. have a main stem which terminates growth at the end of each season. Kaveriamma (2007) reported that the phenological leaf and floral characters of monopodial orchids were important due to their long lasting nature.

Leaf phenology also has important role in the selection of varieties for commercial traits. Interval of leaf production is indicative of leaf yield. The lowest interval of leaf production was observed in *Mok. Sayan* × *Ascda. Doung Porn* (32.17 days) and the highest in *Mok. Sayan* × *Ascda. Bangkuntein Gold* (108.44 days). Maximum leaf longevity was observed in *Vasco. Aroonsri Beauty* (292.20 days). *Mok. Chao Praya Sunset Yellow Spot* was observed to have the minimum leaf longevity (129.02 days).

Flowering phenology varied among different varieties. Tropical low land growing hybrid orchids were found to be as day neutral plants which were not influenced by day length (Soon, 1980). It was noted that the hybrids showed acropetal succession of flowers. This is in the conformity with the finding of Goh (1977) that flowers in the inflorescence of *Vanda* and *Arachnis* tribe opened acropetally in one day interval.

In the present study the variety *Vasco*. Pine Rivers Fuchsia Delight was early with respect to floret opening and had taken 14.99 days to open the first floret from spike emergence. Once the flower bud formation started, the development time depended upon the temperature and genetic constitution (Lopez and Rankle, 2005). Dressler (1981) reported that rainfall had a direct correlation with the flowering phenology in tropics. Flower bud initiation occurred after the spike had reached a certain length under the required environmental conditions.

Usually the inflorescence has to be harvested, when 75 per cent florets were opened for cut flower purpose. *Vasco*. Pine Rivers Fuchsia Delight was the variety earliest to harvest among all the varieties, whereas *Vasco*. Pine River Pink took maximum days to attain harvestable stage. De *et al.*, (2014) reported that *Cymbidium* hyb. 'PCMV', harvested at two bud opened stage, had got maximum vase life (66.8 days). The finding of the present study that different varieties had different harvestable stage is in line with the finding of De *et al.*, (2011).

The duration between the spike emergence to opening of all florets also showed significant differences among varieties. The maximum number of days for opening of all florets was recorded in *Mok*. Chark Kuan Pink, whereas the minimum was in *Ascda*. Yip Sum Wah  $\times$  V. JVB. This is in confirmation with the results of Deepa (2017) that different days were taken from spike emergence to opening of all florets in vandaceous orchids.

Spike longevity on plant is closely related with growth and quality of plants. In the present study three varieties *viz.*, *Mok*. Sayan  $\times$  *Ascda*. Doung Porn, *Mok*. Chark Kuan Pink and *Kag*. Youthong Beauty were observed to have spike longevity more than 35 days. Spike longevity on the plant is generally an indicative of the longevity after harvest, a major criterion for increasing the commercial value of orchids.

A sharp increase in ethylene emission was found during flower maturation, opening and senescence. Ethylene played an important role in the regulation and coordination of senescence in climacteric flowers (Lopez and Runkle, 2005). In the present study life of individual floret was highest in *Mok. Chark Kuan Pink*, whereas it was lowest in *Vasco. Blue Bay White* might be due to the differences in the senescence of flowers resulting from the changes in the ethylene emission.

#### **5.1.4 Post harvest characters**

Post harvest characters like fresh weight of spike, wilting of the first floret, life span of floret, spike longevity, water uptake, and physiological loss in weight were recorded in all the varieties. Vase life or longevity of a cut flower could be assessed on the basis of attributes like diameter and length of florets, opening of flowers, changes in fresh weight, diameter or length of stem/pedicel, senescence pattern, colour of petals, total longevity and foliage burning.

Fresh weight of spike was highest in *Mok. Omayaiy Yellow* which was significantly superior to other hybrids except *Mok. Sayan × Ascda. Doung Porn* and *Mok. Omayaiy Orange* (Fig. 22). More fresh weight of spike is a favorable character in which all the parents might have contributed for the better commercial trait.

Kaveriamma (2010) reported that *Mokara*, being a trigeneric hybrid between *Ascocentrum × Arachnis × Vanda* and some of the *Mokara* hybrids offer the very best of all the three parents. Flowering behavior of *Makara*, is unlike other vandaceous orchids, since *Arachnis* is one of the parents in *Mokara*; most of *Makara* hybrids also retain the flowering pattern of both classic climbing orchid and other vandaceous orchids thus offering good qualitative attributes of flowers of the intermediate climbing monopodial orchids.

Spike longevity in vase is determined by senescence and wilting of petals or wilting of first floret. Variety *Mok. Chark Kuan Pink* recorded significantly high

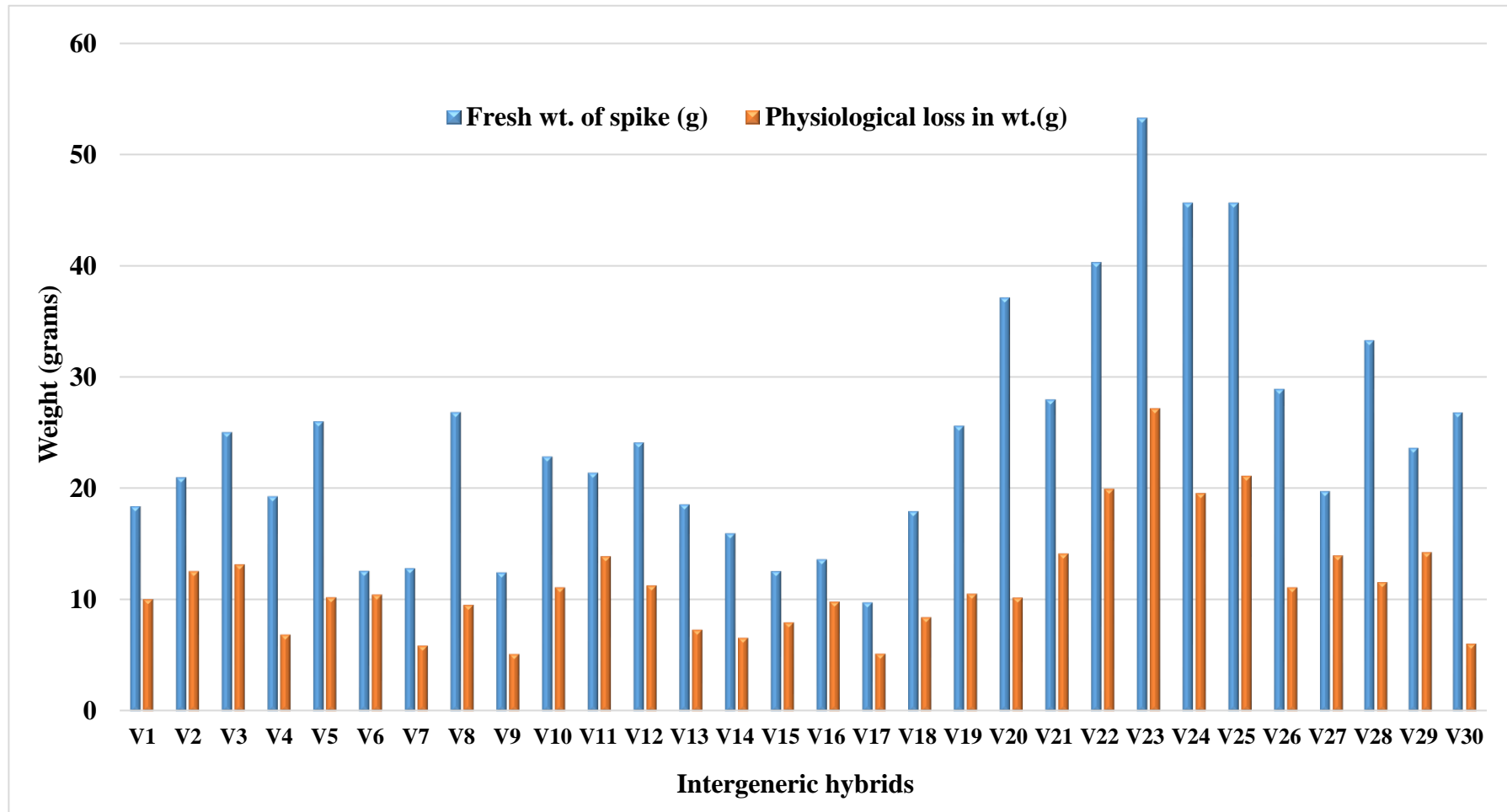


Fig. 22. Fresh weight of spikes and physiological loss in weight of *Ascozentrum* hybrids

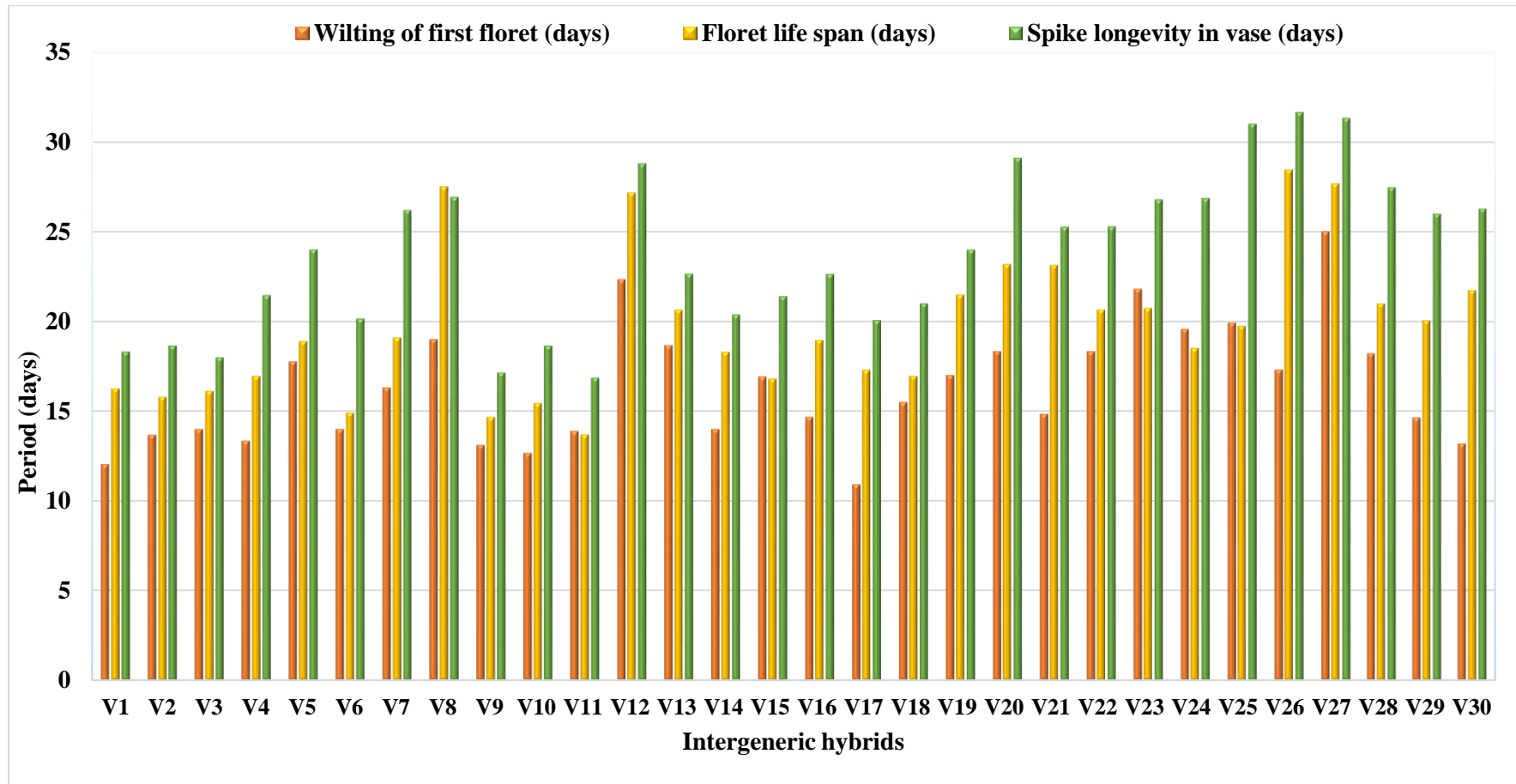


spike longevity among all the selected hybrids. The variety *Vasco*. Blue Bay White recorded the minimum value for spike longevity which was on par with 11 other hybrids (Fig. 23). Spike longevity varied in the different hybrids of the *Ascocentrum*. This is in line with the finding of De *et al.* (2014) that the hybrids of *Dendrobium*, *Vanda* and *Mokara* remained perfect from 7 days to 30 days, all the flowers of *Cattleya* and *Phalaenopsis* remained fresh for 1 to 4 weeks and *Aranda* lasted for 18 to 28 days.

Marked variations were noted in post-harvest characters as well as lasting quality of flowers in the *Ascocentrum* hybrids/varieties. Ethylene is considered to be the main hormone responsible for early senescence. In *Cymbidium* hyb. 'Red Princess,' pulsing with 5% sucrose increased vase life upto 56 days (De *et al.*, 2014a).

In the present study *Vasco*. Blue Bay White took the minimum duration (13.76 days) for wilting after bud opening of first floret, even though it was on par with 21 other hybrids/varieties. The period taken for wilting of the first floret was maximum in *Kag*. Youthong Beauty (25.00 days) (Fig. 23). Bud opening in vase might be related to hormonal functions and active growth of internal tissues in the plant, hence differences were noticed among varieties.

In the present study, floret life span was maximum in the variety *Mok*. Chark Kuan Pink (28.44 days), which was on par with 6 other varieties. Variety *Vasco*. Blue Bay White had the shortest floret life span (13.76 days) (Fig. 23). This is in conformity to the findings of Deepa (2017), wherein varietal difference was reported with respect to floret life span; she reported maximum floret life in *Neostylis* Lou Sneary followed by *Vasco*. Crown fox Red Gem. A sharp increase in ethylene emission was found during flower maturation, opening and senescence. Ethylene played an important role in regulating and co ordinating the senescence in climacteric flowers as reported by De *et al.* (2014a). Production of this hormone was found less



**Fig.23.** Post harvest floral characters of intergeneric hybrids of *Ascozentrum*

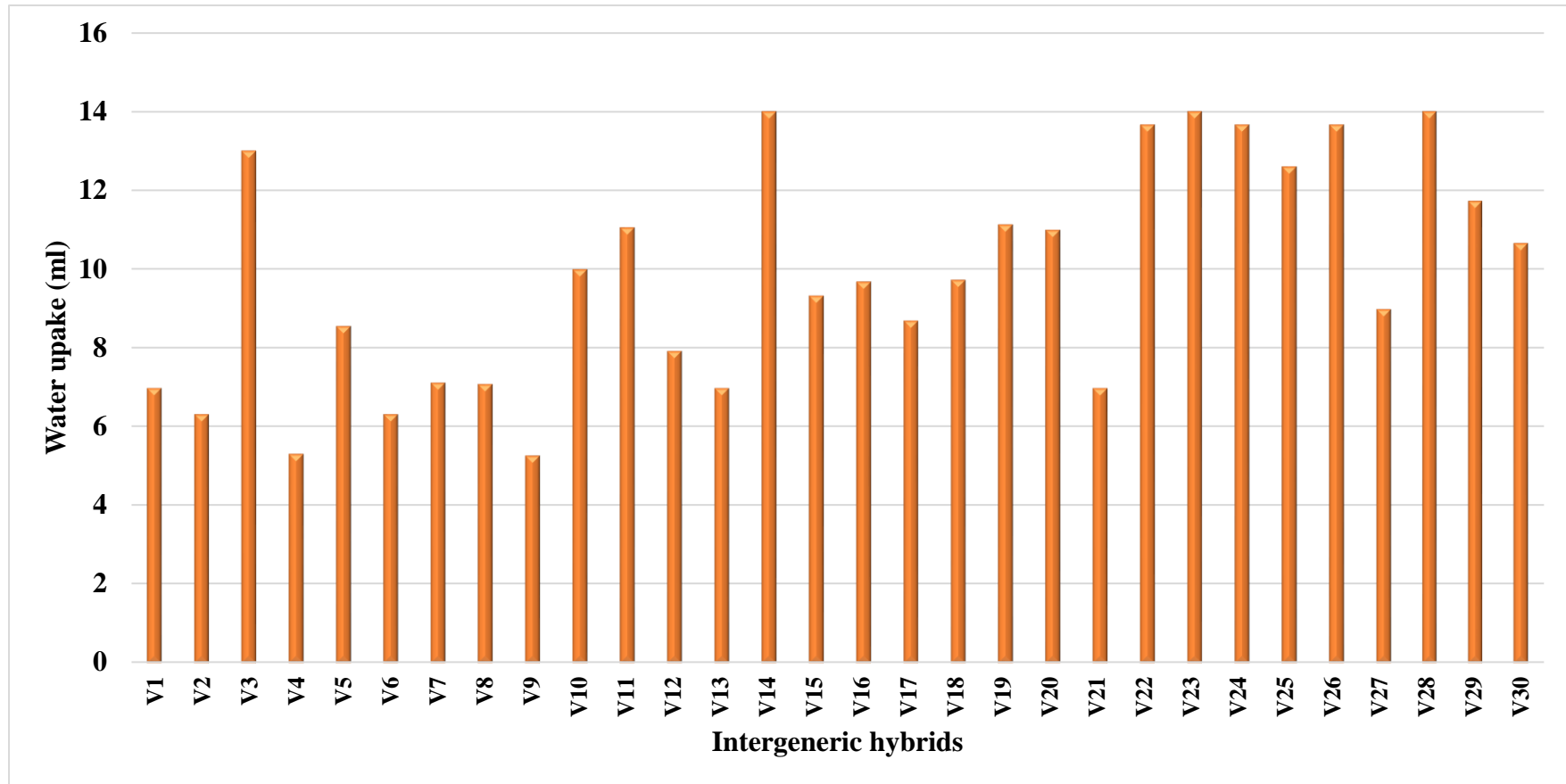
and stable in floral buds and young flowers, and the increase in ethylene concentration in the opened flowers led to wilting (De *et al.*, 2014a). The floret life or freshness of flowers is also one of the attributes deciding the post-harvest quality of spike when used as a cut flower.

Water absorption indicates metabolic activities as well as the retention of turgidity of the tissues. Even though the variety *Mok. Omayaiy Yellow* was observed to have maximum water uptake (14.00 ml), it was on par with 20 other varieties. The variety *Vasco. Aroonsri Beauty* had the minimum water uptake (5.29ml) (Fig. 24). This could be related to the physiological loss in weight of spike when it was kept in vase. Deepa, (2017) also reported similar results of varietal differences in water uptake; and it was reported minimum in *Vasco. Crownfox Red Gem*. According to De *et al.*, (2014a) the failure of water uptake as a result of stem blockage, might be due to air blockage, microbial growth or physiological plugging.

In the present study *Mok. Omayaiy Yellow* had minimum physiological loss of weight (PLW) which was on par with *Mok. Sayan × Ascda. Doung Porn, Mok. Sunspot* and *Mok. Omayaiy Orange*. PLW was maximum in *Vasco. Aroonsri Beauty* which was on par with 21 other hybrids/varieties (Fig. 22).

Physiological loss of weight is connected with freshness and turgidity of cut flowers and has prominent role in deciding the vase life and in turn the value of cut flowers. Hence those flowers with minimum PLW are to be selected for getting maximum vase life which can be exploited for commercial use. Post harvest characters of any flower have profound influence on the quality and cut flower value.

Sebastian (2015) stated that PLW of the spike under vase condition was maximum in *Vanda Lumpini Red x V. Taweewan* and this was related to water absorption and retention in the tissues.



**Fig. 24. Water uptake of intergeneric hybrids of *Ascozentrum* in vase**

Deepa (2017) reported minimum physiological loss in *Neostylis* Lou Sneary. This is in line with the finding that different varieties behaved differently in vase with respect to PLW.

### 5.1.5 Qualitative plant characters

The qualitative plant characters of thirty *Ascocentrum* hybrids/varieties were observed. These were recorded with respect to shoot characters *viz.*, nature of growth, nature, colour and branching; leaf characters *viz.*, shape, texture, margin, apex, colour, pigmentation, orientation, nature of sheath, colour of sheath and leaf base; and root characters *viz.*, origin, branching, colour, and nature.

Two types of growth habits were observed in the selected varieties. Intermediate climbing *Ascocentrum* hybrids showed hanging nature of growth while tall climbing varieties were found with prostrate nature of growth.

Shoot characters have important role in determining the strength of the plants as well as appearance. In the present study, the shoots were found to be as medium thick semi sturdy, sturdy, or stout. They seemed as brown and greenish brown coloured with little or no branching. Branching of the shoots was observed in *Vasco*. Aroonsri Beauty, *Vasco*. Pine Rivers Fuchsia Delight, *Mok*. Rassmatozz, *Mok*. Khaw Phiak Suan × *Ascda*. Bicentennial Yellow Spot, *Mok*. Sunspot, *Mok* Calipso Jumbo, and *Mok*. Sayan × *Ascda*. Doung Porn. According to Kaveriamma *et al.*, (2010), branching in monopodial orchids affected the flowering behavior and reported it suggested that it was better to have less branching. Hence in the present study the varieties with less branching could be selected for good flowering. However, the varieties with more branching, may be recommended for indoor display purpose due its appealing nature.

Leaves of *Ascda*. Udomochai and *Ascda*. Kultana × *Vanda* Bitzs Heartthrob and *Kag*. Samrong were observed as deeply channeled towards the base. *Mok*.

Calypso Pink and *Mok*. Calypso Jumbo were found with linear leaves. *Ascda*. Yip Sum Wah  $\times$  V. JVB, *Vasco*. Pine River Blue, *Vasco*. Pine River Pink, *Kag*. Christie Low and *Kag* Boon Ruby were found to have semiterete leaf and rest of the varieties with quarter terete leaves. *Vasco*. Aroonsri Beauty was the only variety with terete leaves.

Sin *et al.* (2002) reported that the terete vanda had little genetic relationship with the strap leaved vanda. Phuekvilai *et al.* (2009) grouped vandaceous orchids based on morphological and botanical characteristics, showing distinctive difference between strap leaved vandas and terete leaved vandas, and further distinguished the closely related cultivars of the vanda orchids.

Peyachoknagul *et al.* (2014) also reported the different shapes of leaves *viz.*, strap leaves, terete, semi terete, and quarter terete leaves which could be considered as important markers to distinguish the genetic characteristics.

In orchids, leaf quality attributes also have an important role in the selection of plants for ornamental traits. High variation was seen among the leaves of epiphytic orchids and also found diversity in the leaf markers of various sympodial and monopodial orchids. In the present study, less variation was observed with respect to leaf texture and was observed to have smooth and rigid leaf texture along with entire leaf margin among the thirty *Ascocentrum* hybrids/varieties. Leaf apex was acute in *Ascda*. Yip Sum Wah  $\times$  V. JVB and *Vasco*. Aroonsri Beauty and was emarginated in rest of the hybrids/varieties. Leaf colour was found green and leaf sheath colour dark green and with the pigmentation (colour changes with maturity) in some varieties. Leaf orientation was straight, arching and horizontal among the varieties while the leaf base was sheathed and nature of leaf sheath was found membranous, thick and fleshy. This is in conformity with the reports by Kaveriamma, (2007); Rajeevan *et al.* (2009); Sebastian (2015) and Deepa (2017) that

various monopodial vandaceous orchids showed diversity among the hybrids with respect to their qualitative leaf characters.

Root origin was basal and along the stem in all the selected *Ascocentrum* hybrids/varieties. Profused growth and branching were noticed in a large number of roots among most of the hybrids/varieties and was cylindrical in shape in all the varieties. The roots were mostly greenish grey and old roots were greyish brown in some varieties. If root branching was more in any variety, it could be inferred that absorption rate of water and nutrients will also be higher leading to enhanced growth and flower production. According to Rajeevan *et al.* (2002) grey or greenish grey or whitish green coloured roots with cylindrical growth were the signs of healthy aerial roots. Hence it could be concluded that in the present study the hybrids/varieties had healthy roots.

#### **5.1.6 Qualitative floral characters**

The qualitative floral characters of the hybrids/varieties recorded were spike characters- blooming period, spikes per plant at a time, orientation and nature of inflorescence, colour, colour of inflorescence; floret characters- fragrance, colour of florets and pigmentation; petal characters like shape, curvature, apex, margin, base, margin, apex colour and colour pattern; lebellum/lip characters, shape at the apical lobe region, nature of apical lip, shape at the lateral lobe region, nature, apex, surface, base colour, margin colour, apex colour and colour pattern; column characters-colour, colour pattern and length as well as spur length and type of spur.

While formulating DUS guidelines in *Oncidium* from the 60 morphological characters. (Geetha *et al.*, 2014) and DUS guidelines in monopodial orchids (PPV & FRA 2012) and description for qualitative characteristics in dendrobium (PPV & FRA 2012a) and also, it was observed that column and spur orientation showed uniqueness. These types of characters have relevance for the identification and

protection of plant varieties, hybrids, species and genera and in turn useful for the breeding programmes.

In general the plants showed better performance during the entire period of study. Several tropical low land orchid flora showed year round flowering which was mainly controlled by their genetic constitution (Goh, 1984). Stanford (1971) conducted a study on phenology in West African orchids and found that flowering phenology of some orchids were genetically controlled. In the present study, peak flowering was noticed during May-Jun. after the commencement of rainy season. Least flowering was observed during Nov.-Feb. It might be because of short day length prevailing during Nov.-Feb., which might not have supported flowering in *Ascocentrum* hybrids/varieties. Thus flowering could be related with the environmental factors especially day length.

In the present study different *Ascocentrum* hybrids which behaved identically in a particular phenophase were also observed. By observing, flowering phenology of these orchids, eight phenophases viz., Dec.-Jan., Feb.-Mar., Apr.-May, Jun.-Jul., Aug.-Sept. and Oct.-Nov. were recorded and they were categorized to respective phenophases. This is in agreement with the results of Deepa, (2017) that there were four phenophases of different flowering seasons in vandaceous orchids. According to Yong and Hew (2004), photoperiodism, vernalisation and juvenility were the three important factors that determined the flowering season and ontogeny and all the studied species have flowered during March-July.

Apart from the year round flowering, upto five blooming periods were observed during the entire period of study. It was five in *Mok. Sayan* × *Ascda. Doung Porn* (Nov.-Dec., Feb.-Mar., Apr.-May., Jun.-Jul. and Aug.-Sept.) and four in *Mok. Sunspot* (Jun.-Jul., Nov.-Dec., Apr.-May and Aug. –Sept) and *Kag. Youthong Beauty* (Nov. Dec., Jan.-Feb., Apr.-May. and Jun.-Jul.). And other varieties showed single, twice and trice, blooming periods in a year. Multiple number of spikes per



plant at a time was observed in *Mok. Omayaiy Yellow* and *Mok. Sayan × Ascda. Doung Porn*. Three spikes per plant at a time were observed in *Ascda. Sirichi Fragrance*, *Vasco. Pine Rivers Fuchsia Delight*, *Mok. Sunspot* and *Mok. Omayaiy Orange*.

Amin (2004) found that *Aerides multiflorum* had longest inflorescence with maximum flowering area and number of florets per inflorescence, the qualities suitable for spike to use as a cut flower. According to Kaveriamma (2007) and Kaveriamma *et al.* (2010), high yield of spikes per plant per year was an additional advantage for commercially growing orchid varieties to be used as cut flowers. In the present study, the varieties *Mok. Omayaiy Yellow* and *Mok. Sayan × Ascda. Doung Porn* had multiple spikes and hence could be selected as good varieties for cut flower production. In the present study flowering was observed to appear acropetally. This is in confirmation with Lehnebach (2003) who reported acropetal flower opening in *Chloraea lamellate* orchid. Flowering behavior of different *Ascocentrum* and vandaceous orchids were also reported by Kaveriamma (2007); Kaveriamma *et al.* (2010); Sebastian (2015) and Deepa (2017). According to Goh and Arditti (1985), the flowering season of orchids and its duration were determined genetically.

Regarding the nature of inflorescence, dense and lax nature were observed in the varieties studied. The spike arose from lateral position and were oriented in arching or erect manner. Arching orientation of spikes with short stalk and dense nature are good for indoor display particularly in the hanging baskets or as pot plants while the erect spike is an added advantage as cut flower adding to the ornamental quality. Amin (2004) reported that inflorescence of *Vanda teres* was stout and erect and was very suitable for keeping in vase, but inflorescence of *Rhynchostylis retusa* was drooping type and not suitable as a cut flower for vase and also found that flowering nature of the plant decided the method of display especially in orchids.

With respect to orientation of florets, they were faced in all directions. Spikes were green in colour at peduncle or flower stalk and rachis, whereas, in *Vasco*. Aroonsri Beauty it was green with brown streaks and was different from other varieties. High variation was observed with respect to colour of inflorescences which was recorded by observing predominant colour and shades (violet, yellow, white, purple, red, pink, orange, green and lavender). Floret colour also showed abundant variation when observed using RHS colour chart. Variation in colour might be due to the pigmentations with respect to different genetic constitution of parental and non-parental genera of the intergeneric hybrids.

Fragrance in flowers is another character for general acceptance. It was observed that pigmentation and fragrance were negatively related within the varieties studied; wherever mild and sweet fragrance was present, pigmentation (colour changes during maturation) was absent. Highly scented orchid flowers add immeasurably to their overall appeal. *Brassovola cuculata*, the ghost orchid, bloomed during autumn and was highly fragrant at night (Brian and Wilma, 2014). Kaveriamma, (2007) conducted a study in forty monopodial orchids and found that *Vanda* Prolific had sweet fragrance. Fragrant substances produced in osmophores of many orchids serve as attractant for pollinators and had impact in plant reproduction (Huber *et al.*, 2005). Flach *et al.* (2004) reported that major chemical class of compounds present in the labellar secretions was triterpenoid. Deepa (2017) reported that while scoring fragrance, *Vanda* Mimi Palmer recorded the highest mean total score followed by *Neostylis* Lou Sneary.

Petals showed high variations in their shape (obovate, orbicular and lanceolate) and curvature (straight, deflexed with incurved apex, deflexed with deflexed apex, deflexed with straight apex and incurved with incurved apex). Petal apex was acute, obtuse and truncate, petal margin was entire and undulate; petal colour at base, margin and apex region were predominantly observed by different as

single, double, triple and multiple, and different shades (violet, yellow, white, purple, red, pink, orange, green and lavender), colour pattern (shaded, uniform, spotted, netted and tessellated). Size, shape and pattern of lateral sepals were also observed. Cares-Suárez *et al.* (2011), stated that colour was not always a good indicator of odour and that colour scent association may be complex, depending on concerned pollination ecology of the population.

Labellum is a modified petal which is the most attractive and highlighting part of an orchid flower. It is usually glabrous with bilobed apex. Among the tested varieties/ hybrids labellum showed different colours like white, purple, green, red, violet, yellow and orange with uniform, shaded and streaked pattern of colour. Column colour pattern was uniform in almost all the varieties. It was reported that the sexual portions of the orchid flower were quite different from other flowers, and they were used to characterize the family (Anon., 2019). The filaments, anthers, style, and stigma were reduced in number and were usually fused into a single structure called the column. It was observed that majority of the orchids retain only a single anther at the apex of the column. Most of the orchids could be distinguished according to their spur length and spur varied from short, medium and long. Whereas, spur types were conical, tubular and saccate spur types among the hybrids/varieties. It was long and tubular in certain varieties.

### **5.1.7 Visual evaluation**

Selection of good and healthy plant and flower by visual observation accounts to a great extent in judging the cut flower. Rating of market acceptability and consumer appealness is an important step which has to be done before introducing a new variety into trade, especially in flower crops. Orchids are used to create an interior plant scape as well as outdoor garden theme with amazing colours in

landscapes. They also contribute for creating better ambience and comfort in indoor garden.

Visual evaluation was done in thirty intergeneric hybrids. To find out their suitability for cut flowers and indoor display. There was no significant variation among the varieties for spike for use as a cut flower and plant for use in the indoor display. The highest total mean score for visual observation was obtained in *Mok. Omayaiy Yellow* for spike to use as cut flower; whereas, for indoor display the highest mean total score was recorded in *Vasco. Pine River Pink*. Similar visual scoring for use as cut flower and indoor display was reported by Deepa (2017), Sebastian (2015) and they reported the best outcome.

Fragrant orchids also have better scope as natural room freshener. Tiny miniature orchids were grown in pots as small as 3 cm in diameter by Brian and Wilma (2014) and tall growing monopodials were grown in clay pots and *Cymbidium* and *Paphiopedilum* in deep pots. *Vanda*, *Arachnis* and *Rhynchostylis* having pendant flower spikes and long dangling roots were found suitable for basket culture (De and Medhi, 2015).

### **5.1.8 Incidence of pest and diseases**

Incidence of pests and diseases was observed on 43 plants of 16 different varieties throughout the study period but could be controlled by suitable remedial measures. The pests and diseases infected the various plant and floral parts. De *et al.* (2014a) reported that the occurrence of bacterial and fungal infections and insect pests which affected the quality of cut flowers by producing higher amounts of ethylene. They also reported that microbes accelerated flower senescence by the plugging of xylem vessels with pectin degraded products, by producing ethylene and toxic compounds. Meera (2012) reported the cataloguing and management of major diseases on monopodial orchids (*Vanda*, *Ascocenda*, *Mokara*, *etc.*).

## 5.2 COMPATIBILITY STUDIES

### 5.2.1 Anthesis and Stigma receptivity

Knowledge regarding anthesis time, stigma receptivity and pollen viability is a prerequisite for a successful compatibility studies and further crop improvement programme. In the present study, eleven hybrids were selected for compatibility studies and observations were made on anthesis time, stigma receptivity, pollen production and viability. Pollen studies were carried out to choose two best male parents for cross compatibility studies and crossing was done using the selected two male parents. Selfing was attempted in all the eleven varieties. Post pollination changes *viz.*, flower fall, enlargement of pedicel, pod development, percentage of pod set, days taken for maturity, seed germination (%) and days for planting out were also recorded.

Anthesis was observed to be slow during the day time but faster during morning and evening time. However, *Ascocentrum* flowers opened very slowly and took almost full day for complete opening of the flower. In the selected eleven *Ascocentrum* hybrids flower opened during morning and evening; flower opening started from 5.30 am and continued up to 11.30 am and from 3.00pm to 6.00 pm. However, in *Vasco. Aroonsri Beauty* anthesis was observed comparatively early and faster (5.30 am to 6.15 am and 4.00 pm to 4.30 pm) compared to the rest of the varieties. According to Varghese (1995), the flower opened acropetally in an inflorescence with maturation initiating from the basal portion and there was floral resupination and the study could find out the peak anthesis period in *Dendrobium* as between 9 am and 10 am and also between 3pm to 4 pm. Sobhana (2000) reported about resupination in *Dendrobium* and also observed flower anthesis time in *Dendrobium* from 7.30 am to 2.30 pm. floral resupination in orchids was also reported by Anon. (2019a).

In the present study, the longest duration of stigma receptivity was found in *Ascda. Kultana* × *V. Bitzs Heartthrob* from 2<sup>nd</sup> to 14<sup>th</sup> day which varied among the 11 different *Ascocentrum* hybrids/varieties. This is in confirmation with In *Phalaenopsis* Croat, (1980) who reported a direct relationship of anthesis time and stigma receptivity in *Phalaenopsis*. Correct time of stigma receptivity was identified by the presence of secretory cell *i.e.*, eleutherocytes (Calder and Slater, 1985). This was confirmed by observing the shiny secretion on stigmatic surface during receptive period. Yeung (1988) observed that a mature stigma of *Epidendrum ibaguense* was covered by a lipid layer at anthesis. Devi and Deka (1992) observed that stigma remained receptive for different days in different orchids; 3 days after anthesis in *Spathoglottis plicata*, 4 days in *Aerides odoratum*, 5 days in *Dendrobium* and 11 days in *Phaius tankervilleae*. Sobhana (2000) reported stigma receptivity after second day of anthesis in *Dendrobium*. Rahi (2017) reported that the maximum number of days of stigma receptivity (3-12 days) was found in *Phalaenopsis* variety Winter Spot and least in Violet (2-5 days).

### **5.2.2 Pollen studies**

Pollen production per pollinium was recorded to be highest in *Ascda. Kultana* × *V. Bitzs Heartthrob* (1,98,611.1) and the minimum pollen production per pollinium was noticed in *Kag. Youthong Beauty* (40,740.7). Prasad *et al.* (1999) reported that pollen production had a positive linear relationship with fruit set. According to Varghese (1995) highest pollen production in *Dendrobium* Kasem White (1,93,750) whereas lowest in *D. Sonia 28 mutant B* (38,282). Sobhana (2000) and Rahi (2017) studied the pollen production, pollen germination and pollen viability in different varieties of *Dendrobium* and *Phalaenopsis* respectively and obtained good results for successful pollination. In the present study, percentage of pollen viability was observed to be highest in *Ascda. Sirichi Fragrance* and *Mok. Sayan* × *Ascda. Doung Porn* (100%). According to Zirkle (1937), pollen grains which stained well, looked plumpy and well-shaped were considered as fertile and those unstained, small or

shrivelled as sterile or non-viable. There are many reports of pollen studies in different orchids by different scientists which is in agreement with the present findings. Pollen viability was found reduced considerably one day after anthesis in *Vanilla* (Nair and Mathew, 1986). Das and Ghoshal (1988) reported a low percentage of pollen fertility in *Dendrobium chrysotoxum* and *D. transparens* and also revealed the tetrad nature of pollen. According to Varghese (1995) maximum pollen viability was observed in the variety *Dendrobium* Kasem White. Rahi (2017) reported highest pollen viability of 100 per cent in *Phalaenopsis*.

In the present study pollen germination was observed to be highest in *Ascda*. Sirichi Fragrance (86.33 %) followed by *Mok*. Sayan × *Ascda*. Doung Porn while the lowest in *Kag*. Youthong Beauty. Varghese (1995) reported pollen germination in *Dendrobium* orchid, using hanging drop technique. There are reports on pollen germination studies using boron. Pollen is generally considered to be deficient in boron, therefore its addition could increase pollen germination and tube growth (O'Kelley, 1955) and Vasil (1960) reported that the boron helps in oxygen uptake, in addition to synthesis of pectic substances required for formation germination tube wall. Similarly, Pollen germination and tube growth were further observed by the addition of 75 ppm boric acid in the medium containing sucrose and agar and was reported in cocoa (Ravindran, 1977) and *Hibiscus* (Markose, 1984).

### **5.2.3 Post pollination changes**

Post pollination changes after selfing were observed. Flower fall was found within a week in *Vasco*. Aroonsri Beauty, *Mok*. Calypso Pink and *Kag*. Youthong. Enlargement of pedicel was not found in *Mok*. Calypso Pink, *Vasco*. Aroonsri Beauty and *Kag*. Youthong Beauty. Pod set was found in *Ascda*. Udomochai, *Ascda*. Kultana × *V*. Bitzs Heartthrob, *Ascda*. Sirichi Fragrance, *Vasco*. Pine River Blue, *Vasco*. Pine River Pink and *Mok*. Sayan × *Ascda*. Doung Porn while the pod development was not observed in *Vasco*. Pine Rivers Fuchsia Delight and *Mok*. Chao Praya Sunset Yellow

Spot. The highest percentage of pod set was found in *Ascda. Kultana* × *V. Bitz* Heartthrob and the lowest of in *Vasco. Pine River Blue* (33.30 %). Minimum number of days was taken for maturity of pods in *Ascda. Udomochai*. However, in *Ascda. Sirichi Fragrance* and *Mok. Sayan* × *Ascda. Doung Porn* pods were not matured fully, they were shrivelled and dried on plant, after the pod development (dried after 2- 3 months). The number of days taken for planting out in *Vasco. Pine River Pink* was 268.3days. Highest percent of seed germination was found in *Vasco. Pine River Pink* (87.26%). Eventhough, protocorms were initiated, they showed declined growth and dried after a while in *Ascda. Udomochai*.

Two varieties, viz., *Ascda. Sirichi Fragrance* and *Mok. Sayan* × *Ascda. Doung Porn* were selected as the male parents based on pollen viability and germination studies for compatibility studies. Flower fall, enlargement of pedicel, pod development, percentage of pod set, days taken for maturity, seed germination and days for planting out were recorded in the crosses with the two male parents and there were 10 different cross combinations. Post pollination behaviour of orchids varied according to compatibility of the plants and also the environmental condition. According to Nair and Mathew (1986) fruit set was noticed even when the flowers were forced to open just before opening and hand pollinated with the pollen of the same flower of *Vanilla*. According to Varghese (1995) the pods of *Dendrobium* hybrids matured in 85-100 days after pollination. Nath (2003) reported that post pollination changes occurred in monopodial orchids viz., flower fall, pedicel swelling, pod development, extent of germination, protocorm development, greening of protocorm, leaf initiation, shoot initiation, root initiation, days for planting out were recorded in different monopodial orchids. Sobhana (2000) reported the post pollination changes in *Dendrobium* sp. and varieties and could observe the shriveling and falling of the developed pods after 1-2 months; she also reported the unsuccessful seed germination of some pods from successful crosses.



In the present study, the varieties *Ascda. Udomochai*, *Ascda. Kultana* × *Vanda Bitzs Heartthrob* and *Vasco. Pine River Pink* were found to be self-compatible and also cross compatible with both male parents. The male parents *Ascda. Sirichi Fragrance* and *Mok. Sayan* × *Ascda. Doung Porn* were also found to be self-compatible and cross compatible with each other. The variety *Vasco. Pine River Blue* was also found to be self-compatible and cross compatible with *Ascda. Sirichi Fragrance* but was found cross incompatible with *Mok. Sayan* × *Ascda. Doung Porn*, the male parent.

The varieties *Kag. Youthong Beauty* and *Vasco. Aroonsri Beauty* were found to be self-incompatible and cross incompatible with both the male parents. However, the rest of the varieties *viz.*, *Vasco. Pine Rivers Fuchsia Delight*, *Mok. Calypso Pink* and *Mok. Chao Praya Sunset Yellow Spot* were found to be self-incompatible but cross compatible with both the male parents. Nath (2003) reported about the self compatibility and cross compatibility in nonparent genera (*Arachnis* and *Vanda*) and parental genera (*Aranda* and *Aranthera*) with *Arachnis*, *Vanda* and *Renanthera* and also stated that if 2-4 months old pods were aborted, the variety could be considered as self or cross compatible. Rahi (2017) reported successful development of pods after hybridization in different *Phalaenopsis* cultivars.

Out of the crosses made, among the ten varieties, seven were found with pod set, but six were fallen at different times after pod set. However, the seedlings from the pods of crosses *Ascda. Udomochai* × *Ascda. Sirichi Fragrance* reached the planting out stage. Orchids have post pollination changes different from other crops. Even if it seemed to be set successfully, with swelling of the ovary, it might not be a successful cross, and may fall after some time. This has been reported by many researchers (Sobhana, 2000; Nath, 2003 and Rahi, 2017). The same phenomenon was noticed in the present study also, hence is in conformity with the findings of Sobhana (2000); Nath (2003) and Rahi (2017).

### 5.3 MOLECULAR CHARACTERIZATION

Assessment of genetic diversity is a prerequisite for genetic improvement of any agricultural/horticultural crops. Knowledge of genetic diversity within the available germplasm is a key for the successful breeding programme in crop improvement, characterization and grouping and also for the conservation. Even though genetic variation can be assessed by morphological parameters, but these are more influenced by environment and thus need not be accurate and reliable. Therefore, genotypic markers are more reliable which are not affected by environment and are more informative. Genus *Ascocentrum* and its hybrids/varieties exhibited broad range of variations and hence it is difficult to evaluate the genetic relationships.

Most of the morphological traits are influenced by environmental factors and many quantitative traits are of polygenic inheritance and expressed only after several years of growth, (Hamrick *et al.*, 1992). As a result, the level and pattern of genetic diversity determined by morphological traits and characterisation are not accurate, although the characterisation should be based on high heritability characters. Molecular markers have greater dependability and utility compared to morphological markers. According to Lim *et al* (1999) markers could be used for indicating the genetic closeness of orchid species and hybrids easily and thus help to predict the outcome of a cross, based on genotypic information.

Molecular characterisation was also utilised for identifying intergeneric hybrids of *Renanthera* and *Vanda* (Kishor and sharma, 2010), ornamental *Vanda* species in Thailand (Tanee *et al.*, 2012), medicinal *Dendrobium* species (Chattopadhyay *et al.*, 2012), hybrids of roses (Che Dai-di *et al.*, 2013) and lily hybrids (Zhao *el al.*, 2014). Genetic constitution of the plants plays important role with respect to the flowering attributes of orchids. Plants showed better performance during the entire period of the present study. Several tropical low land orchid flora

showed year round flowering which was mainly controlled by their genetic constitution as reported by Goh (1984).

### **5.3.1 Genomic DNA isolation**

Molecular characterisation was done in twenty selected hybrids belonging to different intergeneric groups of *Ascocentrum*. Total genomic DNA was isolated from orchid leaves using the procedure suggested by Lim *et al.* (1997).

### **5.3.2 SSR and ISSR Assay**

The DNA samples of 20 selected orchid hybrids were subjected to SSR and ISSR assays and the genomic DNA was amplified with 50 primers (ISSR and SSR) so as to amplify ISSR and SSR regions.

Microsatellite or SSR marker is ubiquitous and widely used but it has a disadvantage that it requires prior knowledge of the sequence to design the primer. ISSR molecular marker system can overcome this limitation as primers are based on a repeat sequence and no prior sequence knowledge was required (Godwin *et al.*, 1997). Molecular characterization is done to distinguish between varieties/hybrids so as to use them in the future breeding programme. The present study was designed to indicate the ISSR based PCR reaction amplify region between the SSRs, giving a multilocus marker system, and SSRs, giving a monolocus marker system molecular markers so as to understand the genetic relationship between 20 selected hybrids of *Ascocentrum*.

Peyachoknagul *et al.* (2014a) reported that SSR primers FJ539056, FJ539057, FJ539058, FJ539059, FJ539060, FJ539061 and FJ539051 were selected for the DNA amplification from 76 vandaceous orchid samples and were selected to evaluate polymorphism and discriminating potential of each primer in 2 and 3 parental genera, *viz.*, *Ascocenda*, *Mokara* and *Kagawara* and another non parent genera *Vanda* and *Rhyncostylis*. To expand their transferability to vandaceous orchids, microsatellite

marker was developed from *Mokara*. Three primers *viz.*, FJ539056, FJ539057 and FJ539058, could also amplify the DNA of all samples from 12 different genera, namely *Mokara*, *Ascocenda*, *Kagawara*, *Ascocentrum*, *Vanda*, *Rhynchostylis*, *Aranda*, *Phalaenopsis*, *Rhynchovanda*, *Renanstylis*, *Rhynchorides* and a hybrid *Arachnostylis* × *Ascocenda*.

Phuekvilai *et al.* (2009) clearly confirmed that the microsatellite markers have high potential in cultivar identification and evaluation of cultivar purity in commercial orchids. Chung and Nason (2007) reported that the microsatellite loci developed for *Cymbidium sinense* might have a broad applicability within the Orchidaceae family, as previous allozyme and random amplified polymorphic DNA markers used in the genus *Cymbidium*.

Peyachoknagul *et al.* (2014) reported that the SSR primers *viz.*, FJ539056, FJ539057, FJ539058, FJ539059, FJ539060, FJ539061 and FJ539051 were having the highest transferability among the 12 different orchid genera. These markers could become powerful tool in identifying several vanda orchids specifically non parental genera like *Ascocentrum*, *Rhynchostylis*, *Vanda*, *etc.* and intergeneric hybrids like *Mokara*, *Ascocenda*, *Kagawara*, *etc.* These markers were also used for evaluating purity in commercial orchid genera and assessing genetic diversity and conservation of different samples. Peyachoknagul *et al.* (2014) also reported that SSR marker FJ539051 could amplify DNA from terete leaf vandaceous orchid.

The high PIC of primers in the present study indicated the highly informative nature of the SSR primers and the diversity of the used population. The PIC value ranged from 0.095 to 0.800. Primers FJ539050 and JN375713 were observed to have the lowest PIC value (0.095), whereas, FJ539054 recorded the highest PIC value (0.800).

Xia *et al.* (2008) confirmed that twenty five microsatellite loci which were isolated from a SSR enhanced genomic library of *Cymbidium sinense*. Nine of these loci displayed genetic diversity that ranged from one to 15 alleles per locus, and they observed heterozygosity of six PIC value, ranged from 0.18 to 0.90. From the results of the study by Ko *et al.* (2017) it was concluded that high PIC value indicated high polymorphism in *Phalaenopsis* genotypes and the presence of a rare allele or alleles at one marker locus and shows the high discriminatory and differentiation power of that marker.

ISSR primer (GACAC)<sub>4</sub> observed was with low PIC value, 0.594; whereas UBC810 was observed with the highest PIC value, 0.926. It was also observed that 17 out of 20 ISSR markers have recorded PIC value more than 0.8. This indicates high discriminatory and differentiation power of these markers for the whole population of orchids used in this study. ISSR based PCR reaction amplifies region between the SSRs, giving a multilocus marker system.

Rahi (2017) reported that the study was designed to harness these characters of ISSR molecular markers so as to find out the genetic relationship between six selected cultivars of *Phalaenopsis*.

The highest Jaccard's similarity value (0.82) for SSR markers was observed between *Kag. Christie Low* and *Vasco. Pine River Pink*. The least Jaccard's similarity value (0.05) was observed in between *Kag. Samrong* and *Ascda. Suksamran Sunlight*, *Ascda. Yip Sum Wah* × *V. JVB* and *Vasco. Aroonsri Beauty*. The highest Jaccard's similarity value (0.44) was observed between *Mok. Khaw Phiak Suan* × *Ascda. Bicentennial Yellow Spot* and *Mok. Chao Praya Sunset Yellow Spot* whereas, the least value (0.03) was between *Vasco. Blue Bay White* and *Mok. Khaw Phiak Suan* × *Ascda. Bicentennial Yellow Spot*, which indicated that these hybrids are dissimilar to themselves.

According to Phuekvilai *et al.* (2009) the genetic relationship of 33 *Vanda* related orchid cultivars had indicated 0.75 similarity index value and formed four different clusters, first cluster was comprised of 25 *Vanda* and 2 *Ascocenda* samples. The second and third clusters were distinctively separated from first cluster, which indicated that *Vasco*. Pine River Blue, *Vanda denisoniana* and *Rhynchovanda* Colmari and *Vanda* Bangkok White had little genetic relationship with other *Vanda* cultivars/species. The fourth cluster was terete *Vanda* consisting two varieties of *Vanda teres* and *Vanda* Miss Joaquim. The fourth cluster was clearly clustered out from the other *Vanda* and related orchid cultivars because terete *Vanda* had little genetic relationship with the strap leaved *Vanda* and had been reclassified in the genus *Papilionanthe* (Sin *et al.*, 2002).

Moraes *et al.* (2014) evaluated the ISSR markers *viz.*, 7, 901, AW3, DAT, GOOFY, MAO, OMAR, UBC814, UBC843 and UBC899 which was effective to analyse genetic diversity in *Cattleya* and obtained high polymorphism rate (96.9 %) and could justify by the use of the three species as well as the different accession of the species.

In the present study UPGMA clustering algorithm grouped the varieties into 2 main clusters. *Kag*. Samrong clustered was different from all others and other members were grouped in another cluster. At 50 per cent level of similarity, the hybrids were grouped into 13 clusters. It was also observed that the ISSR primers showed very high level of diversity, and at 50 per cent level of similarity all 20 hybrids/varieties were grouped into a separate cluster. Hence, the clustering was done with minimum of 30 per cent similarity. At this similarity, all 20 hybrids/varieties were grouped into 14 different clusters. Six clusters observed with two members each, whereas all remaining clusters were observed with only one member in each.

Pillai (2003) reported molecular characterization of fifteen *Dendrobium* varieties was carried out using molecular marker technique and they were grouped into six clusters on drawing a vertical line in the dendrogram at a distance of 0.425.

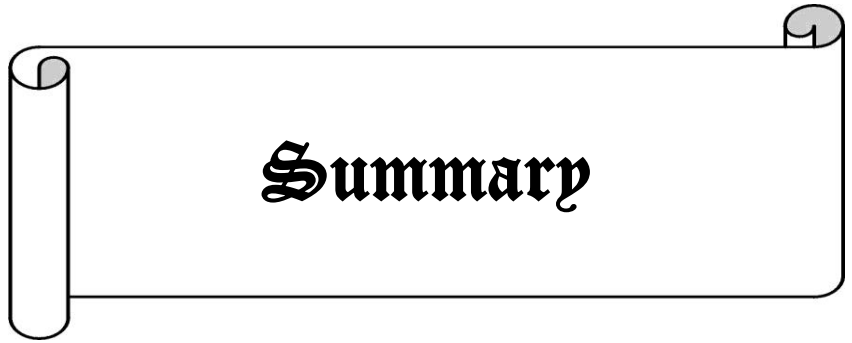
According to Krishnapriya (2005), morphological and cyto-molecular characterization of 12 *Dendrobium* cultivars revealed that morphologically distinct and superior lines were genetically differentiable.

Parab *et al.* (2008) suggested that dendrogram constructed from ISSR marker formed two major clusters and obtained polymorphic bands which helped in better understanding of the genetic profile that can be used to develop strategies for conservation and sustainable utilisation of epiphytic orchid.

Morphological and molecular analysis of genetic variability was investigated by using SDS PAGE and molecular markers in *Vanda tessellata* (Roxb) Hook ex G.Don, an epiphytic orchid from eastern part of Andhra Pradesh (Khasim and Ramesh, 2010).

According to Moraes *et al.* (2014), dendrogram was plotted for germplasm variation based on the dendrogram and genetic distance (GD), in *Cattleya guttata*, *C. leopoldii* and *C. tigrina*; ISSR markers showed high variation among the specimens.

Rahi (2017) reported that ISSR primers were used to distinguish between morphological and molecular characteristics in *Phalaenopsis* and dendrogram indicated variations in different varieties of *Phalaenopsis*.



# Summary





## 6. SUMMARY

The present study on ‘morpho-molecular characterisation of intergeneric hybrids of *Ascocentrum*’ was conducted at the Department of Floriculture and Landscaping, College of Horticulture, Vellanikkara, Thrissur during the period from April 2016 to September 2019. The main objective was to assess the variability at morphological and molecular levels for commercial exploitation.

Thirty tall to intermediate climbing hybrids of *Ascocentrum* were used for the study. The varieties exhibited wide variation with respect to vegetative, floral and molecular characters and also differed in their compatibility pattern. The salient findings are summarized hereunder.

- Maximum plant height was recorded in *Mok. Omayaiy Yellow* followed by *Mok. Sayan × Ascda. Doung Porn* which were found to be significantly superior to all other varieties throughout the study period.
- *Mok. Omayaiy Yellow* also had highest internodal length and leaf breadth. Whereas, plant spread, leaf length and leaf area were maximum in *Kag. Youthong Beauty*.
- *Vasco. Aroonsri Beauty* had the least values in all vegetative characters including plant height, plant spread, shoot, leaf and root characters except the number of leaves which was the least in *Mok. Khaw Phiak Suan × Ascda. Jiraprapa*.
- The number of leaves was observed to be the highest in *Mok. Sayan × Ascda. Doung Porn. Mok. Khaw Phiak Suan × Ascda. Jiraprapa* was observed with the highest root length and *Mok. Chao Praya Sunset Yellow Spot* with the highest root girth.
- *Ascda. Kultana × V. Bitz’s Heartthrob* was found to have maximum peduncle length, flower length, flower width, lip length and lip width, whereas *Vasco. Blue Bay White* produced the highest number of florets/spike.

- In post harvest studies, the variety *Mok. Omayaiy Yellow* recorded highest fresh weight of spike and *Kag. Youthong Beauty* took maximum days to start wilting of floret. Variety *Mok. Chark Kuan Pink* was observed to have longer floret life span, spike longevity and higher water uptake.
- *Mok. Sayan × Ascda. Doung Porn* produced the maximum number of spikes per plant per year. Highest spike length and peduncle length were noticed in *Mok. Omayaiy Yellow*, whereas it was the minimum in *Vasco. Aroonsri Beauty*. Minimum peduncle length was recorded in *Ascda. Kultana × V. Bitzs Heartthrob*.
- *Kag. Samrong* had maximum rachis length, whereas it was minimum in *Mok. Khaw Phiak Suan × Ascda. Jiraprapa*. Highest spike girth was found in *Mok. Chao Praya Sunset Orange*, it was the minimum in *Vasco. Blue Bay White*.
- Maximum pedicel length, floret length, floret width, lip length and lip width were recorded in *Ascda. Kultana × V. Bitzs Heartthrob* whereas, these were the lowest in *Vasco. Blue Bay White*.
- Cluster analysis with 14 different floral characters revealed 12 clusters at 75 per cent similarity. It was observed that cluster 2 and cluster 5 were least similar with each other, whereas, the highest inter-cluster distance was observed in cluster 6 and cluster 10.
- Cluster 6 which included *Ascda. Sirichi Fragrance* and *Vasco Blue Bay White* was found to have the lowest internodal length with the highest value for number of florets per spike, also observed to have lower flower length and flower width. Cluster 10 was found to have the high mean values for spike length, flower length and flower width.
- Cluster analysis revealed that *Vasco. Blue Bay White* had least floret length, floret width and internodal length and with highest number of flowers; this variety also produced compact and dense inflorescence and would be best for indoor display. This variety also had least values in most of the vegetative characters and had the highest leaf longevity; also performed well with respect

to phenological characters; which also add to its value as an indoor display orchid.

- In phenological characters, interval of leaf production was least in *Mok. Sayan × Ascda. Doung Porn* (32.17 days) and the highest in *Mok. Sayan × Ascda. Bangkuntein Gold* (108.44 days). Maximum leaf longevity was observed in *Vasco. Aroonsri Beauty* (292.20 days) and it was the least *Mok. Chao Praya Sunset Yellow Spot* (129.02 days).
- *Vasco. Pine Rivers Fuchsia Delight* was the earliest with respect to opening of the first floret. *Vasco. Pine Rivers Fuchsia Delight* was harvested first at 75 percent opening of florets, whereas *Vasco. Pine River Pink* attained harvestable stage very late. Maximum number of days was taken for the opening of all florets in *Mok. Chark Kuan Pink Blue*, whereas it was minimum in *Ascda. Yip Sum Wah × Vanda JVB*.
- *Mok. Sayan × Ascda. Doung Porn*, *Mok. Chark Kuan Pink* and *Kag. Youthong Beauty* were observed to have spike longevity more than 35 days. Life of individual floret was highest in *Mok. Chark Kuan Pink*, and lowest in *Vasco. Blue Bay White*.
- In post harvest studies fresh weight of spike was highest in *Mok. Omayaiy Yellow* which was significantly superior to the remaining hybrids. *Mok. Chark Kuan Pink* recorded maximum spike longevity in vase among all the selected hybrids. Whereas, *Vasco. Blue Bay White* took the minimum days for spike longevity and also for wilting of first floret.
- Longest life span was observed in *Mok. Chark Kuan Pink* and *Vasco. Blue Bay White* had the shortest life span. *Mok. Omayaiy Yellow* was observed to have maximum water uptake and PLW whereas, the variety *Vasco. Aroonsri Beauty* had least water uptake.
- In qualitative characters two types of growth habits were noticed viz., hanging and prostrate. *Vasco. Aroonsri Beauty* was the only variety with terete leaves;

others had leaf shape such as semi terete, quarter terete, deeply channeled, strap shaped and channeled at base or tip.

- Less variation was observed with respect to leaf texture and they had smooth and rigid leaf surface, leaves with entire leaf margin. Leaf apex was acute in *Ascda*. Yip Sum Wah × *Vanda* JVB and *Vasco*. Aroonsri Beauty and was emarginated in rest of the hybrids.
- Leaf colour and leaf sheath colour was found green and dark green and with the pigmentation (colour changes with maturity) in some varieties.
- Leaf orientation was straight, arching and horizontal in different varieties; leaf base was sheathed and nature of leaf sheath was found membranous, thick and fleshy.
- Root origin was basal and along the stem. Profused growth, branching and a large number of roots were noticed among most of the *Ascocentrum* hybrids/varieties and was found either at base or along the roots. They were cylindrical in all the varieties.
- The roots were mostly greenish grey in all varieties and old roots were found with greyish brown colour in some varieties.
- Five flowering seasons were observed during the two years of study in *Mok*. Sayan × *Ascda* Doung Porn, four were noticed in *Mok*. Sunspot and *Kag*. Youthong Beauty. In the rest of the varieties there were single to three flowering seasons.
- Multiple number of spikes per plant at a time was observed in *Mok*. Omayaiy Yellow and *Mok*. Sayan × *Ascda*. Doung Porn. Three spikes per plant at a time was observed in *Ascda*. Sirichi Fragrance, *Vasco*. Pine Rivers Fuchsia Delight, *Mok*. Sunspot and *Mok*. Omayaiy Orange.
- Regarding the nature of inflorescence, it was dense and lax in different varieties, it arose from lateral position and oriented in arching or erect manner. The florets were faced to all directions.

- In all the varieties, spikes were green in colour except in *Vasco*. Aroonsri Beauty where it was green with brown streaks. High variation was observed with respect to colour of inflorescences like shades of violet, yellow, white, purple, red, pink, orange, green and lavender.
- A negative relation was observed between pigmentation and fragrance; wherever a mild and sweet fragrance was present, pigmentation (colour change during maturation) was not properly observed.
- Petals showed variations in their shape, curvature, petal apex, petal margin and colour. Petal at base, margin, apex region were observed with different colour shades. Different colour patterns, size, shape of petals and colour pattern of lateral sepals were also observed among the varieties.
- Labellum, the modified petal and the most attractive part of an orchid flower was glabrous with bilobed apex. Among the varieties used in the study, labellum showed different colours like white, purple, green, red, violet, yellow and orange, with uniform, shaded and streaked patterns. Column colour was uniform in all the varieties.
- Colour, pigmentation, texture, shape, pattern, , size of florets and arrangement of florets on spike were considered during the visual evaluation for use as a cut flower for commercial exploitation. The highest total mean score in visual evaluation of spike to use as a cut flower was observed in *Mok*. Omayai Yellow (54.6 out of 60).
- Plant quality was rated on the basis of its fullness, growth pattern, visual appearance, flower colour/pigmentation, spike longevity, as well as shape and arrangement of foliage for use in indoor display. Highest mean total score was obtained for the variety *Vasco*. Pine River Pink (53.83) for use in indoor display.
- According to visual evaluation of all quantitative as well as qualitative morphological characters, phenological and post harvest characters, varieties which were free flowering, along with high longevity on plant and in vase,

higher yields and with highest visual scoring are best for commercial exploitation (as cut flower as well as for hanging baskets and pot plants). *Mok. Omayai yellow, Mok. Sayan × Ascda. Doung Porn, Kag. Youthong Beauty, Mok. Chark Kuan Pink and Mok. Chao Praya Sunset Yellow Spot* performed best with respect to vegetative, floral and post-harvest characters and also visual scoring; hence suitable for use as cut flowers.

- The intermediate climbing varieties showed hanging nature of growth while tall climbing varieties were found to grow in prostrate manner.
- Arching orientation of spikes with short stalk and dense nature of spike is a preferable character for indoor display either in the hanging baskets or as pot plants while the erect spike is an added advantage for cut flower purpose. The varieties studied could be grouped into these categories based on the observations.
- Hanging *Asctm.* varieties, with deflexed flowering, short spike length and stalk length and slow growing, with minimum number of florets *Viz., Vasco Blue Bay White, Vasco. Aroonsri Beauty, Ascda. Kultana × V. Bitzs Heartthrob, Vasco. Pine River Pink, Mok. Rassmatozz* are excellent for use in the indoor display.
- Eleven *Ascocentrum* hybrids were selected for compatibility studies. Anthesis time, stigma receptivity and pollen studies were carried out prior to compatibility studies and flower opening was observed during morning time and evening time. Earliest flower opening and short period of anthesis were noticed in *Vasco. Aroonsri Beauty* (5.30 am to 6.15 am and 4.00 pm to 4.30 pm) compared to other varieties.
- The longest duration of stigma receptivity was found in *Ascda. Kultana × V. Bitzs Heartthrob* (2<sup>nd</sup> to 14<sup>th</sup> day) which varied among the selected 11 *Ascocentrum* hybrids.
- Even though pollen production per pollinium was highest in *Ascda. Kultana × Vanda Bitzs Heartthrob*, percentage of pollen viability and germination were

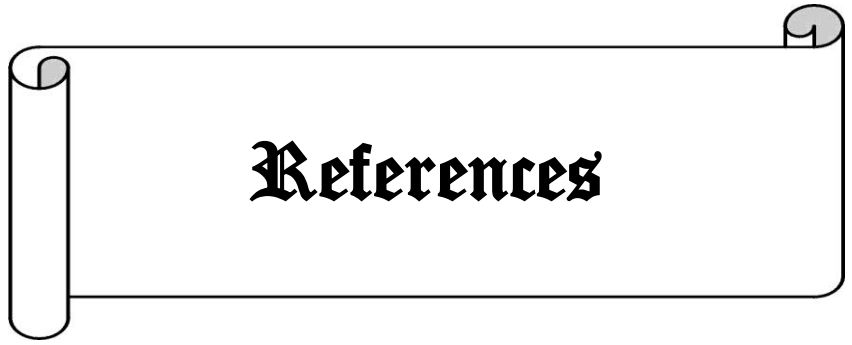
higher in *Ascda. Sirichi Fragrance* and *Mok. Sayan × Ascda. Doung Porn*. Hence these two were selected as male parents for further crossing. Selfing was attempted in all the 11 varieties.

- The selected male parents *Ascda. Sirichi Fragrance* and *Mok. Syan × Ascda. Doung Porn* were found to be self and cross compatible with each other.
- *Ascda. Udomochai, Ascda. Kultana × V. Bitzs Heartthrob* and *Vasco. Pine River Pink* were found to be self-compatible and cross compatible with both the male parents. *Vasco. Pine River Blue* was found cross incompatible with *Mok. Sayan × Ascda. Doung Porn* whereas, rest of the varieties were found cross compatible with both the male parents.
- *Ascda. Kultana × V. Bitzs Heartthrob, Mok. Sayan × Ascda. Doung Porn* and *Ascda. Sirichi Fragrance* were found best with respect to pollen characters. *Ascda. Udomochai, Ascda. Kultana × V. Bitzs Heartthrob* and *Vasco. Pine River Pink* were found best compatibility with both male parents.
- In molecular characterisation ten SSR markers produced polymorphic bands. The number of amplicons detected varied from 2 to 7. Higher number of alleles was found in the FJ539054, FJ539061 and JN375718. Primers DQ501383, DQ501384, DQ501387 and FJ539057 had amplified 4 amplicons. Primers DQ494847 (3) observed to have less number of amplicons.
- One unique band was produced by JN375713 and FJ539050 primers in *Kag. Samrong* and *Vasco. Aroonsri Beauty*, respectively.
- The PIC value ranged from 0.095 to 0.800. Primers FJ539050 and JN375713 were observed to have lower PIC values, whereas, FJ539054 recorded the highest PIC value. Primers DQ501387, JN375718, DQ501383, DQ494847, DQ501384, FJ53906 and FJ539057 also produced good amount of polymorphism.
- The highest Jaccard's similarity value was observed between *Kag. Christie Low* and *Vasco. Pine River Pink*. Whereas, the least Jaccard's similarity value

was observed in between *Kag. Samrong* and *Ascda. Suksamran Sunlight*, *Ascda. Yip Sum Wah* × *V. JVB* and *Vasco. Aroonsri Beauty*.

- The UPGMA clustering algorithm grouped the varieties into 2 main clusters according to dendrogram. *Kag. Samrong* clustered was different from all others and other members were grouped in another cluster. At 50 per cent level of similarity, the hybrids were grouped into 13 clusters.
- The total number of amplicons detected by an individual ISSR primer ranged from 11 to 31. ISSR primer (GACAC)<sub>4</sub> was generated 11 amplicons with 20 varieties/ hybrids, whereas ISSR 901 generated 31 amplicons. Four ISSR primers *viz.*, DAT, MAO, UBC808 and UBC858 were generated more than 25 amplicons. The amplicon size ranged from 126 bp to 2487 bp. Primer ISSR 7 was observed with the least size of amplicon (126 bp-1780 bp) and ISSR primer OMAR with the highest (155 bp-2487 bp.).
- ISSR primer (GACAC) 4 had lowest PIC value, and UBC810, the highest. The highest Jaccard's similarity value was observed between *Mok. Khaw Phiak Suan* × *Ascda. Bicentennial Yellow Spot* and *Mok. Chao Praya Sunset Yellow Spot* whereas, the least was between *Vasco. Blue Bay White* and *Mok. Khaw Phiak Suan* × *Ascda. Bicentennial Yellow Spot*.
- A UPGMA-based dendrogram separated the 20 hybrids into two main clusters each with 10 members. At 30 per cent level of similarity all hybrids were grouped into 14 different clusters. Six clusters were observed with two members each.
- Genetic variation among varieties with SSR and ISSR analyses will be useful for deciding parents in future breeding programmes.
- Both SSR and ISSR marker systems have diversified hybrids. The primers' unique banding pattern can be used for further identification of orchid population and for DNA fingerprinting, as in the unique banding pattern in JN375713 and FJ539050 SSR markers.





**References**



## 7. REFERENCES

- Abraham, A. and Vatsala, P. 1981. *Introduction to Orchids*. Tropical Botanical Garden and Research Institute, Trivandrum, 533p.
- Abraham, A. and Vatsala, P. 1981a. *Introduction to Orchids: With Illustrations and descriptions of 150 South Indian Orchids*. Tropical Botanical Garden and Research Institute, Trivandrum, 250p.
- Agarwal, M., Shrivastava, N., and Padh, H. 2008. Advances in molecular marker techniques and their applications in plant sciences. *Plant Cell Rep.* 27: 617-631.
- Ajibade, S. R., Weeden, N. F., and Michite, S. 2000. Inter simple sequence repeat analysis of genetic relationships in the genus *Vigna*. *Euphytica* 111(1): 47-55.
- Akshata, A. S., Nataraj, S. K., Jadeyegowda, M., Nair, S. A. and Kantharaj, Y. 2018. Morphological characterization of wild orchids of Western Ghats. *J. Farm Sci. Special Issue* 31(5): 618-619.
- Amin, M. M. U., Mollah, M. S., Tania, S. A., Ahmad, M. R., and Khan, F. N. 2004. Performance study of six indigenous epiphytic monopodial orchids of Bangladesh. *J. Biol. Sci.* 4(2): 87-89.
- Anonymous, 2019. <http://www.theplantlist.org/browse/A/Orchidaceae/Ascocentrum/>, [retrieved on July 21, 2019].
- Anonymous, 2019a. <http://www.theplantlist.org/tp11.1/search?q=Vanda>, [retrieved on July 21, 2019].
- Anonymous, 2019b. <http://www.theplantlist.org/1.1/browse/A/Orchidaceae/> [retrieved on July 21, 2019].

- Arditti, J. 1992. *Fundamentals of Orchid Biology*. John Wiley and Sons. Inc., New York, 691p.
- Arora, R. K. 1983. Plant Diversity in the Indian gene centre. In: Paroda, R. S. and Arora, R. K. (eds), *Plant Genetic Resources Conservation and Management Concepts and Approaches*. International Board for Plant Genetic Resources Regional Office for South and Southeast Asia, New Delhi, pp. 245-261.
- Baudino, S., Caissard, J. C., Bergougnoux, V., Jullien, F., Magnard, J. L., Scalliet, G., and Hugueney, P. 2007. Production and emission of volatile compounds by petal cells. *Plant Signaling Behav.* 2(6): 525-526.
- Behera, D., Chandi, C. R., and Mohapatra, U. 2013. Medicinal Orchids in India and their Conservation: A Review. *Floriculture Ornamental Biotechnol.* 7(1): 53-59.
- Bhattacharjee, S. K., Pushkar, S. and Kumar, P. N. 2002. *Passport Data of Flower Crops Germplasm*. AICRP on Floriculture. Technical Bulletin No. 23. ICAR, New Delhi. 210 p.
- Bleasdale, J. K. A. 1973. *Plant Physiology in Relation to Horticulture*. English language book Society and Mac Millan Press Ltd., London, 139p.
- Bose, T. K. and Bhattacharjee, S. K. 1980. *Orchids in India*. Naya Prakash Publishers, Calcutta, West Bengal, India, 538p.
- Bose, T. K., Bhattacharjee, S. K., Das, P., and Basak, U. C. 1999. *Orchids of India*. Naya Prokash, Kolkatta, 487p.
- Brain, R. and Wilma, R., 1979. *Orchids in colour*. Blandford press, New York, 332p.

- Brian, R. and Wilma, R. 2014. *The Practical Illustrated Encyclopedia of Orchids: A Complete Guide to Orchids and their Cultivation*. Southwater Publisher, 431p.
- Briard, M., LeClerc, V., Grzebelus, D., Senalik, D., and Simon, P. W. 2000. Modified protocols for rapid carrot genomic DNA extraction and AFLP analysis using silver stain or radioisotopes. *Plant Mol. Biol. Rep.* 18: 235-241.
- Brown, R. 1833. Observations on the organs and mode of fecundation in Orchideae and Asclepiadeae. Transactions of the Linnean Society of London. *Bot.*, 16: 685-720.
- Bulpitt, C., Li, Y., Bulpitt, F. P. and Wang, J. 2007. The use of orchids in Chinese medicine. *J. Royal Soc. Med.* 63: 100-558.
- Cai, X., Feng, Z., Hou, B., Xing, W., and Ding, X. 2012. Development of microsatellite markers for genetic diversity analysis of *Dendrobium loddigesii* Rolfe, an endangered orchid in China. *Biochemical Syst. Ecol.* 43: 42-47.
- Calder, D. M. and Slater, A. T. 1985. The stigma of *Dendrobium speciosum* Sm. (Orchidaceae): a new stigma type comprising de-tached cells within a mucilaginous matrix. *Ann. Bot.* 55: 297-307.
- Cares-Suárez, R., Poch, T., Acevedo, R. F., Acosta-Bravo, I., Pimentel, C., Espinoza, C., and Botto-Mahan, C. 2011. Do pollinators respond in a dose-dependent manner to flower herbivory?: An experimental assessment in *Loasa tricolor* (Loasaceae). *Gayana. Botánica* 68(2): 176–181.
- Castillo, A., Budak, H., Martin, A., Dorado, G., Borner, A., Roder, M., and Hernandez, P. 2010. Interspecies and intergenus transferability of barley and wheat D-genome microsatellite markers. *Ann. Appl. Biol.* 156: 347-356.

- Chakravarthi, B. K. and Naravaneni, R. 2006. SSR Marker Based DNA fingerprinting and diversity study in rice (*Oryza sativa*. L.). *Afr. J. Biotechnol.* 5: 684-688.
- Chatterji, A. K. 1986. Chromosomes in Orchid Phylogeny and Classification. In: Vij, S. P. (ed.), *Biology, Conservation and Culture of Orchids*. The Orchid Society of India, pp. 181-188.
- Chattopadhyay, P., Banerjee, N and Chaudhary, B. 2012. Genetic characterization of selected medicinal *Dendrobium* (Orchidaceae) species using molecular markers. *J. Biol.* 2(4): 117-125.
- Che Dai-de, Su Cheng -Yuan, Zhang Jin-zhu., Chen Xue, Ma Xue., Wang Na and Li Wen-tian 2013. ISSR Analysis of hybrid descendants of roses. *J. Northeast Agric. University.* 20(1):1-4.
- Chen, J. C., and Fang, S. C. 2016. The long pollen tube journey and *in vitro* pollen germination of *Phalaenopsis* orchids. *Plant Rep.* 29(1–2): 179-188.
- Chen, W. H., Chen, T. M., Fu, Y. M., Hsieh, R. M., and Chen, W. S. 1998. Studies on somaclonal variation in *Phalaenopsis*. *Plant Cell Rep.* 18: 7-13.
- Chen, W. H., Tsai, W. T., Chyou, M. S., Fu, Y. M., Chen, Y. H., Lin, Y. S., and Lin, K.C. 2000. The breeding behaviour of *Phalaenopsis equestris* (Schauer) Rchb.f. *Taiwan Sug.* 47(1): 11-14.
- Chen, X. W. and Jeffrey, J. 2009. *Phalaenopsis*. In: *Flora of China, Volume* Science Press, 25: 478-483.
- Chen, X., Hoon, S., Man, S., Hwa, Y., Kuo, J., Wing, T., and Lin, J. 1999. Amplified fragment length polymorphism analysis of vandaceous orchids. *Plant Sci.* 141: 183-189.

- Chen, Y. Y., Bao, Z. X., Qu, Y., Li, W., and Li, Z. Z. 2014. Genetic diversity and population structure of the medicinal orchid *Gastrodia elata* revealed by microsatellite analysis. *Biochemical Syst. Ecol.* 54: 182-189.
- Chowdhery, H. J. 2001. Orchid diversity in North-East India. *J. Orchid Soc. India* 15: 1-17.
- Christenhusz, M. J. M. and Byng, J. W. 2016. The number of known plants species in the world and its annual increase. *Phytotaxa.* 261(3): 201-217.
- Chung, M. Y. and Nason, J. D. 2007. Spatial demographic and genetic consequences of harvesting within populations of the terrestrial orchid *Cymbidium goeringii*. *Biol. Conserv.* 13(7): 125-137.
- Croat, T. B. 1980. Flowering behavior of the neotropical genus *Anthurium* (Araceae). *Am. J. Bot.* 67: 888-904.
- Darwin, C. R. 1862. The Correspondence of Charles Darwin. *Letter 3560—Darwin, C. R., to More, A. G., 18 May 1862. In: Burkhardt, Frederick, et al., eds. 2003. Vol. 10. Cambridge University Press, Cambridge, 209 p.*
- Dash, P.K., Sahoo, S. and Bal, S. 2008. Ethnobotanical Studies on Orchids of Niyamgiri Hill Ranges, Orissa, India. *Ethnobotanical Leaflets.* 12: 70-78.
- De, L. C. and Debnath, N. G. 2011. *Vision 2030*. NRC for Orchids, Pakyong, Sikkim, 43p.
- De, L. C. and Medhi, R. P. 2015. Orchid: A diversified component of farming systems for profitability and livelihood security of small and marginal farmers. *J. Glob. Biosci.* 4(2): 1393-1406.
- De, L. C., Khan, A. M., Kumar, R., and Medhi, R. P. 2014. Orchid farming a remunerative approach for farmer livelihood. *Int. J. Sci. Res.* 3: 468-471.

- De, L. C., Vij, S. P., and Medhi, R. P. 2014a. Postharvest physiology and technology in orchids. *J. Hortic.* 1: 222-230.
- De, L.C., Rao A.N., Singh, D.R., Dhiman, S.R., Prakash, R. and Singh, R. 2018. DUS Test Guidelines in *Mokara* Orchids. *Int. J. Hortic.* 8(4) 29-35.
- De, L.C., Rao, A.N., Rajeevan, P.K., Srivastava, M. and Chhetri, G. 2015. Morphological Characterization in *Vanda* species. *Int. J. Sci. Res.* 1(4): 26-32.
- Deepa, T. 2017. Evaluation of Fragrant vandaceous orchids for ornamental traits. M. Sc. (Hort) thesis, Kerala Agricultural University, Thrissur, 123p.
- Delle-Vedove, R., Schatz, B., and Dufay, M. 2017. Understanding intraspecific variation of floral scent in light of evolutionary ecology. *Ann. Bot.* 120(1): 1-20.
- Devi, J. and Deka, P. C. 1992. Pollen viability, stigma receptivity and cross compatibility of some Indian orchids. *J. Orchid Soc. India* 6(1-2): 79-84.
- Dharmaraj, K., Ezhilkumar, S., and Dinesh, R. 2018. Studies on varietal identification of rice genotypes using ISSR markers. *J. Pharmacognosy Phytochem.* 7(1): 2808-2812.
- Dharmarathna, T., Herath, V., and Herath, D. 2018. Inter Simple Sequence Repeat markers for analysis of molecular diversity and genetic structure of eighteen *Dendrobium* cultivars in Sri Lanka. *Int. J. Innov. Edu. Res.* 6(3): 91-102.
- Dongre, A., Raut, M., Bhandarkar, M., and Meshram, K. 2011. Identification and genetic purity testing of cotton F<sub>1</sub> hybrid using molecular markers. *Indian J. Biotechnol.* 10(3): 15-20.
- Dressler, R. L. 1993. *Phylogeny and Classification of the Orchid Family*. Cambridge University Press, 314p.

- Dressler, R. L. 1981. *The Orchids, Natural History and Classification*. Harvard University Press, Cambridge, USA, 171p.
- Elliot, J. 1994. *Orchid Growing in the Tropics*. Orchid Society of South East Asia, Time Editions Pte. Ltd., Singapore, 207p.
- Fatimah, and Sukma, D. E. W. I. 2011. Development of sequence-based microsatellite marker for *Phalaenopsis* orchid. *Hayati J. Biosci.* 18(2): 71-76.
- Fatokun, C. A., Danesh, D., Young, N. D., and Stewart, E. L. 1993. Molecular taxonomic relationships in the genus *Vigna* based on RFLP analysis. *Theor. Appl. Genet.* 86: 97-104.
- Ferreira, J. R., Pereira, J. F., Turchetto, C., Minella, E., Consoli, L., and Delatorre, C. A. 2016. Assessment of genetic diversity in Brazilian barley using SSR markers. *Genet. Mol. Biol.* 39(1): 86-96.
- Flach, A., Dondon, R. C., Singer, R. B., Koehler, S., Amaral, M. D. C. E., and Marsaioli, A. J. 2004. The chemistry of pollination in selected Brazilian maxillariinae orchids: Floral rewards and fragrance. *J. Chem. Ecol.* 30(5): 1045–1056.
- Frary, A., Xu, Y., Liu, J., Mitchell, S., Tedeschi, E., and Tanksley, S. 2005. Development of a set of PCR-based anchor markers encompassing the tomato genome and evaluation of their usefulness for genetics and breeding experiments. *Theor. Appl. Genet.* 111: 291-312.
- Frowine, S. A. 2005. *Fragrant Orchids: A Guide to Selecting, Growing, and Enjoying*. Timber Press, United Kingdom, 200p.
- Fu, P. C., Zhang, Y. Z., Ya, H., and Gao, Q. 2016. Characterization of SSR genomic abundance and identification of SSR markers for population genetics in Chinese jujube (*Ziziphus jujuba* Mill.). *Peer J.* 4: 1735p.



- Geetha, C. K., Rajeevan, P. K. and Valsalakumari, P. K. 2009. Diversity analysis in *Arachnis* genotypes under warm humid tropical conditions of Kerala. National Conference on Floriculture for Livelihood and Profitability. (November 16-19, 2009), IARI, New Delhi, *Abstracts* p 39-40.
- George, J. 2019. Response of *Ascocenda* orchid to growth regulator and micronutrients. M.Sc. (Hort.) thesis, Kerala Agricultural University, Thrissur, 46p.
- Godini, A. 1981. Counting pollen grains of some almond cultivars by means of an haemocytometer. *Rivista Di Ortoflorofrutticoltura Italiana*, 65(3): 173-178.
- Godwin, I. D., E. Aitken, A. B., and Smith, L. W. 1997. Application of inter-simple sequence repeat (ISSR) markers to plant genetics. *Electrophoresis* 18: 1524–1528.
- Goh, C. J. 1977. Regulation of floral initiation and development in orchid hybrid *Aranda Deborah*. *Ann. Bot.* 41: 763-769.
- Goh, C. J. 1983. Aerial root production in *Aranda* orchids. National University of Singapore. *Ann. Bot.* 51: 145-147.
- Goh, C. J. 1984. Physiology of flowering in orchids. In: Tan, K. N. (ed.), *Proceedings of the Eleventh World Orchid Conference, Florida*, pp.166-173.
- Goh, C. J. and Arditti, J. 1985. Orchidaceae, In: A.H. Halevy (ed.), *Handb. flowering*: Vol. 1. CRC Press, pp. 309-336.
- Griesbach, R. J. 2005. A Scientific approach to breeding blue orchids: exploring new frontiers in search of elusive flower colours. *Orchids* 74(5): 378-379.

- Grove, D. L. 1995. *Vandas and Ascocendas and their combinations with other genera*. Timber Press, Portland, 241p.
- Gupta, M., Chyi, Y. S., Romero-Severson, J. and Owen, J. L. 1994. Amplification of DNA markers from evolutionarily diverse genomes using single primers of simple-sequence repeats. *Theor. Appl. Genet.* 89: 998-1006.
- Gupta, P. K. and Varshney, R. K. 2000. The development and use of microsatellite markers for genetic analysis and plant breeding with emphasis on bread wheat. *Euphytica* 113(3): 163-185.
- Gupta, S. and Prasad, M. 2009. Development and characterization of genic SSR markers in *Medicago truncatula* and their transferability in leguminous and non leguminous species. *Genome* 52(9): 761-771.
- Hamrick, J. L., Godt, M. J. W., and Sherman-Broyles, S. L. 1992. Factors influencing levels of genetic diversity in woody plant species. *New Forests* 6:95-124.
- Harkema, H. and Struijlaart, P. F. 1989. Effect of amino-oxyacetic acid on coloration of the labellum and longevity of cut cymbidium flowers. *Acta Hortic.* 261: 293-304.
- He, C., Poysa, V., and K. Yu, 2003. Development and characterization of simple sequence repeat (SSR) markers and their use in determining relationships among *Lycopersicon esculentum* cultivars. *Theor. Appl. Genet.* 106(2): 363-373.
- Heikal, A., Hadia, H. S., Abdel-Razzak and Hafez, E. E. 2008. Assessment of genetic relationships among and within cucurbita species using RAPD and ISSR markers. *J. Appl. Sci. Res.* 4: 515-525.
- Heslop-Harrison, Y. 2000. Control gates and micro-ecology: the pollen-stigma interaction in perspective. *Ann. Bot.* 85: 5-13.

- Hew, C. S., Clifford, P. E., and Yong, J. W. H. 1996. Aspects of carbon partitioning in tropical orchids. *J. Orchid Soc. India* 10(1-2): 53-81.
- Hicks, D. M., Ouvrard, P., Baldock, K. C. R., Baude, M., Goddard, A., Kunin, W. E., and Robertson, K. M. 2016. Food for pollinators: quantifying the nectar and pollen resources of Urban Flower Meadows. *PLoS ONE* 11(6): e0158117.
- Ho, C. K. and Chen, C. C. 2003. Moscatillin from the orchid *Dendrobium loddigesii* is a potential anticancer agent. *Cancer Invest.* 21: 729-736.
- Hokanson, S. C., Szewc-McFadden, A., Lamboy, W. F., and Mcferson, J. 1998. Microsatellite (SSR) markers reveal genetic identities, genetic diversity and relationships in a *Malus domestica* Borkh. core subset collection. *Theor. Appl. Genet.* 97: 671-683.
- Holton, T. A., Christopher, J. T., McClure, L., Harker, N., and Henty, R. J. 2002. Identification and mapping of polymorphism SSR markers from expressed sequences of barley and wheat. *Mol. Breed.* 9: 63-71.
- Hossain, M. M. 2011. Therapeutic orchids: traditional uses and recent advances-an overview. *Fitoterapia* 82: 102-140.
- Huang, Y., Li, F. and Chen, K. 2010. Analysis of diversity and relationships among Chinese orchid cultivars using EST-SSR markers. *Biochemical Syst. Ecol.* 38: 93-102.
- Huber, F. K., Kaiser R., Sauter, W., and Schiest, F. P. 2005. Floral scent emission and pollinator attraction in two species of *Gymnadenia* (Orchidaceae). *Oecologia* 142: 564-575.
- Hyde, H. A. and Williams, D. A. 1944. The right word. *Pollen Anal. Circular* 8: 6p.
- Jain, S. K. 1986. Orchid wealth of India. In: Vij, S.P. (ed.), *Biology, Conservation and Culture of Orchids*. The Orchid Society of India, pp. 319-322.

- Jakha, H. Y., Deb, C. R., Jamir, N. S., and Dey, S. 2015. *Arachnis labrosa* var. *zhaoi* (Orchidaceae): A new record for India. *Rheedea*. 25(2): 120-122.
- Jan, S. J. K. 2002. PIC calculator. Available: <http://www.liv.ac.uk/~kempsj/pic.html> [08 October 2013].
- Jonah, P. M., Bello, L. L., Lucky, O., Midau, A., and Moruppa, S. M. 2011. Review: The importance of molecular markers in plant breeding programmes. *Glob. J. Sci. Frontier Res.* 11(5): 5-12.
- Joshi, S. P., Gupta, V. S., Aggarwal, R. K., Ranjekar, P. K., and Brar, D. S. 2000. Genetic diversity and phylogenetic relationship as revealed by inter-simple sequence repeat (ISSR) polymorphism in the genus *Oryza*. *Theor. Appl. Genet.* 100: 1311-1320.
- Kaga, A., Tomooka, N., Egawa, Y., Hosaka, K., and Kamijima, O. 1996. Species relationships in the subgenus *Ceratotropis* (genus *Vigna*) as revealed by RAPD analysis. *Euphytica*. 88: 17-24.
- Kang, J. Y., Lu, J. J., Qiu, S., Chen, Z., Liu, J. J., and Wang, H. Z. 2015. Dendrobium SSR markers play a good role in genetic diversity and phylogenetic analysis of Orchidaceae species. *Sci. Hortic.* 183: 160-166.
- KAU [Kerala Agricultural University]. 2011. *Package of Practices Recommendations: Crops* (14<sup>th</sup> Ed.). Kerala Agricultural University, Thrissur, 360p.
- Kaveriamma, M. M. 2007. Evaluation of monopodial orchids for cut flower. M.Sc. (Hort) thesis, Kerala Agricultural University, Thrissur, 123p.
- Kaveriamma, M. M., Geetha, C. K., Rajeevan, P. K. and Valsalakumari, P. K. 2008. Vegetative and floral diversity in tall climbing monopodials. National Conference on Orchids. Science and Society, Bangalore. April 10-12, 2008. *Souvenir and Abstracts*. p 6.

- Kaveriamma, M. M., Geetha, C. K., Rajeevan, P. K., and Valsalakumari, P. K. 2010. Flowering behaviour of monopodial orchids. In: *Proceedings of National Symposium on Lifestyle Floriculture Challenges and Opportunities* (March 19-21, 2010), ISOH at Y. S. Parmar University of Horticulture and Forestry, Abstracts p. 106.
- Khan, H., M., Belwal, T., Mohd Tariq, Atanasov, A. G., and Devkota, H. P. 2019. Genus *Vanda*: A review on traditional uses, bioactive chemical constituents and pharmacological activities. *J. Ethno. pharmacology* 229: 46-53.
- Khasim, S. M and Ramesh, G 2010. Molecular and morphological studies in *Vanda tessellata*, an epiphytic orchid from Eastern Ghats of India *Acta Hortic.* 878: 63-70.
- Khasim, S. M. and Rao, P. R. M. 1986. Anatomical studies in relation to habitat tolerance in some epiphytic orchids. In: Vij, S.P. (ed), *Biology, Conservation and Culture of Orchids*. The Orchid Society of India, pp. 49-57.
- Kishor, R. K. and Sharma, G. J. 2010 Morphological molecular characterization of Intergeneric Hybrids between the Orchid Genera *Renanthera* and *Vanda*. *Acta Hortic.* 855: 169-178.
- Knudson, L. 1922. Nonsymbiotic germination of orchid seeds. *Bot. Gaz.* 73:1-25.
- Ko, Y. Z., Shih, H. C., Tsai, C. C., Ho, H. H., Liao, P. C., and Chiang, Y. C. 2017. Screening transferable microsatellite markers across genus *Phalaenopsis* (Orchidaceae). *Bot. Stud.* 58(1): 1-9.
- Krishnapriya, M. 2005. Morpho- cytomolecular characterization of *Dendrobium* Sw cultivars MSc, thesis, Kerala Agricultural University, Vellanikkara. 91p.

- Kumar, C. N. P., Somashekar, R. K., Nagaraja, B. C., Shivaprasad, D., and Info, P. 2015. Pollination ecology and reproductive biology of *Canarium strictum Roxb.* from evergreen forests of Central Western Ghats, India. *J. Environ. Biol.* 36: 1131-1136.
- Kumar, P., Pandey, A. K., Rawat, G. S. and Jalal, J. S. 2005. Diversity and conservation of orchids in state of Jharkhan. In: *Plant Taxonomy: Advances and Relevance*. CBS Publication, New Delhi, pp. 345-353.
- Kumar, R., Deka, B. C., and Roy, A. R. 2012. Evaluation of Orchid species under Sub-tropical Mid-hills of Meghalaya. *Hortic. Flora Res. Spectrum* 1(1): 24-28.
- Kumaria, S. and Tandon, P. 2010. Orchids: the world's most wondrous plants. Available at: <http://dspace.nehu.ac.in/handle/1/2009> [Accessed on 1 Nov. 2019].
- Lawler, L. J. 1984. Ethnobotany of the Orchidaceae. In: *Orchid Biology: Reviews and Perspectives, III<sup>ed</sup>*. Comstock Publishing Associates - a division of Cornell University Press, pp. 101-130.
- Lehnebach, C. and Riveros, M. 2003. Pollination biology of the Chilean endemic orchid *Chloraea lamellata*. *Ann. Bot.* 93: 773-781.
- Leite, C. A., Ito, R. M., Lee, G. T. S., Ganelevin, R., and Fagnani, M. A. 2008. Light spectrum management using colored nets to control the growth and blooming of *Phalaenopsis*. *Acta Hortic.* 770: 177-184.
- Lenz, L.W. and Wimber, D. E. 1959. Hybridization and inheritance in orchids. In Withner, C. L. (ed.), *The Orchids: A Scientific Survey*. The Ronald Press Company Ltd., New York, pp. 261-313.
- Li, C., Fatokun, C. A, Ubi, B., Singh, B., and Scoles, G. J. 2001. Determining genetic similarities and relationships among cowpea breeding lines and cultivars by microsatellite markers. *Crop Sci.* 41(1): 189-197.

- Libby, W. J., Steller, R. F., and Seitz, F. W. 1969. Forest genetics and forest breeding. *Ann. Rev. Genet.* 3: 469-494.
- Lim, S. H., Liew, C. F., Lim, Y. H., Lee, Y. H., and Goh, C. J. 1997. A simple and efficient method of DNA isolation from orchid species and hybrids. *Biologia. Planta.* 41(2): 313-316.
- Lim, S. H., Tang, P. C. P., Lee, Y. H., Croh, C. J. 1999. RAPD analysis of some species in the genus *Vanda* (Orchidaceae). *Ann. Bot.* 83: 193-196.
- Litt, M. and Luty, J. A. 1989. A hypervariable microsatellite revealed by *in vitro* amplification of a dinucleotide repeat within the cardiac muscle actin gene. *Am. J. Hum. Genet.* 44: 397-401.
- Loksha, R. and Vasudeva, R. 1994. Do floral features determine the endangered status of Indian. Orchids. *J. Orchid Soc. India.* 8(1): 53-54.
- Lopez Roberts, M., Almeida, P. R., Oliveira, E., and van den Berg, C. 2012. Microsatellite marker development for the threatened orchid *Masdevallia solomonii* (Orchidaceae). *Am. J. Bot.* 8: 66-99.
- Lopez, R. G. and Runkle, E. S. 2004. *The flowering of orchids* [on-line]. Available:<http://www.hrt.msu.edu/faculty/Runkle/Orchid/Articles/AOS%20Lopez%20and%20Runkle.pdf>. [26 Sep. 2019].
- Lopez, R. G. and Runkle, E. S. 2005. Environmental physiology of growth and flowering of orchids. *Hortic. Sci.* 40(7): 1969-1973.
- Lu, J., Hu, X., Liu, J., and Wang, H. 2011. Genetic diversity and population structure of 151 *Cymbidium sinense* cultivars. *Int. J. Hortic. Floriculture.* 7(8): 001-011.
- Markose, B. L. 1984. Pollen production, fertility and compatibility studies in shoe flower (*Hibiscus Rosa Sinensis* L). M.Sc. (Ag) thesis, Kerala Agricultural University, Thrissur, 130p.

- McCouch, S. R., Chen, X., Panaud, O., Temnykh, S., Xu, Y., Cho, Y. G., and Blair, M. 1997. Microsatellite marker development, mapping and applications in rice genetics and breeding. *Plant Mol. Biol.* 35(1): 89-99.
- Meera, T. M. 2012. Cataloguing and management of major diseases of monopodial orchids. M.Sc. (Ag) thesis, Kerala Agricultural University, Thrissur, 90p.
- Mehra, P. N. and Vij, S. P. 1974. Some observations on the ecological adaptations and distribution pattern of the East Himalayan Orchids. *Am. Orchid Soc. Bull.* 43(4): 301-315.
- Melendez-Ackerman, E. J. and Ackerman, J. D. 2001. Density - dependent variation in reproductive success in a terrestrial orchid. *Plant Syst. Evol.* 227(1-2): 27-36.
- Meyer, W., Mitchell, T. G., Freedman, E. Z., and Vilgays, R, 1993. Hybridization probes for conventional DNA fingerprinting used as single primers in the polymerase chain reaction to distinguish strains of *Cryptococcus neoformes*. *J. Clinical Microbiol.* 31: 2274-2280.
- Moraes, M. C., Bertao, M. R., Loose, P. V, Cordeiro, A. F, and Palmieri, D.A. 2014. Molecular study on endemic *Cattleya* species from Brazilian flora. *Am. Int. J. Biol.* 2(3): 77-84.
- Mukherjee, S. K. 1983. *Orchids*. Indian Council of Agricultural Research, New Delhi, 102p.
- Mukherjee, S. K. 1990. *Orchids*. Indian Council of Agricultural Research, New Delhi, 94p.
- Nair, P. C. S. and Mathew, L. 1986. Observations on the floral biology and fruit set in Vanilla. *Agric. Res. J. Kerala* 24(2): 46-47.



- Nash, N. and Frownie, S. 2008. *Complete Guide to Orchids*. Meredith Publishing Group, 122p.
- Nath, N. C. 2003. Compatibility studies in monopodial orchids. M.Sc. (Hort) thesis, Kerala Agricultural University, Thrissur, 100p.
- Ng, W. L. and Tan, S. G. 2015. Inter-Simple Sequence Repeat (ISSR) Markers: Are We Doing It Right? *ASM Sci. J.* 9(1): 30-39.
- Niimoto, D. H. and Sagawa, Y. 1961. Ovule development in *Dendrobium*. *Am. Orchid Soc. Bull.* 30: 813-819.
- Oberle, G. D. and Goertzen, K. L. 1952. A method for evaluating pollen production of fruit varieties. *Proc. Am. Soc. Hortic. Sci.* 59: 263-265.
- O'Kelley, J. C. 1955. External carbohydrates in growth and respiration of pollen tubes *in vitro*. *Am. J. Bot.* 42: 322-327.
- Panaud, O., Chen, X., and McCouch, S. R. 1995. Frequency of microsatellite sequences in rice (*Oryza sativa* L.). *Genome* 38(6): 1170-1176.
- Parab, G. V., Krishnan, S., Janarthnam, M. K., Sivaprakash, K. R. and Parida, A. 2008. Short communication ISSR and RAPD markers assessed genetic variation of *Aerides maculosum* an epiphytic orchid from Goa, India. *J. Plant Biochem. Biotechnol.* 17(1): 107-109.
- Peyachoknagul, S., Mongkolsiriwatan, C., Wannapinpong, S., Huehne, P. S., and Srikulnath, K. 2014. Identification of native *Dendrobium* species in Thailand by PCR-RFLP of rDNA-ITS and chloroplast DNA. *Sci. Asia* 40(2): 113-120.
- Peyachoknagul, S., Nettuwakul, C., Phuekvilai, P., Wannapinpong, S., and Srikulnath, K. 2014a. Development of microsatellite markers of vandaceous orchids for species and variety identification. *Genet. Mol. Res.* 13(3): 5441-5445.

- Phuekvilai, P., Pongtongkam, P., and Peyachoknagul, S. 2009. Development of microsatellite markers for Vanda orchid. *Kasetsart J. Nat. Sci.* 43: 497–506.
- Pijl, L. and Dodson, C. H. 1966. *Orchid Flowers: Their Pollination and Evolution*. Coral Gables, Fla: Published jointly by the Fairchild Tropical Garden and the University of Miami Press, 214p.
- Pillai, N. P. 2003. Morpho-anatomical and molecular characterization of *Dendrobium* Sw cultivars Ph.D (Hort) thesis, Kerala Agricultural University, Thrissur.122p.
- Powell, W., Morgante, M., Doyle, J. J., McNicol, J. W., Tingey, S. V., and Rafalski, A. J. 1996. Genepool variation in genus *Glycine* subgenus Soja revealed by polymorphic nuclear and chloroplast microsatellites. *Genet.* 144: 793-803.
- PPV & FRA. 2012. Guidelines for conduct of test for distinctiveness, uniformity and stability on orchid (*Vanda Jones ex R. Br.*). PPV & FR Authority, Gov. India, New Delhi, 54-58p.
- PPV & FRA. 2012a. Guidelines for conduct of test for distinctiveness, uniformity and stability on *Dendrobium*. PPV & FR Authority, Gov. India, New Delhi, 120p.
- Prasad, P. V. V., Craufurd P. Q., and Summerfield, R. J. 1999. Fruit number in relation to pollen production and viability in groundnut exposed to short episodes of heat stress. *Ann. Bot.* 84: 381-386.
- Pridgeon, A. M., Cribb, P., Chase, M. W. and Rasmussen, F. N. 2001. *Genera Orchidacearum: Epidendroideae*. Oxford University Press, 464p.
- Raguso, R. A. 2008. Wake up and smell the roses: the ecology and evolution of floral scent. *Ann. Rev. Ecol. Evol. Syst.* 39(1): 549-569.

- Rahi, D. 2017. Induction of genetic variability in Phalaenopsis orchids through hybridization and embryo culture. M.Sc. (Ag) thesis, Kerala Agricultural University, Thrissur, 93p.
- Raina, S. N., Rani, V., Kojima, T., Ogihara, Y., Singh, K. P., and Devarumath, R. M. 2001. RAPD and ISSR fingerprints as useful genetic markers for analysis of genetic diversity, varietal identification, and phylogenetic relationships in peanut (*Arachis hypogaea*) cultivars and wild species. *Genome* 44(5): 763-772.
- Rajeevan, P. K. 1995. The scenario of orchid industry in Kerala: Retrospective of a decade. *J. Orchid Soc. India* 9(1-2): 1-5.
- Rajeevan, P. K. 2003. *Orchids*. In: Chadha, K. L. (ed.), *Handbook of Horticulture*. I.C.A.R., New Delhi, pp. 573-577.
- Rajeevan, P. K., Geetha, C. K. and Kaveramma, M. M. 2009. Adorn monopodial orchids. *Indian Hortic.* 54(1): 15-18.
- Rajeevan, P. K., Geetha, C. K. and Kaverramma, M. M. 2009a. For long lasting aesthetic beauty: Adorn monopodial orchid. *Indian Hortic.* 15: 1-19.
- Rajeevan, P. K., Sobhana, A. Bhaskar, J., Swapna, S. and Bhattacharjee, S. K. 2002. *Orchids*. AICRP on Floriculture, Technical Bulletin No.22, ICAR, New Delhi. 62p.
- Rajeevan, P. K., Sobhana, A., Bhaskar, J., Swapna, S., and Bhattacharjee, S. K. 2002. *Orchids*. All India Co-ordinated Research Project on Floriculture, Indian Agricultural Research Institute, New Delhi, 62p.
- Rani, L. C. 2002. Intra and interspecific hybridization in *Dendrobium* sp. Ph.D. thesis, Kerala Agricultural University, Thrissur, 360p.
- Rao, V. N. M. and Khader, J. B. M. 1962. Estimation of pollen production in fruit crops. *Madras Agric. J.* 49(5):152-156.

- Ravindran, P. N. 1977. *In vitro* germination and growth of cacao pollen. *J. Plant. Crops* 5: 109-111.
- Reddy, P. M., Sarla, N., and Siddiq, E. A. 2002. Inter simple sequence repeat (ISSR) polymorphism and its application in plant breeding. *Euphytica* 128: 9-17.
- Rhodehamel, W. A. 1994. Pollination of orchid flowers. *Am. Orchid Soc. Bull.* 63: 534-539.
- Rice, R. 2008. *Renanthera* of Borneo. *Malesian Orchid J.* 1: 71-76.
- Rittershausen, W. and Rittershausen, B. 1999. *Orchids*. Quadrille Publishing Ltd., Great Britain, 224p.
- Saghai Maroof, M. A., Biyashev, R. M., Yang, G. P., Zhang, Q., and Allard, R. W. 1994. Extraordinarily polymorphic microsatellite DNA in barley: species diversity, chromosomal locations, and population dynamics. *Proc. Nat. Acad. Sci. USA* 91: 5466-5470.
- Sankar, I. B., Rao, K. M., and Krishna, A. G. 2010. Prediction of heat transfer coefficient of steel bars subjected to Tempcore process using nonlinear modeling. *Int. J. Adv. Manufacturing Technol.* 47(9-12): 1159-1166.
- Sarikamis, G., Yanmaz, R., Ermis, S., Bakir, M., and Yuksel, C. 2010. Genetic characterisation of pea (*Pisum sativum*) germplasm from Turkey using morphological and SSR markers. *Genet. Mol. Res.* 9(1): 591-600.
- Sebastian, M. 2015. Evaluation of *Vanda* orchid for commercial traits. M. Sc. (Hort) thesis, Kerala Agricultural University, Thrissur, 120p.
- Semagn, K., Bjornstad, A., and Ndjioudjop, M. N. 2006. An overview of molecular marker methods for plants. *Afr. J. Biotechnol.* 5: 2540-2568.
- Sharma, M. D., Dhobal, U., Singh, P., Kumar, S., Gaur, K., Singh, S. P., Jeena, A. S., Koshy, E. P., and Kumar, S. 2014. Assessment of genetic

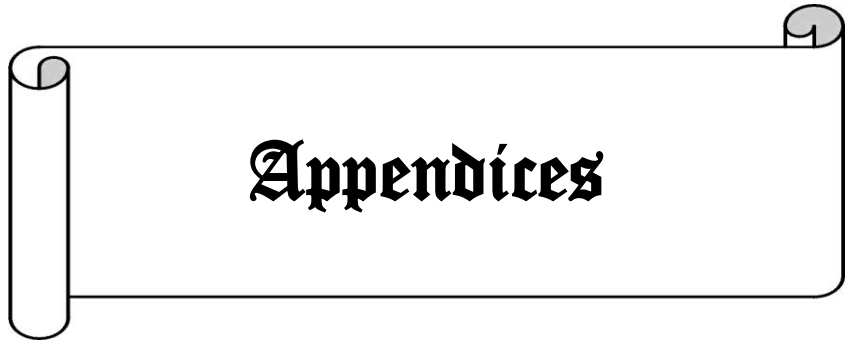
- diversity among sugarcane cultivars using novel microsatellite markers. *Afr. J. Biotechnol.* 13(13): 1444-1451.
- Sharma, V., Rana, M., Katoch, M., Sharma, P. K., Ghani, M., Rana, J. C., and Chahota, R. K. 2015. Development of SSR and ILP markers in horsegram (*Macrotyloma uniflorum*), their characterization, cross-transferability and relevance for mapping. *Mol. Breed.* 35(4): 102-110.
- Simioniuc, D., Uptmoor, R., Friedt, W., and Ordon, F. 2002. Genetic diversity and relationships among pea cultivars revealed by RAPDs and AFLPs. *Plant Breed.* 121: 429-435.
- Sin, H. C., Wing, Y. T., and Arditti, J. 2002. *Biology of Vanda Miss Joaquim* (1<sup>st</sup> ed.). National University of Singapore Press, Singapore.
- Singh, F. 1990. Indian orchids. *Indian Hort.* 35(1): 14-16.
- Singh, F. 1991. Enchanting orchids. *Vatika* 1(3): 9-14.
- Singh, S. S. 2019. India is home to 1,256 species of orchid, says first comprehensive survey. Available at: <https://www.thehindu.com/sci-tech/energy-and-environment/india-is-home-to-1256-species-of-orchid-says-first-comprehensive-survey/article28429797.ece> [Accessed on 10 October 2019].
- Sobhana, A. 2000. Improvement of *Dendrobium* through hybridization and *in vitro* mutagenesis. Ph.D. (Hort) thesis, Kerala Agricultural University, Thissur, 230p.
- Soon, T. E. 1980. *Asian Orchids*. Times Books International, Singapore, 287p.
- Souframanien, J. and Gopalakrishna, T. 2004. A comparative analysis of genetic diversity in blackgram genotypes using RAPD and ISSR markers. *Theor. Appl. Genet.* 109(8): 1687-1693.

- Stanford, W. W. 1971. The flowering time of West African orchids. *Bot. J. Linnear Soc.* 64: 163-181.
- Stpiczyńska, M. 2003. Floral longevity and nectar secretion of *Platanthera chlorantha* (Custer) Rchb. (Orchidaceae). *Ann. Bot.* 92(2): 191-197.
- Sudeep, H.P., and Seetharamu, G. K., Aswath, C., Munikrishnappa, P. M., Sreenivas, K. N., Basavaraj, G. and Gowda, D. M., 2018. Standardization of embedding media and drying temperature for superior quality of dry orchid flower var. Sonia 17. *Int. J. Pure App. Biosci.* 6(2): 69-73.
- Tanee, T., Chadmuk, P., Sudmoon, R., Chaveerach, A and Noikotr, K. 2012. Genetic analysis for identification, genomic template stability in hybrids and barcodes of the *Vanda* species (Orchidaceae) of Thailand. *Afr.J. Biotechnol.* 11:11772-11781.
- Tanksley, S. D., Ganal, M. W., Prince, J. P., de Vicente, M. C., Bonierbale, M. W., Broun, P., and Martin, G. B. 1992. High density molecular linkage maps of the tomato and potato genomes. *Genet.* 132(4): 1141-1160.
- Tatsuzawa, F., Saito, N., Seki, H., Yokoi, M., Yukawa, T., Shinoda, K., and Honda, T. 2004. Acylated anthocyanins in the flowers of *Vanda* (Orchidaceae). *Biochem. Syst. Ecol.* 32(7): 651-664.
- Tautz, D. 1989. Hyper variability of simple sequences as a general source for polymorphic DNA markers. *Nucleic Acids Res.* 17: 6463-6471.
- Tchinda, N. D., Wanjala, B. W., Muchugi, A., Fotso, Nzweundji, G., Ndoumou, D. O., and Skilton, R. 2016. Genetic diversity and gene flow revealed by microsatellite DNA markers in some accessions of African Plum (*Dacryodes edulis*) in Cameroon. *Afr. J. Biotechnol.* 15(13): 511-517.
- Thomas, B. and Rani, C. L. 2008. Assessment of floral characters in commercial varieties of monopodial orchids. *J. Ornamental Hortic.* 11(1): 15-20.

- Tian, H. Z., Han, L. X., Zhang, J. L., Li, X. L., Kawahara, T., Yukawa, T., and Chung, M. Y. 2018. Genetic diversity in the endangered terrestrial orchid *Cypripedium japonicum* in East Asia: Insights into population history and implications for conservation. *Sci. Rep.* 8(1): 6467.
- Tungalag, M., Ariungerel, M., Otgonbayar, B., and Myagmarsuren, Ya. 2018. Varietal identification study of six wheat varieties using ISSR markers. *Mongolian J. Agric. Sci.* 23(01): 14-17.
- Ueno, S., Rodrigues, J. F., Alves-Pereira, A., Pansarin, E. R., and Veasey, E. A. 2015. Genetic variability within and among populations of an invasive, exotic orchid. *AoB Plants* 7: 55-55.
- Varghese, S. 1995. Floral biology and compatibility studies in *Dendrobium*. M. Sc. (Hort) thesis, Kerala Agricultural University, Thrissur, 76p.
- Vasil, I. K. 1960. Studies on pollen germination of certain cucurbitaceae. *Bot.* 47(4): 239-247.
- Warren, R. 1981. Orchids from seed-Part I: Pollination. *Orchid Rev.* 89: 103-105.
- WCSP, 2019. 'World Checklist of Selected Plant Families'. Facilitated by the Royal Botanic Gardens, Kew. Published on the Internet; <http://wmsp.science.kew.org/> [Retrieved 21 July 2019].
- Wikipedia, 2019. Vanda. Available at: [https://en.wikipedia.org/wiki/Vanda#cite\\_note-4](https://en.wikipedia.org/wiki/Vanda#cite_note-4) [Accessed on 1 Nov. 2019].
- Williams, J. G. K., Kubelik, A. R., Lavak, K. J., Rafalski, J. A., and Tingey, S. V. 1990. DNA polymorphism amplified by arbitrary primers are useful as genetic markers. *Nucleic Acids Res.* 18: 6531-6535.
- Wu, K. S., Jones, R., Danneberger, L., and Scolnik, P. A. 1994. Detection of microsatellite polymorphisms without cloning. *Nucleic Acids Res.* 22: 3257-3258.

- Xanthopoulou, A., Ganopoulos, I., Kalivas, A., Nianiou-Obeidat, I., Ralli, P., Moysiadis, T., Tsaftaris, A., and Madesis, P. 2015. Comparative analysis of genetic diversity in Greek Genebank collection of summer squash (*Cucurbita pepo*) landraces using start codon targeted (SCoT) polymorphism and ISSR markers. *Aust. J. Crop Sci.* 9(1): 14-21.
- Xia, K., Ye, X. and Zhang, M. 2008. Isolation and characterization of nine microsatellite markers for *Cymbidium sinense*. *Hortic. Sci.* 43(6):1925-1926.
- Xu, Y. and Crouch, J. H. 2008. Marker-assisted selection in plant breeding: from publications to practice. *Crop Sci.* 48(2): 391-407.
- Yokoi, M. 1975. Colour and pigment distribution in the cultivars of selected ornamental plants, with special reference to their contribution to the ornamental value of plants. *Transactions Fac. Hortic. Chiba Univ.* 14: 20-28.
- Yong, J. W. H. and Hew, C. S. 2004. *The Physiology of Tropical Orchids in Relation to the Industry*. World scientific, 370p.
- Zhao, R., Fan, J. P., Zhang, H. and Liu, Y. B. 2014 Analysis on genetic relationships in lily hybrids based on ISSR molecular markers. *Acta Hortic.* 1035: 215-222.
- Zhao, X. and Kochert, G. 1993. Phylogenetic distribution and genetic mapping of a (GGC)<sub>n</sub> microsatellite from rice (*Oryza sativa* L.). *Plant Mol. Biol.* 21(4): 607-614.
- Zietkiewicz, E., Rafalski, A., Labuda, D. 1994. Genome fingerprinting by simple sequence repeat (SSR)-anchored polymerase chain reaction amplification. *Genomics* 20: 176-183.
- Zirkle, C. 1937. Acetocarmine mounting KKmedia. *Sci.* 85: 528.





# Appendices



**APPENDIX I. Plant height of *Ascocentrum* hybrids/varieties during the period 2016-17, cm**

Var.No.	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	17.26 (4.256)	18.20 (4.368)	19.80 (4.54)	20.76 (4.645)	22.20 (4.797)	22.40 (4.821)	23.30 (4.915)	23.40 (4.925)	24.24 (5.009)	24.86 (5.071)	25.60 (5.144)	26.34 (5.217)
V <sub>2</sub>	15.76 (4.082)	16.66 (4.189)	17.36 (4.27)	18.28 (4.375)	19.16 (4.47)	19.84 (4.543)	20.34 (4.599)	20.64 (4.634)	21.18 (4.69)	21.62 (4.737)	22.18 (4.795)	22.74 (4.853)
V <sub>3</sub>	24.00 (4.98)	24.70 (5.045)	25.60 (5.125)	26.60 (5.222)	27.90 (5.34)	28.72 (5.419)	29.40 (5.483)	30.00 (5.536)	30.62 (5.592)	31.02 (5.628)	31.60 (5.68)	28.20 (5.401)
V <sub>4</sub>	41.20 (6.408)	43.20 (6.58)	44.20 (6.664)	45.00 (6.727)	46.30 (6.828)	47.20 (6.894)	47.90 (6.947)	48.50 (6.993)	48.88 (7.019)	49.68 (7.075)	50.02 (7.099)	50.48 (7.131)
V <sub>5</sub>	19.40 (4.502)	20.70 (4.647)	21.28 (4.711)	22.28 (4.813)	22.92 (4.882)	22.76 (4.863)	22.70 (4.857)	23.80 (4.969)	24.52 (5.042)	25.18 (5.109)	25.78 (5.167)	26.54 (5.239)
V <sub>6</sub>	19.00 (4.467)	20.00 (4.578)	21.34 (4.723)	22.16 (4.81)	22.80 (4.877)	22.70 (4.865)	23.90 (4.988)	24.10 (5.008)	24.66 (5.064)	25.42 (5.139)	26.12 (5.207)	26.66 (5.259)
V <sub>7</sub>	17.60 (4.309)	18.70 (4.436)	20.20 (4.599)	21.44 (4.73)	22.50 (4.841)	22.70 (4.862)	23.56 (4.951)	23.90 (4.985)	25.00 (5.095)	25.68 (5.16)	26.62 (5.249)	27.02 (5.287)
V <sub>8</sub>	29.90 (5.557)	31.30 (5.68)	31.90 (5.732)	33.00 (5.828)	33.50 (5.87)	33.80 (5.895)	34.60 (5.963)	34.60 (5.963)	35.38 (6.028)	35.88 (6.07)	36.52 (6.123)	37.10 (6.17)
V <sub>9</sub>	8.70 (3.107)	11.20 (3.458)	10.68 (3.4)	11.80 (3.563)	13.00 (3.729)	14.10 (3.881)	14.70 (3.958)	15.02 (3.996)	15.70 (4.081)	16.40 (4.166)	16.96 (4.233)	17.62 (4.31)
V <sub>10</sub>	20.00 (4.559)	21.20 (4.69)	21.86 (4.76)	23.00 (4.869)	23.86 (4.957)	24.56 (5.024)	25.92 (5.15)	26.68 (5.225)	27.42 (5.296)	27.86 (5.338)	28.36 (5.384)	28.88 (5.431)
V <sub>11</sub>	10.20 (3.342)	11.40 (3.517)	12.12 (3.618)	12.92 (3.726)	13.78 (3.837)	13.92 (3.855)	14.66 (3.948)	15.50 (4.054)	16.06 (4.124)	16.44 (4.169)	17.06 (4.243)	17.52 (4.296)
V <sub>12</sub>	27.10 (5.295)	28.60 (5.435)	29.20 (5.49)	30.50 (5.608)	31.50 (5.696)	31.80 (5.722)	32.20 (5.756)	32.60 (5.792)	33.52 (5.871)	34.06 (5.918)	34.50 (5.955)	35.32 (6.024)
V <sub>13</sub>	26.60 (5.244)	27.40 (5.32)	27.80 (5.359)	28.60 (5.431)	29.80 (5.542)	31.00 (5.649)	31.62 (5.703)	31.78 (5.717)	32.24 (5.757)	32.74 (5.801)	33.36 (5.854)	33.46 (5.862)
V <sub>14</sub>	16.60 (4.181)	17.80 (4.325)	19.20 (4.477)	20.30 (4.603)	20.64 (4.641)	21.16 (4.696)	21.90 (4.772)	22.70 (4.855)	23.44 (4.931)	23.76 (4.963)	24.50 (5.038)	25.36 (5.121)
V <sub>15</sub>	18.60 (4.387)	19.62 (4.504)	20.00 (4.547)	20.60 (4.613)	21.38 (4.7)	21.90 (4.754)	23.10 (4.874)	23.60 (4.928)	24.20 (4.989)	24.54 (5.02)	25.10 (5.076)	25.52 (5.118)
V <sub>16</sub>	15.80 (4.059)	16.76 (4.172)	17.74 (4.282)	18.90 (4.406)	19.80 (4.508)	20.18 (4.55)	20.78 (4.615)	21.16 (4.656)	21.92 (4.735)	22.40 (4.783)	23.40 (4.887)	24.18 (4.966)

**APPENDIX I. continue**

Var. No.	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	16.70 (4.188)	18.20 (4.36)	18.80 (4.435)	19.16 (4.477)	20.20 (4.593)	21.10 (4.689)	21.22 (4.702)	21.56 (4.739)	22.04 (4.79)	22.58 (4.846)	23.14 (4.905)	23.64 (4.956)
V <sub>18</sub>	23.60 (4.957)	24.44 (5.041)	24.50 (5.047)	25.50 (5.145)	26.50 (5.241)	27.48 (5.333)	28.36 (5.414)	28.62 (5.439)	29.52 (5.519)	29.98 (5.561)	30.44 (5.602)	31.14 (5.663)
V <sub>19</sub>	41.44 (6.513)	42.18 (6.571)	42.88 (6.623)	43.98 (6.706)	44.84 (6.77)	45.04 (6.785)	45.72 (6.835)	46.56 (6.896)	47.54 (6.966)	48.38 (7.026)	49.30 (7.091)	50.30 (7.16)
V <sub>20</sub>	38.60 (6.288)	39.48 (6.357)	40.08 (6.403)	40.76 (6.457)	41.58 (6.518)	42.20 (6.565)	42.92 (6.62)	43.82 (6.687)	45.02 (6.774)	45.40 (6.803)	46.56 (6.888)	47.54 (6.958)
V <sub>21</sub>	50.90 (7.2)	51.76 (7.258)	52.14 (7.284)	54.95 (7.47)	56.36 (7.564)	57.26 (7.623)	58.00 (7.673)	58.66 (7.716)	59.72 (7.784)	60.66 (7.844)	61.56 (7.902)	62.40 (7.955)
V <sub>22</sub>	50.54 (7.172)	51.48 (7.238)	52.32 (7.296)	53.44 (7.374)	54.52 (7.447)	55.04 (7.483)	55.80 (7.533)	56.90 (7.606)	57.86 (7.668)	59.06 (7.747)	59.72 (7.789)	60.34 (7.83)
V <sub>23</sub>	105.80 (10.318)	107.30 (10.391)	109.20 (10.48)	111.16 (10.573)	112.40 (10.632)	113.32 (10.673)	114.36 (10.72)	115.60 (10.777)	117.40 (10.859)	119.20 (10.941)	120.86 (11.018)	123.20 (11.122)
V <sub>24</sub>	83.64 (9.198)	84.40 (9.239)	85.20 (9.282)	86.28 (9.341)	87.60 (9.41)	87.84 (9.423)	88.40 (9.453)	89.52 (9.512)	90.32 (9.554)	91.02 (9.59)	91.88 (9.635)	92.96 (9.691)
V <sub>25</sub>	98.80 (9.979)	100.00 (10.04)	100.60 (10.07)	102.20 (10.149)	103.26 (10.2)	103.94 (10.232)	105.12 (10.291)	107.00 (10.38)	108.40 (10.445)	109.94 (10.516)	112.48 (10.637)	114.76 (10.742)
V <sub>26</sub>	32.92 (5.807)	33.93 (5.895)	34.32 (5.928)	35.28 (6.007)	36.58 (6.113)	37.44 (6.184)	36.30 (6.071)	36.98 (6.126)	37.48 (6.165)	38.30 (6.229)	39.16 (6.302)	39.96 (6.365)
V <sub>27</sub>	38.34 (6.161)	39.42 (6.265)	40.50 (6.367)	41.72 (6.467)	42.24 (6.499)	42.96 (6.555)	43.58 (6.605)	44.18 (6.654)	45.38 (6.743)	45.92 (6.788)	46.08 (6.799)	48.52 (6.991)
V <sub>28</sub>	49.70 (7.104)	50.20 (7.141)	51.40 (7.227)	52.56 (7.308)	54.06 (7.412)	55.10 (7.482)	55.81 (7.529)	56.48 (7.573)	57.26 (7.624)	58.11 (7.68)	58.76 (7.723)	59.82 (7.791)
V <sub>29</sub>	37.74 (6.208)	38.82 (6.295)	39.80 (6.373)	41.20 (6.482)	42.10 (6.551)	43.46 (6.646)	44.14 (6.698)	44.86 (6.753)	46.30 (6.859)	46.86 (6.901)	47.80 (6.966)	48.98 (7.056)
V <sub>30</sub>	24.74 (5.041)	25.74 (5.136)	26.42 (5.211)	28.22 (5.392)	28.88 (5.455)	29.52 (5.514)	30.34 (5.588)	32.34 (5.766)	32.84 (5.81)	33.53 (5.867)	35.68 (6.046)	36.54 (6.116)
C.D.	0.65	0.637	0.618	0.614	0.617	0.625	0.636	0.631	0.638	0.638	0.636	0.611
SE(M)±	0.232	0.227	0.22	0.219	0.22	0.223	0.227	0.225	0.228	0.228	0.227	0.218
C.V.	9.213	8.854	8.476	8.28	8.203	8.241	8.31	8.163	8.169	8.099	7.99	7.607

Note: The data in parenthesis indicate square root transformed values

**APPENDIX II. Plant height of *Ascocentrum* hybrids/varieties during the period 2017-18, cm**

Var. No.	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	28.09 (5.38)	29.31 (5.488)	29.88 (5.541)	30.43 (5.592)	30.95 (5.636)	31.29 (5.664)	31.95 (5.72)	32.69 (5.783)	33.18 (5.824)	33.54 (5.852)	33.07 (5.822)	33.72 (5.881)
V <sub>2</sub>	24.25 (4.998)	25.37 (5.108)	26.19 (5.18)	26.64 (5.221)	27.37 (5.282)	27.75 (5.317)	28.59 (5.397)	29.09 (5.442)	29.45 (5.473)	29.77 (5.503)	30.36 (5.558)	31.17 (5.63)
V <sub>3</sub>	29.63 (5.529)	30.15 (5.576)	30.91 (5.644)	31.66 (5.708)	32.36 (5.771)	33.03 (5.827)	33.35 (5.855)	33.91 (5.902)	34.76 (5.973)	35.51 (6.035)	36.60 (6.126)	37.85 (6.229)
V <sub>4</sub>	51.21 (7.18)	51.80 (7.224)	52.43 (7.268)	53.24 (7.322)	54.15 (7.382)	54.67 (7.417)	55.00 (7.44)	55.94 (7.504)	56.69 (7.554)	57.50 (7.607)	58.06 (7.644)	58.78 (7.692)
V <sub>5</sub>	29.41 (5.502)	31.01 (5.645)	31.62 (5.7)	32.28 (5.758)	32.95 (5.815)	33.27 (5.842)	33.79 (5.886)	34.43 (5.943)	34.89 (5.982)	35.23 (6.011)	36.08 (6.082)	36.22 (6.094)
V <sub>6</sub>	30.12 (5.561)	31.55 (5.689)	31.93 (5.72)	33.27 (5.837)	33.58 (5.864)	34.20 (5.916)	34.73 (5.96)	35.63 (6.034)	35.87 (6.053)	36.43 (6.1)	37.10 (6.156)	37.63 (6.201)
V <sub>7</sub>	27.95 (5.375)	29.59 (5.52)	30.03 (5.562)	31.07 (5.654)	31.65 (5.705)	32.66 (5.794)	33.44 (5.863)	33.87 (5.901)	34.66 (5.968)	35.19 (6.014)	36.00 (6.08)	36.20 (6.097)
V <sub>8</sub>	38.06 (6.247)	38.68 (6.296)	39.45 (6.356)	40.14 (6.41)	40.35 (6.427)	41.01 (6.478)	41.66 (6.528)	42.27 (6.575)	42.95 (6.626)	43.47 (6.665)	43.76 (6.687)	44.41 (6.736)
V <sub>9</sub>	19.17 (4.484)	19.23 (4.494)	19.38 (4.513)	19.87 (4.568)	20.13 (4.595)	20.41 (4.626)	21.04 (4.694)	21.34 (4.726)	21.56 (4.749)	21.69 (4.763)	21.93 (4.788)	22.21 (4.816)
V <sub>10</sub>	29.83 (5.517)	30.77 (5.593)	31.48 (5.661)	32.75 (5.767)	33.40 (5.828)	33.61 (5.846)	34.53 (5.923)	35.50 (6.004)	35.96 (6.04)	36.75 (6.102)	37.41 (6.155)	38.17 (6.217)
V <sub>11</sub>	18.33 (4.386)	18.72 (4.431)	18.83 (4.443)	19.29 (4.491)	19.98 (4.567)	20.24 (4.594)	20.94 (4.669)	21.49 (4.729)	21.97 (4.779)	22.33 (4.816)	22.36 (4.825)	23.09 (4.901)
V <sub>12</sub>	36.41 (6.114)	37.79 (6.227)	38.39 (6.275)	39.23 (6.34)	39.34 (6.349)	39.75 (6.381)	40.41 (6.433)	41.41 (6.51)	42.58 (6.599)	43.35 (6.657)	43.77 (6.688)	44.51 (6.742)
V <sub>13</sub>	34.78 (5.974)	35.99 (6.074)	36.70 (6.131)	37.20 (6.172)	37.76 (6.218)	38.71 (6.295)	40.07 (6.401)	41.09 (6.479)	41.45 (6.507)	41.84 (6.537)	42.67 (6.599)	43.57 (6.667)
V <sub>14</sub>	26.48 (5.231)	27.50 (5.328)	28.24 (5.397)	29.78 (5.537)	30.86 (5.633)	31.52 (5.692)	32.00 (5.735)	32.60 (5.787)	33.06 (5.826)	33.62 (5.874)	34.12 (5.915)	34.98 (5.987)
V <sub>15</sub>	26.26 (5.189)	27.08 (5.269)	28.34 (5.392)	28.88 (5.437)	29.23 (5.464)	29.60 (5.498)	30.27 (5.554)	31.58 (5.667)	32.34 (5.725)	32.04 (5.701)	34.67 (5.911)	35.48 (5.978)
V <sub>16</sub>	24.88 (5.038)	25.91 (5.144)	27.53 (5.301)	28.73 (5.414)	29.93 (5.516)	31.54 (5.654)	32.39 (5.727)	34.06 (5.86)	34.67 (5.911)	34.82 (5.922)	35.53 (5.982)	36.21 (6.036)

**APPENDIX II. continued**

Var. No.	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	25.00 (5.094)	25.78 (5.17)	26.60 (5.25)	27.22 (5.309)	28.06 (5.387)	28.76 (5.452)	29.62 (5.53)	30.46 (5.607)	31.16 (5.669)	31.68 (5.714)	32.80 (5.81)	34.22 (5.932)
V <sub>18</sub>	31.94 (5.733)	32.91 (5.818)	33.41 (5.86)	34.22 (5.93)	35.23 (6.015)	36.05 (6.083)	36.91 (6.153)	37.95 (6.236)	38.49 (6.278)	39.05 (6.324)	39.90 (6.39)	40.34 (6.424)
V <sub>19</sub>	51.30 (7.23)	52.12 (7.287)	53.06 (7.351)	53.55 (7.384)	54.82 (7.468)	55.26 (7.497)	56.32 (7.568)	58.21 (7.691)	59.02 (7.743)	59.88 (7.798)	60.44 (7.834)	61.08 (7.874)
V <sub>20</sub>	47.98 (6.992)	48.94 (7.059)	49.90 (7.128)	50.54 (7.172)	51.71 (7.251)	52.29 (7.291)	53.26 (7.357)	54.80 (7.459)	55.82 (7.527)	57.06 (7.608)	58.68 (7.712)	60.30 (7.817)
V <sub>21</sub>	63.16 (8.003)	64.40 (8.081)	64.90 (8.113)	66.34 (8.201)	67.74 (8.287)	69.44 (8.391)	70.40 (8.449)	71.82 (8.533)	73.06 (8.605)	73.66 (8.64)	75.06 (8.721)	76.12 (8.782)
V <sub>22</sub>	64.18 (8.064)	65.46 (8.144)	66.70 (8.221)	69.36 (8.377)	70.22 (8.43)	71.78 (8.522)	72.78 (8.58)	73.34 (8.614)	75.52 (8.741)	76.58 (8.802)	77.86 (8.874)	79.58 (8.97)
V <sub>23</sub>	125.07 (11.207)	126.81 (11.284)	128.00 (11.336)	129.72 (11.411)	131.38 (11.483)	133.08 (11.556)	134.83 (11.632)	136.65 (11.71)	138.59 (11.793)	140.27 (11.865)	142.01 (11.938)	144.11 (12.026)
V <sub>24</sub>	93.90 (9.739)	95.08 (9.799)	95.98 (9.844)	97.50 (9.921)	99.22 (10.007)	100.22 (10.056)	101.94 (10.139)	103.80 (10.23)	105.76 (10.326)	107.71 (10.42)	109.61 (10.512)	111.50 (10.602)
V <sub>25</sub>	114.13 (10.704)	115.69 (10.775)	117.12 (10.841)	118.44 (10.9)	119.68 (10.956)	121.20 (11.024)	122.75 (11.093)	124.11 (11.154)	125.45 (11.213)	127.19 (11.29)	128.59 (11.349)	130.55 (11.437)
V <sub>26</sub>	41.05 (6.452)	41.97 (6.524)	42.48 (6.562)	43.30 (6.621)	43.66 (6.647)	44.30 (6.695)	45.70 (6.797)	46.60 (6.861)	47.47 (6.92)	48.25 (6.973)	50.01 (7.089)	51.19 (7.168)
V <sub>27</sub>	49.11 (7.028)	50.10 (7.094)	50.99 (7.154)	51.96 (7.221)	52.47 (7.258)	53.09 (7.304)	54.00 (7.364)	55.10 (7.44)	56.20 (7.515)	57.10 (7.574)	57.76 (7.618)	58.98 (7.695)
V <sub>28</sub>	60.90 (7.862)	61.92 (7.927)	62.58 (7.969)	63.28 (8.012)	64.08 (8.062)	64.80 (8.106)	65.44 (8.145)	66.30 (8.197)	67.12 (8.246)	68.36 (8.321)	68.22 (8.312)	69.52 (8.39)
V <sub>29</sub>	49.95 (7.126)	50.91 (7.195)	51.78 (7.255)	51.61 (7.243)	53.43 (7.357)	53.82 (7.383)	54.91 (7.456)	55.71 (7.508)	56.48 (7.558)	57.64 (7.634)	58.81 (7.709)	60.17 (7.796)
V <sub>30</sub>	38.21 (6.255)	39.94 (6.39)	41.10 (6.481)	42.39 (6.58)	43.85 (6.693)	45.73 (6.83)	46.88 (6.913)	47.46 (6.955)	48.91 (7.056)	50.24 (7.15)	50.97 (7.201)	51.73 (7.253)
C.D.	0.638	0.643	0.642	0.661	0.677	0.682	0.691	0.702	0.715	0.72	0.73	0.729
SE(M)±	0.228	0.229	0.229	0.236	0.241	0.243	0.247	0.25	0.255	0.257	0.26	0.26
C.V.	7.825	7.785	7.702	7.846	7.964	7.961	7.992	8.037	8.114	8.113	8.16	8.069

Note: The data in parenthesis indicate square root transformed values

**APPENDIX III. Plant spread of *Asocentrum* hybrids/varieties during the period 2016-17, cm**

Var. No.	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	38.00 (6.244)	38.37 (6.273)	38.73 (6.302)	39.20 (6.339)	39.67 (6.376)	40.07 (6.407)	40.43 (6.435)	40.80 (6.464)	41.27 (6.5)	41.60 (6.525)	42.20 (6.571)	42.47 (6.591)
V <sub>2</sub>	35.50 (6.04)	35.90 (6.073)	36.17 (6.095)	36.57 (6.128)	37.20 (6.179)	37.60 (6.212)	37.93 (6.238)	38.40 (6.276)	38.73 (6.303)	39.03 (6.326)	39.37 (6.353)	39.73 (6.381)
V <sub>3</sub>	46.33 (6.878)	46.70 (6.905)	46.97 (6.924)	47.25 (6.946)	47.60 (6.97)	47.93 (6.993)	48.40 (7.027)	48.80 (7.055)	49.37 (7.095)	49.73 (7.121)	50.07 (7.144)	50.40 (7.167)
V <sub>4</sub>	32.83 (5.813)	33.23 (5.847)	33.53 (5.873)	33.80 (5.896)	34.20 (5.93)	34.43 (5.95)	34.97 (5.994)	35.23 (6.016)	35.53 (6.04)	35.67 (6.051)	35.93 (6.074)	36.50 (6.119)
V <sub>5</sub>	36.67 (6.134)	36.93 (6.155)	37.27 (6.183)	37.63 (6.213)	38.00 (6.243)	38.53 (6.286)	39.10 (6.331)	39.27 (6.344)	39.57 (6.367)	39.83 (6.388)	40.10 (6.408)	40.40 (6.432)
V <sub>6</sub>	31.47 (5.694)	31.73 (5.717)	32.37 (5.772)	32.83 (5.812)	33.17 (5.841)	33.60 (5.877)	33.90 (5.902)	34.23 (5.931)	34.60 (5.961)	35.00 (5.994)	35.37 (6.025)	35.70 (6.053)
V <sub>7</sub>	29.67 (5.536)	29.97 (5.562)	30.33 (5.595)	30.60 (5.619)	31.10 (5.663)	31.70 (5.716)	32.17 (5.756)	32.60 (5.794)	32.93 (5.822)	33.40 (5.862)	33.77 (5.893)	34.23 (5.933)
V <sub>8</sub>	41.00 (6.477)	42.17 (6.565)	42.67 (6.603)	43.10 (6.636)	43.57 (6.671)	44.03 (6.705)	44.43 (6.734)	44.80 (6.762)	45.17 (6.789)	45.53 (6.816)	45.83 (6.838)	46.13 (6.86)
V <sub>9</sub>	14.33 (3.913)	14.77 (3.968)	15.17 (4.018)	15.43 (4.051)	16.03 (4.125)	16.33 (4.161)	16.77 (4.212)	17.20 (4.263)	17.47 (4.294)	17.93 (4.348)	18.70 (4.433)	19.03 (4.47)
V <sub>10</sub>	41.40 (6.511)	41.83 (6.544)	42.70 (6.61)	42.83 (6.62)	43.27 (6.653)	43.83 (6.695)	44.57 (6.75)	44.83 (6.77)	45.27 (6.802)	45.40 (6.812)	45.77 (6.839)	46.10 (6.863)
V <sub>11</sub>	17.53 (4.303)	17.87 (4.342)	18.17 (4.376)	18.40 (4.403)	18.57 (4.422)	18.90 (4.459)	19.13 (4.485)	19.57 (4.534)	20.27 (4.61)	20.53 (4.638)	20.77 (4.664)	21.03 (4.693)
V <sub>12</sub>	20.67 (4.653)	21.03 (4.692)	21.37 (4.727)	21.63 (4.756)	22.03 (4.798)	22.40 (4.836)	22.67 (4.864)	23.00 (4.898)	23.33 (4.932)	23.67 (4.965)	24.03 (5.002)	24.30 (5.029)
V <sub>13</sub>	39.67 (6.375)	40.10 (6.409)	40.67 (6.453)	41.17 (6.492)	41.53 (6.52)	41.93 (6.55)	42.37 (6.583)	42.80 (6.616)	43.23 (6.649)	43.50 (6.669)	43.90 (6.699)	44.17 (6.719)
V <sub>14</sub>	43.67 (6.68)	44.27 (6.726)	44.87 (6.77)	45.23 (6.797)	45.57 (6.822)	45.93 (6.849)	46.30 (6.876)	46.67 (6.902)	47.13 (6.936)	47.47 (6.96)	47.70 (6.977)	48.10 (7.006)
V <sub>15</sub>	46.50 (6.889)	46.83 (6.913)	47.23 (6.942)	47.90 (6.99)	48.30 (7.019)	48.67 (7.045)	48.93 (7.063)	49.13 (7.077)	49.47 (7.101)	49.67 (7.114)	49.93 (7.133)	50.10 (7.144)
V <sub>16</sub>	34.33 (5.935)	34.70 (5.966)	35.00 (5.991)	35.30 (6.016)	35.80 (6.058)	36.17 (6.088)	37.33 (6.184)	37.80 (6.222)	38.13 (6.249)	38.33 (6.265)	38.90 (6.31)	39.07 (6.323)

**APPENDIX III. continued**

Var. No.	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	27.00 (5.28)	27.43 (5.322)	27.87 (5.362)	28.13 (5.386)	28.47 (5.417)	28.83 (5.452)	29.03 (5.47)	29.40 (5.503)	29.83 (5.543)	30.10 (5.567)	30.40 (5.595)	30.80 (5.63)
V <sub>18</sub>	38.83 (6.305)	39.53 (6.361)	40.37 (6.427)	40.80 (6.46)	41.20 (6.491)	41.40 (6.506)	41.73 (6.532)	42.20 (6.568)	42.67 (6.603)	42.97 (6.626)	43.43 (6.661)	43.87 (6.693)
V <sub>19</sub>	29.33 (5.499)	29.70 (5.531)	30.03 (5.561)	30.30 (5.586)	30.57 (5.609)	31.07 (5.653)	31.53 (5.695)	31.87 (5.724)	32.17 (5.75)	32.53 (5.782)	32.60 (5.789)	32.80 (5.807)
V <sub>20</sub>	33.00 (5.819)	33.33 (5.846)	33.80 (5.887)	33.87 (5.889)	34.57 (5.951)	34.93 (5.983)	35.10 (5.996)	35.50 (6.03)	35.93 (6.066)	36.43 (6.107)	36.77 (6.135)	37.10 (6.162)
V <sub>21</sub>	46.33 (6.878)	46.70 (6.904)	47.40 (6.954)	47.70 (6.976)	48.10 (7.004)	48.50 (7.033)	48.90 (7.061)	49.23 (7.084)	49.50 (7.103)	49.93 (7.133)	50.27 (7.156)	50.57 (7.176)
V <sub>22</sub>	52.67 (7.323)	52.67 (7.323)	52.97 (7.344)	53.43 (7.375)	53.77 (7.398)	54.33 (7.436)	54.80 (7.467)	55.07 (7.484)	55.47 (7.511)	55.70 (7.527)	56.00 (7.547)	56.20 (7.56)
V <sub>23</sub>	52.00 (7.274)	52.30 (7.295)	52.63 (7.318)	53.03 (7.345)	53.40 (7.37)	54.10 (7.418)	54.33 (7.434)	54.97 (7.477)	55.30 (7.499)	55.53 (7.514)	56.20 (7.557)	56.43 (7.573)
V <sub>24</sub>	35.33 (5.997)	35.33 (6.011)	36.03 (6.055)	37.03 (6.096)	37.40 (6.129)	37.83 (6.164)	38.07 (6.18)	38.07 (6.199)	38.17 (6.224)	38.33 (6.251)	38.53 (6.28)	38.70 (6.304)
V <sub>25</sub>	34.33 (5.97)	34.63 (5.982)	35.07 (6.032)	35.30 (6.093)	35.77 (6.129)	36.13 (6.159)	36.33 (6.177)	36.43 (6.166)	36.73 (6.174)	37.03 (6.185)	37.40 (6.2)	37.67 (6.211)
V <sub>26</sub>	32.67 (5.795)	32.73 (5.8)	33.00 (5.823)	33.40 (5.858)	33.77 (5.888)	34.20 (5.925)	34.60 (5.959)	34.87 (5.982)	35.10 (6.003)	35.07 (6)	35.13 (6.006)	35.30 (6.021)
V <sub>27</sub>	57.33 (7.637)	57.70 (7.661)	58.20 (7.694)	58.47 (7.711)	58.83 (7.735)	59.10 (7.752)	59.47 (7.776)	59.67 (7.789)	59.90 (7.804)	60.17 (7.821)	60.50 (7.842)	60.93 (7.87)
V <sub>28</sub>	54.67 (7.457)	54.67 (7.457)	55.23 (7.495)	55.67 (7.524)	56.23 (7.561)	56.47 (7.577)	56.97 (7.61)	57.23 (7.628)	57.50 (7.646)	57.67 (7.657)	57.87 (7.67)	57.97 (7.677)
V <sub>29</sub>	51.83 (7.266)	52.00 (7.277)	52.43 (7.306)	53.00 (7.345)	53.33 (7.368)	53.67 (7.391)	54.07 (7.418)	54.37 (7.438)	54.80 (7.467)	55.10 (7.487)	55.57 (7.519)	55.97 (7.545)
V <sub>30</sub>	36.00 (6.073)	36.23 (6.092)	36.67 (6.127)	37.10 (6.163)	37.50 (6.195)	37.73 (6.213)	38.33 (6.262)	38.90 (6.308)	39.20 (6.331)	39.47 (6.352)	39.57 (6.36)	39.57 (6.359)
<b>C.D.</b>	0.452	0.455	0.453	0.455	0.449	0.447	0.445	0.442	0.44	0.441	0.441	0.445
<b>SE(m)±</b>	0.159	0.161	0.16	0.16	0.158	0.158	0.157	0.156	0.155	0.155	0.156	0.157
<b>C.V.</b>	4.488	4.497	4.445	4.444	4.367	4.327	4.283	4.236	4.194	4.189	4.173	4.192

Note: The data in parenthesis indicate square root transformed values

**APPENDIX IV. Plant spread of *Asocentrum* hybrids/varieties during the period 2017-18, cm**

Var. No.	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	42.83 (6.618)	43.13 (6.641)	43.47 (6.666)	43.80 (6.691)	44.07 (6.711)	44.47 (6.741)	45.00 (6.78)	43.90 (6.7)	44.37 (6.735)	44.60 (6.752)	45.20 (6.796)	47.83 (6.983)
V <sub>2</sub>	40.13 (6.413)	40.47 (6.438)	40.67 (6.454)	40.90 (6.472)	41.17 (6.493)	41.40 (6.511)	41.53 (6.522)	41.63 (6.529)	41.73 (6.537)	41.90 (6.55)	42.00 (6.557)	42.00 (6.557)
V <sub>3</sub>	50.80 (7.195)	51.23 (7.225)	51.60 (7.251)	52.37 (7.304)	52.60 (7.321)	53.23 (7.364)	53.87 (7.407)	54.33 (7.439)	54.73 (7.465)	54.93 (7.479)	54.93 (7.479)	54.93 (7.479)
V <sub>4</sub>	37.03 (6.161)	37.53 (6.199)	37.77 (6.218)	38.53 (6.281)	38.93 (6.313)	40.03 (6.401)	40.50 (6.438)	40.73 (6.456)	40.93 (6.472)	41.33 (6.502)	41.77 (6.536)	41.93 (6.549)
V <sub>5</sub>	40.77 (6.461)	41.10 (6.486)	41.53 (6.519)	41.90 (6.547)	42.20 (6.57)	42.50 (6.593)	42.93 (6.626)	43.30 (6.653)	43.50 (6.668)	43.83 (6.693)	44.00 (6.706)	44.17 (6.718)
V <sub>6</sub>	36.07 (6.083)	36.47 (6.116)	36.87 (6.149)	37.20 (6.176)	37.47 (6.197)	37.77 (6.222)	38.20 (6.256)	38.53 (6.283)	38.77 (6.302)	39.27 (6.341)	39.57 (6.365)	39.77 (6.381)
V <sub>7</sub>	34.57 (5.961)	34.90 (5.989)	35.27 (6.019)	35.63 (6.049)	36.13 (6.091)	36.40 (6.113)	36.73 (6.14)	37.23 (6.18)	37.50 (6.202)	37.83 (6.229)	38.13 (6.253)	37.97 (6.239)
V <sub>8</sub>	46.50 (6.886)	46.87 (6.913)	47.17 (6.934)	47.43 (6.953)	47.63 (6.968)	47.90 (6.987)	48.33 (7.018)	48.73 (7.047)	49.37 (7.093)	49.70 (7.116)	50.10 (7.145)	50.23 (7.154)
V <sub>9</sub>	19.40 (4.511)	19.83 (4.559)	20.30 (4.609)	20.67 (4.649)	20.70 (4.654)	21.00 (4.687)	21.40 (4.729)	21.60 (4.751)	21.80 (4.772)	21.93 (4.786)	21.00 (4.685)	21.07 (4.692)
V <sub>10</sub>	46.50 (6.892)	47.03 (6.93)	47.40 (6.957)	47.60 (6.971)	47.87 (6.99)	48.13 (7.009)	48.40 (7.028)	48.63 (7.045)	48.90 (7.064)	49.07 (7.076)	49.37 (7.097)	49.50 (7.106)
V <sub>11</sub>	21.70 (4.763)	21.83 (4.777)	22.00 (4.795)	22.23 (4.819)	22.57 (4.854)	23.00 (4.898)	23.37 (4.935)	23.63 (4.962)	23.93 (4.993)	24.10 (5.009)	24.27 (5.026)	24.43 (5.043)
V <sub>12</sub>	24.60 (5.059)	25.00 (5.098)	25.33 (5.131)	25.53 (5.15)	25.70 (5.166)	25.90 (5.185)	26.30 (5.224)	26.53 (5.246)	26.83 (5.274)	27.17 (5.306)	27.50 (5.338)	27.87 (5.372)
V <sub>13</sub>	44.23 (6.724)	44.40 (6.736)	44.67 (6.756)	45.03 (6.783)	45.40 (6.81)	45.90 (6.847)	46.23 (6.871)	46.53 (6.893)	46.97 (6.924)	47.37 (6.953)	47.40 (6.956)	47.53 (6.965)
V <sub>14</sub>	48.47 (7.032)	48.53 (7.037)	48.97 (7.068)	49.17 (7.082)	49.57 (7.11)	49.87 (7.131)	50.23 (7.157)	50.57 (7.18)	51.00 (7.21)	51.67 (7.257)	52.03 (7.282)	52.43 (7.309)
V <sub>15</sub>	50.47 (7.17)	50.83 (7.195)	51.20 (7.221)	51.37 (7.233)	51.60 (7.249)	51.70 (7.256)	51.80 (7.263)	52.20 (7.292)	52.90 (7.34)	53.13 (7.356)	53.37 (7.372)	53.67 (7.393)
V <sub>16</sub>	39.30 (6.342)	39.47 (6.354)	39.93 (6.391)	40.13 (6.407)	40.43 (6.43)	40.80 (6.459)	41.23 (6.493)	41.67 (6.526)	42.10 (6.559)	42.63 (6.599)	42.80 (6.611)	43.00 (6.627)



**APPENDIX IV. continued**

Var. No.	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	31.27 (5.671)	31.63 (5.704)	31.93 (5.73)	32.27 (5.76)	32.20 (5.753)	32.87 (5.813)	33.33 (5.853)	33.57 (5.872)	33.97 (5.906)	34.80 (5.976)	35.13 (6.004)	35.67 (6.048)
V <sub>18</sub>	44.37 (6.73)	44.60 (6.748)	45.00 (6.778)	45.27 (6.798)	45.47 (6.814)	45.83 (6.841)	46.07 (6.858)	46.43 (6.885)	46.67 (6.903)	46.83 (6.915)	47.00 (6.927)	47.00 (6.927)
V <sub>19</sub>	33.07 (5.83)	33.33 (5.853)	33.50 (5.867)	33.73 (5.887)	34.07 (5.915)	34.33 (5.937)	34.83 (5.979)	35.13 (6.003)	35.60 (6.042)	36.03 (6.08)	36.53 (6.12)	37.13 (6.169)
V <sub>20</sub>	37.37 (6.183)	37.73 (6.214)	37.97 (6.233)	38.47 (6.272)	38.90 (6.307)	39.30 (6.339)	39.93 (6.389)	40.33 (6.42)	40.00 (6.397)	40.60 (6.443)	41.00 (6.474)	41.23 (6.492)
V <sub>21</sub>	50.83 (7.194)	51.00 (7.206)	51.10 (7.213)	51.40 (7.234)	51.57 (7.246)	51.63 (7.251)	51.93 (7.272)	51.77 (7.26)	52.07 (7.281)	52.20 (7.29)	52.33 (7.299)	52.33 (7.299)
V <sub>22</sub>	56.60 (7.587)	56.60 (7.587)	56.93 (7.609)	57.10 (7.62)	57.20 (7.626)	57.43 (7.642)	57.57 (7.65)	57.87 (7.67)	58.20 (7.692)	58.40 (7.705)	58.53 (7.714)	58.60 (7.718)
V <sub>23</sub>	56.57 (7.582)	56.67 (7.589)	56.67 (7.589)	56.67 (7.589)	56.67 (7.589)	56.87 (7.602)	56.97 (7.609)	57.20 (7.624)	57.43 (7.64)	57.53 (7.647)	57.63 (7.653)	57.93 (7.673)
V <sub>24</sub>	39.00 (6.33)	39.53 (6.348)	39.87 (6.364)	40.47 (6.392)	40.83 (6.41)	41.20 (6.425)	41.60 (6.455)	41.97 (6.473)	42.23 (6.484)	42.40 (6.507)	42.50 (6.53)	42.60 (6.543)
V <sub>25</sub>	37.90 (6.226)	38.13 (6.269)	38.40 (6.301)	38.83 (6.354)	39.10 (6.385)	39.43 (6.424)	39.87 (6.458)	40.27 (6.499)	40.50 (6.528)	40.63 (6.528)	40.83 (6.528)	40.90 (6.528)
V <sub>26</sub>	35.57 (6.043)	35.77 (6.06)	36.00 (6.079)	36.50 (6.12)	37.03 (6.164)	37.57 (6.207)	38.10 (6.25)	38.53 (6.284)	38.77 (6.303)	39.20 (6.337)	39.70 (6.376)	39.80 (6.384)
V <sub>27</sub>	61.63 (7.914)	62.17 (7.948)	62.60 (7.975)	62.60 (7.975)	62.60 (7.975)	62.60 (7.975)	62.60 (7.975)	62.60 (7.975)	62.60 (7.975)	62.73 (7.983)	62.87 (7.992)	63.00 (8)
V <sub>28</sub>	58.40 (7.704)	58.53 (7.713)	58.67 (7.721)	58.73 (7.726)	59.10 (7.75)	59.53 (7.777)	59.70 (7.788)	60.00 (7.807)	60.10 (7.813)	60.53 (7.84)	60.53 (7.84)	60.53 (7.84)
V <sub>29</sub>	56.30 (7.567)	56.60 (7.587)	56.70 (7.593)	57.13 (7.62)	57.13 (7.62)	57.33 (7.634)	57.37 (7.636)	57.40 (7.638)	57.47 (7.643)	57.77 (7.662)	58.10 (7.684)	59.73 (7.79)
V <sub>30</sub>	39.70 (6.368)	40.00 (6.392)	40.30 (6.415)	40.70 (6.445)	41.03 (6.47)	41.40 (6.5)	41.73 (6.526)	42.17 (6.561)	42.43 (6.582)	42.60 (6.595)	42.60 (6.595)	42.73 (6.606)
<b>C.D.</b>	0.454	0.456	0.454	0.446	0.445	0.43	0.428	0.42	0.402	0.399	0.402	0.404
<b>SE(m)±</b>	0.16	0.161	0.16	0.157	0.157	0.152	0.151	0.148	0.142	0.141	0.142	0.142
<b>C.V.</b>	4.266	4.269	4.234	4.136	4.12	3.967	3.925	3.848	3.671	3.632	3.651	3.652

Note: The data in parenthesis indicate square root transformed values

**APPENDIX V. Shoot girth of *Ascocentrum* hybrids/varieties during 2016-17 the period, cm**

Var. No.	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	3.33 (2.081)	3.40 (2.098)	3.47 (2.113)	3.57 (2.137)	3.57 (2.137)	3.67 (2.16)	3.77 (2.183)	3.73 (2.175)	3.77 (2.183)	3.80 (2.191)	3.83 (2.198)	3.80 (2.191)
V <sub>2</sub>	3.97 (2.228)	4.13 (2.266)	4.30 (2.302)	4.33 (2.309)	4.33 (2.309)	4.37 (2.316)	4.37 (2.316)	4.23 (2.288)	4.27 (2.295)	4.27 (2.295)	4.30 (2.302)	4.33 (2.309)
V <sub>3</sub>	4.77 (2.401)	4.77 (2.401)	4.83 (2.415)	4.93 (2.436)	4.93 (2.436)	5.13 (2.476)	5.10 (2.469)	5.20 (2.49)	5.27 (2.503)	5.30 (2.51)	5.33 (2.516)	5.40 (2.529)
V <sub>4</sub>	4.00 (2.233)	4.10 (2.256)	4.20 (2.278)	4.30 (2.3)	4.33 (2.307)	4.40 (2.322)	4.40 (2.322)	4.40 (2.321)	4.53 (2.349)	4.63 (2.37)	4.80 (2.407)	4.83 (2.414)
V <sub>5</sub>	3.57 (2.137)	3.63 (2.152)	3.73 (2.175)	3.87 (2.205)	3.87 (2.205)	3.97 (2.228)	4.00 (2.235)	4.03 (2.243)	4.07 (2.25)	4.10 (2.257)	4.13 (2.264)	4.17 (2.271)
V <sub>6</sub>	3.13 (2.033)	3.37 (2.09)	3.47 (2.113)	3.57 (2.137)	3.57 (2.137)	3.67 (2.16)	3.80 (2.191)	3.73 (2.176)	3.77 (2.183)	3.80 (2.191)	3.83 (2.198)	3.87 (2.206)
V <sub>7</sub>	4.23 (2.287)	4.40 (2.324)	4.43 (2.331)	4.63 (2.373)	4.70 (2.387)	4.83 (2.415)	4.87 (2.422)	4.93 (2.435)	5.00 (2.449)	5.07 (2.462)	5.20 (2.489)	5.27 (2.503)
V <sub>8</sub>	3.73 (2.176)	3.77 (2.183)	3.87 (2.206)	3.97 (2.229)	3.97 (2.229)	4.00 (2.236)	4.00 (2.236)	3.97 (2.229)	4.03 (2.243)	4.10 (2.257)	4.10 (2.257)	4.13 (2.265)
V <sub>9</sub>	2.10 (1.76)	2.17 (1.778)	2.17 (1.779)	2.23 (1.798)	2.27 (1.807)	2.33 (1.825)	2.33 (1.825)	2.50 (1.871)	2.57 (1.889)	2.63 (1.906)	2.67 (1.915)	2.70 (1.924)
V <sub>10</sub>	3.57 (2.137)	3.67 (2.16)	3.73 (2.176)	3.83 (2.198)	3.90 (2.214)	3.93 (2.221)	4.00 (2.236)	4.03 (2.243)	4.13 (2.265)	4.20 (2.28)	4.30 (2.302)	4.37 (2.317)
V <sub>11</sub>	2.77 (1.941)	2.93 (1.983)	3.03 (2.008)	3.07 (2.016)	3.10 (2.024)	3.23 (2.057)	3.20 (2.049)	3.27 (2.065)	3.37 (2.089)	3.50 (2.121)	3.57 (2.137)	3.63 (2.152)
V <sub>12</sub>	3.73 (2.175)	3.80 (2.19)	3.93 (2.22)	4.00 (2.236)	4.07 (2.251)	4.20 (2.28)	4.20 (2.28)	4.27 (2.294)	4.27 (2.294)	4.30 (2.301)	4.33 (2.308)	4.33 (2.308)
V <sub>13</sub>	3.90 (2.213)	4.03 (2.242)	4.10 (2.257)	4.23 (2.286)	4.27 (2.294)	4.33 (2.309)	4.40 (2.324)	4.37 (2.317)	4.43 (2.331)	4.57 (2.359)	4.67 (2.38)	4.77 (2.401)
V <sub>14</sub>	3.47 (2.113)	3.57 (2.136)	3.67 (2.16)	3.77 (2.183)	3.80 (2.19)	3.90 (2.213)	3.97 (2.228)	3.93 (2.22)	4.03 (2.243)	4.10 (2.258)	4.20 (2.28)	4.27 (2.295)
V <sub>15</sub>	4.00 (2.234)	4.20 (2.278)	4.23 (2.286)	4.30 (2.3)	4.33 (2.308)	4.33 (2.308)	4.30 (2.3)	4.30 (2.3)	4.33 (2.307)	4.33 (2.307)	4.43 (2.33)	4.47 (2.336)
V <sub>16</sub>	3.47 (2.113)	3.57 (2.136)	3.67 (2.159)	3.77 (2.183)	3.77 (2.183)	3.87 (2.205)	3.87 (2.205)	3.87 (2.206)	3.93 (2.22)	4.03 (2.242)	4.07 (2.25)	4.10 (2.257)

**APPENDIX V. continued**

Var. No.	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	2.67 (1.914)	2.73 (1.931)	2.83 (1.957)	2.87 (1.965)	2.90 (1.974)	2.93 (1.982)	2.97 (1.989)	3.00 (1.998)	3.13 (2.03)	3.20 (2.047)	3.27 (2.063)	3.33 (2.078)
V <sub>18</sub>	3.60 (2.143)	3.73 (2.174)	3.80 (2.189)	3.90 (2.212)	4.00 (2.234)	4.07 (2.249)	4.13 (2.264)	4.13 (2.264)	4.17 (2.272)	4.23 (2.286)	4.33 (2.308)	4.37 (2.315)
V <sub>19</sub>	4.50 (2.345)	4.53 (2.352)	4.73 (2.394)	4.80 (2.408)	4.83 (2.415)	4.90 (2.429)	4.93 (2.436)	4.97 (2.442)	5.00 (2.449)	5.07 (2.463)	5.13 (2.476)	5.13 (2.476)
V <sub>20</sub>	4.93 (2.435)	4.97 (2.442)	5.00 (2.449)	5.03 (2.455)	5.10 (2.469)	5.17 (2.482)	5.20 (2.489)	5.20 (2.489)	5.23 (2.495)	5.27 (2.502)	5.40 (2.527)	5.43 (2.534)
V <sub>21</sub>	4.67 (2.38)	4.67 (2.38)	4.77 (2.401)	4.83 (2.415)	4.87 (2.422)	4.93 (2.435)	4.97 (2.442)	5.00 (2.449)	5.13 (2.476)	5.17 (2.483)	5.33 (2.516)	5.27 (2.503)
V <sub>22</sub>	4.67 (2.38)	4.70 (2.387)	4.77 (2.401)	4.87 (2.422)	4.90 (2.429)	5.00 (2.449)	5.00 (2.449)	5.10 (2.47)	5.17 (2.483)	5.23 (2.497)	5.40 (2.53)	5.37 (2.523)
V <sub>23</sub>	5.70 (2.588)	5.77 (2.601)	5.83 (2.614)	5.97 (2.639)	5.97 (2.639)	6.07 (2.658)	6.07 (2.658)	6.10 (2.665)	6.13 (2.671)	6.27 (2.696)	6.27 (2.696)	6.27 (2.696)
V <sub>24</sub>	5.47 (2.542)	5.53 (2.556)	5.57 (2.569)	5.67 (2.588)	5.70 (2.588)	5.70 (2.595)	5.70 (2.595)	5.77 (2.62)	5.90 (2.646)	6.03 (2.658)	6.10 (2.671)	6.17 (2.677)
V <sub>25</sub>	5.60 (2.569)	5.63 (2.575)	5.73 (2.588)	5.83 (2.607)	5.83 (2.614)	5.97 (2.633)	5.97 (2.633)	6.03 (2.632)	6.10 (2.645)	6.17 (2.67)	6.27 (2.689)	6.33 (2.708)
V <sub>26</sub>	4.77 (2.4)	4.83 (2.414)	4.87 (2.421)	4.93 (2.435)	4.93 (2.435)	5.12 (2.476)	5.10 (2.469)	5.13 (2.476)	5.20 (2.489)	5.20 (2.489)	5.30 (2.509)	5.37 (2.522)
V <sub>27</sub>	4.50 (2.345)	4.57 (2.359)	4.63 (2.373)	4.73 (2.394)	4.77 (2.401)	4.90 (2.429)	4.90 (2.429)	4.97 (2.442)	5.03 (2.456)	5.10 (2.47)	5.13 (2.477)	5.23 (2.497)
V <sub>28</sub>	4.57 (2.358)	4.63 (2.373)	4.80 (2.408)	4.97 (2.442)	4.93 (2.435)	5.03 (2.456)	5.03 (2.456)	5.07 (2.463)	5.17 (2.483)	5.13 (2.476)	5.13 (2.476)	5.20 (2.49)
V <sub>29</sub>	4.63 (2.373)	4.63 (2.373)	4.83 (2.415)	5.07 (2.462)	5.10 (2.469)	5.13 (2.476)	5.17 (2.483)	5.20 (2.49)	5.30 (2.51)	5.23 (2.497)	5.33 (2.517)	5.30 (2.51)
V <sub>30</sub>	3.27 (2.064)	3.30 (2.072)	3.37 (2.088)	3.43 (2.104)	3.47 (2.112)	3.57 (2.135)	3.60 (2.143)	3.63 (2.151)	3.70 (2.166)	3.70 (2.166)	3.70 (2.166)	3.73 (2.174)
<b>C.D.</b>	0.11	0.107	0.1	0.102	0.1	0.107	0.108	0.114	0.115	0.116	0.114	0.117
<b>SE(m)±</b>	0.039	0.038	0.035	0.036	0.035	0.038	0.038	0.04	0.04	0.041	0.04	0.041
<b>C.V.</b>	2.993	2.908	2.685	2.72	2.648	2.804	2.831	2.981	2.984	3.01	2.94	2.997

Note: The data in parenthesis indicate square root transformed values

**APPENDIX VI. Shoot girth of *Ascocentrum* hybrids/varieties during the period 2017-18, cm**

Var. No.	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	3.90 (2.214)	4.03 (2.243)	4.03 (2.243)	4.13 (2.266)	4.13 (2.266)	4.07 (2.251)	4.17 (2.273)	4.23 (2.287)	4.47 (2.338)	4.57 (2.359)	4.57 (2.359)	4.57 (2.359)
V <sub>2</sub>	4.37 (2.316)	4.53 (2.352)	4.63 (2.373)	4.83 (2.415)	4.87 (2.422)	5.00 (2.449)	5.07 (2.463)	5.07 (2.463)	5.13 (2.476)	5.17 (2.483)	5.20 (2.489)	5.27 (2.503)
V <sub>3</sub>	5.43 (2.536)	5.53 (2.556)	5.57 (2.562)	5.72 (2.595)	5.70 (2.588)	5.77 (2.601)	5.77 (2.601)	5.87 (2.62)	5.93 (2.633)	5.97 (2.639)	6.03 (2.652)	6.07 (2.658)
V <sub>4</sub>	4.87 (2.42)	4.90 (2.427)	4.97 (2.441)	5.03 (2.454)	5.00 (2.448)	5.10 (2.469)	5.03 (2.455)	5.10 (2.469)	5.20 (2.489)	5.27 (2.502)	5.30 (2.508)	5.30 (2.508)
V <sub>5</sub>	4.03 (2.24)	4.17 (2.27)	4.27 (2.293)	4.30 (2.3)	4.37 (2.314)	4.37 (2.314)	4.37 (2.314)	4.50 (2.343)	4.53 (2.349)	4.57 (2.356)	4.60 (2.363)	4.63 (2.37)
V <sub>6</sub>	3.97 (2.229)	4.33 (2.309)	4.53 (2.351)	4.63 (2.372)	4.63 (2.372)	4.67 (2.378)	4.83 (2.413)	4.87 (2.42)	4.90 (2.427)	4.93 (2.435)	5.00 (2.448)	5.30 (2.51)
V <sub>7</sub>	5.27 (2.503)	5.33 (2.516)	5.42 (2.536)	5.47 (2.543)	5.50 (2.549)	5.53 (2.556)	5.53 (2.556)	5.57 (2.562)	5.60 (2.569)	5.63 (2.575)	5.63 (2.575)	5.63 (2.575)
V <sub>8</sub>	4.20 (2.28)	4.33 (2.309)	4.43 (2.331)	4.47 (2.338)	4.50 (2.345)	4.63 (2.373)	4.63 (2.373)	4.67 (2.38)	4.73 (2.394)	4.77 (2.401)	4.83 (2.415)	4.83 (2.415)
V <sub>9</sub>	2.80 (1.949)	2.90 (1.974)	2.97 (1.991)	3.03 (2.008)	3.07 (2.016)	3.10 (2.024)	3.10 (2.024)	3.17 (2.041)	3.23 (2.057)	3.27 (2.065)	3.27 (2.065)	3.40 (2.096)
V <sub>10</sub>	4.43 (2.331)	4.43 (2.331)	4.67 (2.38)	4.80 (2.408)	4.87 (2.421)	4.87 (2.422)	4.93 (2.435)	4.97 (2.442)	4.97 (2.442)	5.07 (2.463)	5.03 (2.456)	5.03 (2.456)
V <sub>11</sub>	3.70 (2.168)	3.73 (2.175)	3.80 (2.19)	3.83 (2.198)	3.90 (2.213)	3.93 (2.221)	3.93 (2.221)	4.07 (2.25)	4.13 (2.265)	4.17 (2.272)	4.20 (2.279)	4.17 (2.272)
V <sub>12</sub>	4.40 (2.323)	4.47 (2.337)	4.60 (2.366)	4.67 (2.38)	4.67 (2.38)	4.87 (2.421)	4.87 (2.421)	4.97 (2.442)	5.00 (2.449)	5.07 (2.462)	5.13 (2.476)	5.13 (2.476)
V <sub>13</sub>	4.77 (2.401)	4.77 (2.401)	4.83 (2.415)	4.93 (2.436)	4.97 (2.443)	5.07 (2.463)	5.03 (2.456)	5.17 (2.483)	5.23 (2.496)	5.27 (2.503)	5.27 (2.503)	5.30 (2.509)
V <sub>14</sub>	4.30 (2.302)	4.50 (2.345)	4.53 (2.352)	4.60 (2.366)	4.63 (2.373)	4.73 (2.394)	4.80 (2.408)	4.87 (2.422)	4.97 (2.442)	4.97 (2.442)	5.13 (2.476)	5.13 (2.476)
V <sub>15</sub>	4.50 (2.344)	4.77 (2.401)	4.83 (2.415)	4.93 (2.436)	4.93 (2.436)	5.00 (2.449)	5.13 (2.476)	5.13 (2.476)	5.17 (2.483)	5.20 (2.49)	5.30 (2.51)	5.33 (2.516)
V <sub>16</sub>	4.13 (2.264)	4.27 (2.294)	4.27 (2.294)	4.37 (2.316)	4.40 (2.322)	4.40 (2.322)	4.43 (2.329)	4.43 (2.329)	4.63 (2.372)	4.73 (2.394)	4.77 (2.401)	4.77 (2.401)

**APPENDIX VI. continued**

Var. No.	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	3.43 (2.102)	3.50 (2.118)	3.53 (2.125)	3.60 (2.14)	3.67 (2.156)	3.73 (2.17)	3.80 (2.185)	3.83 (2.193)	3.83 (2.193)	3.87 (2.2)	3.90 (2.207)	3.97 (2.222)
V <sub>18</sub>	4.40 (2.322)	4.43 (2.329)	4.50 (2.343)	4.57 (2.357)	4.57 (2.357)	4.60 (2.364)	4.63 (2.372)	4.70 (2.386)	4.77 (2.4)	4.77 (2.4)	4.83 (2.414)	4.87 (2.421)
V <sub>19</sub>	5.27 (2.503)	5.30 (2.51)	5.37 (2.523)	5.43 (2.536)	5.50 (2.549)	5.53 (2.556)	5.57 (2.562)	5.60 (2.569)	5.67 (2.582)	5.77 (2.601)	5.83 (2.614)	5.90 (2.626)
V <sub>20</sub>	5.47 (2.541)	5.47 (2.541)	5.53 (2.554)	5.57 (2.56)	5.67 (2.58)	5.70 (2.586)	5.77 (2.599)	5.80 (2.605)	5.93 (2.631)	5.97 (2.637)	6.00 (2.643)	6.07 (2.656)
V <sub>21</sub>	5.23 (2.497)	5.27 (2.503)	5.37 (2.523)	5.47 (2.543)	5.47 (2.543)	5.53 (2.555)	5.67 (2.582)	5.70 (2.588)	5.77 (2.601)	5.83 (2.614)	5.83 (2.614)	5.97 (2.639)
V <sub>22</sub>	5.40 (2.53)	5.43 (2.536)	5.53 (2.556)	5.60 (2.569)	5.60 (2.569)	5.80 (2.607)	5.80 (2.607)	5.83 (2.614)	5.87 (2.62)	5.93 (2.633)	5.97 (2.639)	6.07 (2.658)
V <sub>23</sub>	6.33 (2.708)	6.37 (2.714)	6.40 (2.72)	6.47 (2.733)	6.53 (2.745)	6.57 (2.751)	6.67 (2.769)	6.70 (2.775)	6.77 (2.787)	6.83 (2.799)	6.90 (2.811)	6.97 (2.823)
V <sub>24</sub>	6.20 (2.689)	6.23 (2.695)	6.30 (2.702)	6.33 (2.708)	6.37 (2.708)	6.40 (2.72)	6.47 (2.732)	6.50 (2.732)	6.57 (2.75)	6.60 (2.75)	6.67 (2.763)	6.73 (2.775)
V <sub>25</sub>	6.43 (2.72)	6.53 (2.738)	6.60 (2.756)	6.60 (2.756)	6.67 (2.774)	6.70 (2.774)	6.80 (2.792)	6.83 (2.804)	6.90 (2.81)	6.93 (2.822)	7.03 (2.84)	7.10 (2.851)
V <sub>26</sub>	5.43 (2.535)	5.50 (2.548)	5.60 (2.568)	5.70 (2.587)	5.70 (2.587)	5.80 (2.607)	5.80 (2.606)	5.83 (2.613)	5.90 (2.626)	6.00 (2.645)	6.03 (2.651)	6.17 (2.676)
V <sub>27</sub>	5.27 (2.503)	5.30 (2.51)	5.37 (2.523)	5.37 (2.523)	5.43 (2.536)	5.53 (2.556)	5.53 (2.556)	5.57 (2.562)	5.63 (2.575)	5.73 (2.594)	5.87 (2.62)	5.93 (2.633)
V <sub>28</sub>	5.23 (2.496)	5.27 (2.503)	5.27 (2.503)	5.32 (2.516)	5.35 (2.523)	5.35 (2.523)	5.40 (2.53)	5.47 (2.543)	5.43 (2.536)	5.50 (2.549)	5.60 (2.568)	5.70 (2.588)
V <sub>29</sub>	5.40 (2.53)	5.37 (2.523)	5.40 (2.53)	5.68 (2.588)	5.77 (2.601)	5.80 (2.607)	5.87 (2.62)	5.87 (2.62)	5.97 (2.639)	6.07 (2.658)	6.17 (2.677)	6.30 (2.701)
V <sub>307</sub>	3.83 (2.198)	3.87 (2.205)	3.90 (2.213)	4.00 (2.235)	4.00 (2.235)	4.03 (2.243)	4.03 (2.243)	4.03 (2.243)	4.13 (2.265)	4.20 (2.279)	4.23 (2.287)	4.27 (2.294)
<b>C.D.</b>	0.122	0.12	0.122	0.128	0.128	0.131	0.132	0.13	0.129	0.131	0.134	0.139
<b>SE(m)±</b>	0.043	0.042	0.043	0.045	0.045	0.046	0.047	0.046	0.045	0.046	0.047	0.049
<b>C.V.</b>	3.11	3.052	3.087	3.205	3.208	3.248	3.276	3.196	3.159	3.2	3.267	3.355

Note: The data in parenthesis indicate square root transformed values

**APPENDIX VII. Shoot diameter *Ascocentrum* hybrids/varieties during the period 2016-17, cm**

Var. No.	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	1.06 (1.435)	1.08 (1.442)	1.10 (1.45)	1.14 (1.461)	1.14 (1.461)	1.17 (1.473)	1.20 (1.483)	1.19 (1.48)	1.20 (1.483)	1.21 (1.487)	1.22 (1.49)	1.21 (1.487)
V <sub>2</sub>	1.26 (1.505)	1.32 (1.522)	1.37 (1.539)	1.38 (1.543)	1.38 (1.543)	1.39 (1.546)	1.39 (1.547)	1.35 (1.533)	1.36 (1.536)	1.36 (1.536)	1.37 (1.539)	1.38 (1.543)
V <sub>3</sub>	1.52 (1.586)	1.52 (1.586)	1.54 (1.593)	1.57 (1.603)	1.57 (1.603)	1.63 (1.624)	1.62 (1.621)	1.66 (1.63)	1.68 (1.636)	1.69 (1.639)	1.70 (1.642)	1.72 (1.65)
V <sub>4</sub>	1.27 (1.507)	1.31 (1.518)	1.34 (1.528)	1.37 (1.539)	1.38 (1.542)	1.40 (1.549)	1.40 (1.549)	1.40 (1.549)	1.44 (1.562)	1.48 (1.572)	1.53 (1.589)	1.54 (1.592)
V <sub>5</sub>	1.14 (1.46)	1.16 (1.467)	1.19 (1.479)	1.23 (1.494)	1.23 (1.494)	1.26 (1.504)	1.27 (1.508)	1.28 (1.512)	1.30 (1.515)	1.31 (1.518)	1.32 (1.522)	1.33 (1.526)
V <sub>6</sub>	1.00 (1.414)	1.07 (1.439)	1.10 (1.45)	1.14 (1.461)	1.14 (1.461)	1.17 (1.473)	1.21 (1.487)	1.19 (1.48)	1.20 (1.483)	1.21 (1.487)	1.22 (1.49)	1.23 (1.493)
V <sub>7</sub>	1.35 (1.533)	1.40 (1.549)	1.41 (1.552)	1.48 (1.574)	1.50 (1.58)	1.54 (1.593)	1.55 (1.596)	1.57 (1.603)	1.59 (1.609)	1.61 (1.616)	1.66 (1.63)	1.68 (1.636)
V <sub>8</sub>	1.19 (1.48)	1.20 (1.483)	1.23 (1.493)	1.26 (1.504)	1.26 (1.504)	1.27 (1.508)	1.27 (1.508)	1.26 (1.504)	1.28 (1.511)	1.31 (1.517)	1.31 (1.517)	1.32 (1.521)
V <sub>9</sub>	0.67 (1.291)	0.69 (1.298)	0.69 (1.3)	0.71 (1.308)	0.72 (1.311)	0.74 (1.321)	0.74 (1.321)	0.80 (1.34)	0.82 (1.349)	0.84 (1.356)	0.85 (1.36)	0.86 (1.364)
V <sub>10</sub>	1.14 (1.461)	1.17 (1.473)	1.19 (1.48)	1.22 (1.49)	1.24 (1.497)	1.25 (1.5)	1.27 (1.508)	1.28 (1.511)	1.32 (1.522)	1.34 (1.529)	1.37 (1.539)	1.39 (1.546)
V <sub>11</sub>	0.88 (1.371)	0.93 (1.39)	0.97 (1.401)	0.98 (1.405)	0.99 (1.409)	1.03 (1.425)	1.02 (1.421)	1.04 (1.429)	1.07 (1.44)	1.11 (1.454)	1.14 (1.46)	1.16 (1.468)
V <sub>12</sub>	1.19 (1.479)	1.21 (1.488)	1.25 (1.501)	1.27 (1.508)	1.30 (1.514)	1.34 (1.528)	1.34 (1.528)	1.36 (1.535)	1.36 (1.535)	1.37 (1.539)	1.38 (1.542)	1.38 (1.542)
V <sub>13</sub>	1.24 (1.496)	1.28 (1.51)	1.31 (1.518)	1.35 (1.532)	1.36 (1.535)	1.38 (1.542)	1.40 (1.549)	1.39 (1.546)	1.41 (1.552)	1.45 (1.566)	1.49 (1.576)	1.52 (1.586)
V <sub>14</sub>	1.10 (1.45)	1.14 (1.462)	1.17 (1.472)	1.20 (1.482)	1.21 (1.485)	1.24 (1.498)	1.26 (1.505)	1.25 (1.501)	1.28 (1.512)	1.31 (1.519)	1.34 (1.528)	1.36 (1.535)
V <sub>15</sub>	1.27 (1.507)	1.34 (1.529)	1.35 (1.532)	1.37 (1.539)	1.38 (1.542)	1.38 (1.542)	1.37 (1.539)	1.37 (1.539)	1.38 (1.542)	1.38 (1.542)	1.41 (1.552)	1.42 (1.555)
V <sub>16</sub>	1.10 (1.45)	1.14 (1.46)	1.17 (1.472)	1.20 (1.483)	1.20 (1.483)	1.23 (1.494)	1.23 (1.494)	1.23 (1.494)	1.25 (1.501)	1.28 (1.511)	1.30 (1.514)	1.31 (1.517)

**APPENDIX VII. continued**

Var. No.	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	0.85 (1.36)	0.87 (1.368)	0.90 (1.379)	0.91 (1.383)	0.92 (1.387)	0.93 (1.39)	0.94 (1.395)	0.96 (1.397)	1.00 (1.413)	1.02 (1.42)	1.04 (1.427)	1.06 (1.434)
V <sub>18</sub>	1.15 (1.465)	1.19 (1.479)	1.21 (1.486)	1.24 (1.496)	1.27 (1.506)	1.30 (1.514)	1.32 (1.522)	1.32 (1.521)	1.33 (1.525)	1.35 (1.532)	1.38 (1.542)	1.39 (1.545)
V <sub>19</sub>	1.43 (1.559)	1.44 (1.562)	1.51 (1.583)	1.53 (1.591)	1.54 (1.594)	1.56 (1.6)	1.57 (1.603)	1.58 (1.607)	1.59 (1.61)	1.61 (1.617)	1.63 (1.624)	1.63 (1.624)
V <sub>20</sub>	1.57 (1.603)	1.58 (1.606)	1.59 (1.609)	1.60 (1.612)	1.62 (1.619)	1.65 (1.626)	1.66 (1.63)	1.66 (1.63)	1.67 (1.633)	1.68 (1.636)	1.72 (1.648)	1.73 (1.651)
V <sub>21</sub>	1.49 (1.577)	1.49 (1.577)	1.52 (1.586)	1.54 (1.594)	1.55 (1.597)	1.57 (1.604)	1.58 (1.607)	1.59 (1.61)	1.63 (1.624)	1.65 (1.627)	1.70 (1.642)	1.68 (1.636)
V <sub>22</sub>	1.49 (1.577)	1.50 (1.58)	1.52 (1.586)	1.55 (1.597)	1.56 (1.6)	1.59 (1.61)	1.59 (1.61)	1.62 (1.621)	1.65 (1.627)	1.67 (1.633)	1.72 (1.649)	1.71 (1.646)
V <sub>23</sub>	1.82 (1.678)	1.84 (1.685)	1.86 (1.691)	1.90 (1.703)	1.90 (1.703)	1.93 (1.712)	1.93 (1.712)	1.94 (1.715)	1.95 (1.718)	2.00 (1.731)	2.00 (1.731)	2.00 (1.731)
V <sub>24</sub>	1.74 (1.656)	1.76 (1.662)	1.77 (1.668)	1.80 (1.678)	1.82 (1.678)	1.82 (1.681)	1.82 (1.681)	1.84 (1.694)	1.88 (1.706)	1.92 (1.713)	1.94 (1.718)	1.96 (1.721)
V <sub>25</sub>	1.78 (1.668)	1.79 (1.671)	1.83 (1.678)	1.86 (1.687)	1.86 (1.691)	1.90 (1.699)	1.90 (1.699)	1.92 (1.699)	1.94 (1.706)	1.96 (1.718)	2.00 (1.728)	2.02 (1.737)
V <sub>26</sub>	1.52 (1.586)	1.54 (1.593)	1.55 (1.597)	1.57 (1.604)	1.57 (1.604)	1.63 (1.621)	1.62 (1.619)	1.63 (1.622)	1.66 (1.629)	1.66 (1.629)	1.69 (1.64)	1.71 (1.646)
V <sub>27</sub>	1.43 (1.559)	1.45 (1.566)	1.48 (1.573)	1.51 (1.584)	1.52 (1.587)	1.56 (1.6)	1.56 (1.6)	1.58 (1.607)	1.60 (1.613)	1.62 (1.62)	1.63 (1.623)	1.67 (1.634)
V <sub>28</sub>	1.45 (1.566)	1.48 (1.572)	1.53 (1.589)	1.58 (1.607)	1.57 (1.604)	1.60 (1.613)	1.60 (1.613)	1.61 (1.616)	1.65 (1.626)	1.63 (1.623)	1.63 (1.623)	1.66 (1.629)
V <sub>29</sub>	1.48 (1.572)	1.48 (1.572)	1.54 (1.593)	1.61 (1.617)	1.62 (1.621)	1.63 (1.624)	1.65 (1.627)	1.66 (1.63)	1.69 (1.639)	1.67 (1.633)	1.70 (1.643)	1.69 (1.64)
V <sub>30</sub>	1.04 (1.429)	1.05 (1.433)	1.07 (1.439)	1.09 (1.446)	1.10 (1.45)	1.14 (1.461)	1.15 (1.465)	1.16 (1.468)	1.18 (1.475)	1.18 (1.475)	1.18 (1.475)	1.19 (1.478)
<b>C.D.</b>	0.052	0.051	0.047	0.048	0.047	0.05	0.051	0.054	0.055	0.055	0.054	0.056
<b>SE(m)±</b>	0.018	0.018	0.017	0.017	0.017	0.018	0.018	0.019	0.019	0.02	0.019	0.02
<b>C.V.</b>	2.103	2.043	1.895	1.921	1.865	1.973	1.991	2.12	2.138	2.163	2.115	2.161

Note: The data in parenthesis indicate square root transformed values

**APPENDIX VIII. Shoot diameter *Ascocentrum* hybrids/varieties during the period 2017-18, cm**

Var. No.	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	1.24 (1.497)	1.28 (1.511)	1.28 (1.511)	1.32 (1.522)	1.32 (1.522)	1.30 (1.514)	1.33 (1.525)	1.35 (1.532)	1.42 (1.556)	1.45 (1.566)	1.45 (1.566)	1.45 (1.566)
V <sub>2</sub>	1.39 (1.546)	1.44 (1.563)	1.48 (1.574)	1.54 (1.594)	1.55 (1.597)	1.59 (1.609)	1.61 (1.617)	1.61 (1.617)	1.63 (1.624)	1.65 (1.627)	1.66 (1.63)	1.68 (1.636)
V <sub>3</sub>	1.73 (1.653)	1.76 (1.662)	1.77 (1.665)	1.82 (1.68)	1.82 (1.678)	1.84 (1.684)	1.84 (1.684)	1.87 (1.693)	1.89 (1.7)	1.90 (1.703)	1.92 (1.709)	1.93 (1.712)
V <sub>4</sub>	1.55 (1.595)	1.56 (1.598)	1.58 (1.606)	1.60 (1.613)	1.59 (1.609)	1.62 (1.619)	1.60 (1.613)	1.62 (1.619)	1.66 (1.629)	1.68 (1.635)	1.69 (1.638)	1.69 (1.638)
V <sub>5</sub>	1.28 (1.511)	1.33 (1.524)	1.36 (1.536)	1.37 (1.539)	1.39 (1.545)	1.39 (1.545)	1.39 (1.545)	1.43 (1.559)	1.44 (1.562)	1.45 (1.565)	1.46 (1.568)	1.48 (1.571)
V <sub>6</sub>	1.26 (1.503)	1.38 (1.541)	1.44 (1.563)	1.48 (1.573)	1.48 (1.573)	1.49 (1.575)	1.54 (1.594)	1.55 (1.597)	1.56 (1.6)	1.57 (1.604)	1.59 (1.61)	1.69 (1.64)
V <sub>7</sub>	1.68 (1.636)	1.70 (1.643)	1.73 (1.651)	1.74 (1.656)	1.75 (1.659)	1.76 (1.662)	1.76 (1.662)	1.77 (1.666)	1.78 (1.669)	1.79 (1.672)	1.79 (1.672)	1.79 (1.672)
V <sub>8</sub>	1.34 (1.527)	1.38 (1.544)	1.41 (1.553)	1.42 (1.557)	1.43 (1.56)	1.48 (1.573)	1.48 (1.573)	1.49 (1.576)	1.51 (1.583)	1.52 (1.587)	1.54 (1.594)	1.54 (1.594)
V <sub>9</sub>	0.89 (1.376)	0.92 (1.387)	0.94 (1.395)	0.97 (1.402)	0.98 (1.406)	0.99 (1.409)	0.99 (1.409)	1.01 (1.416)	1.03 (1.423)	1.04 (1.428)	1.04 (1.428)	1.08 (1.443)
V <sub>10</sub>	1.41 (1.552)	1.41 (1.552)	1.49 (1.576)	1.53 (1.59)	1.55 (1.597)	1.55 (1.597)	1.57 (1.604)	1.58 (1.607)	1.58 (1.607)	1.61 (1.616)	1.60 (1.613)	1.60 (1.613)
V <sub>11</sub>	1.18 (1.476)	1.19 (1.48)	1.21 (1.488)	1.22 (1.491)	1.24 (1.498)	1.25 (1.501)	1.25 (1.501)	1.30 (1.515)	1.32 (1.522)	1.33 (1.525)	1.34 (1.528)	1.33 (1.525)
V <sub>12</sub>	1.40 (1.549)	1.42 (1.556)	1.46 (1.569)	1.49 (1.577)	1.49 (1.577)	1.55 (1.597)	1.55 (1.597)	1.58 (1.606)	1.59 (1.609)	1.61 (1.617)	1.63 (1.624)	1.63 (1.624)
V <sub>13</sub>	1.52 (1.586)	1.52 (1.586)	1.54 (1.594)	1.57 (1.603)	1.58 (1.606)	1.61 (1.616)	1.60 (1.613)	1.65 (1.627)	1.67 (1.633)	1.68 (1.636)	1.68 (1.636)	1.69 (1.639)
V <sub>14</sub>	1.37 (1.538)	1.43 (1.56)	1.44 (1.563)	1.46 (1.57)	1.48 (1.574)	1.51 (1.583)	1.53 (1.589)	1.55 (1.596)	1.58 (1.606)	1.58 (1.606)	1.63 (1.624)	1.63 (1.624)
V <sub>15</sub>	1.43 (1.558)	1.52 (1.586)	1.54 (1.594)	1.57 (1.603)	1.57 (1.603)	1.59 (1.61)	1.63 (1.624)	1.63 (1.624)	1.65 (1.627)	1.66 (1.63)	1.69 (1.64)	1.70 (1.643)
V <sub>16</sub>	1.32 (1.521)	1.36 (1.536)	1.36 (1.536)	1.39 (1.546)	1.40 (1.549)	1.40 (1.548)	1.41 (1.552)	1.41 (1.552)	1.48 (1.572)	1.51 (1.582)	1.52 (1.585)	1.52 (1.585)



**APPENDIX VIII. continued**

Var. No.	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	1.09 (1.446)	1.11 (1.453)	1.13 (1.456)	1.15 (1.462)	1.17 (1.47)	1.19 (1.478)	1.21 (1.486)	1.22 (1.489)	1.22 (1.489)	1.23 (1.493)	1.24 (1.496)	1.26 (1.502)
V <sub>18</sub>	1.40 (1.548)	1.41 (1.552)	1.43 (1.559)	1.45 (1.565)	1.45 (1.565)	1.46 (1.569)	1.48 (1.573)	1.50 (1.581)	1.52 (1.587)	1.52 (1.587)	1.54 (1.593)	1.55 (1.596)
V <sub>19</sub>	1.68 (1.637)	1.69 (1.64)	1.71 (1.646)	1.73 (1.652)	1.75 (1.658)	1.76 (1.661)	1.77 (1.665)	1.78 (1.668)	1.80 (1.675)	1.84 (1.684)	1.86 (1.69)	1.88 (1.696)
V <sub>20</sub>	1.74 (1.655)	1.74 (1.655)	1.76 (1.66)	1.77 (1.664)	1.80 (1.673)	1.82 (1.677)	1.84 (1.683)	1.85 (1.686)	1.89 (1.699)	1.90 (1.702)	1.91 (1.705)	1.93 (1.712)
V <sub>21</sub>	1.67 (1.633)	1.68 (1.637)	1.71 (1.646)	1.74 (1.656)	1.74 (1.656)	1.76 (1.662)	1.80 (1.675)	1.82 (1.678)	1.84 (1.684)	1.86 (1.69)	1.86 (1.69)	1.90 (1.703)
V <sub>22</sub>	1.72 (1.649)	1.73 (1.652)	1.76 (1.662)	1.78 (1.669)	1.78 (1.669)	1.85 (1.687)	1.85 (1.687)	1.86 (1.69)	1.87 (1.693)	1.89 (1.699)	1.90 (1.703)	1.93 (1.711)
V <sub>23</sub>	2.02 (1.758)	2.03 (1.741)	2.04 (1.744)	2.06 (1.749)	2.08 (1.755)	2.09 (1.758)	2.12 (1.766)	2.13 (1.77)	2.15 (1.777)	2.18 (1.783)	2.20 (1.789)	2.22 (1.794)
V <sub>24</sub>	1.97 (1.727)	1.99 (1.73)	2.01 (1.733)	2.02 (1.736)	2.03 (1.736)	2.04 (1.742)	2.06 (1.75)	2.07 (1.75)	2.09 (1.759)	2.10 (1.759)	2.12 (1.764)	2.14 (1.77)
V <sub>25</sub>	2.05 (1.742)	2.08 (1.752)	2.10 (1.76)	2.10 (1.76)	2.12 (1.771)	2.13 (1.771)	2.17 (1.779)	2.18 (1.785)	2.20 (1.788)	2.21 (1.794)	2.24 (1.803)	2.26 (1.808)
V <sub>26</sub>	1.73 (1.652)	1.75 (1.658)	1.78 (1.667)	1.82 (1.678)	1.82 (1.678)	1.85 (1.687)	1.85 (1.688)	1.86 (1.691)	1.88 (1.697)	1.91 (1.705)	1.92 (1.708)	1.96 (1.72)
V <sub>27</sub>	1.68 (1.637)	1.69 (1.64)	1.71 (1.646)	1.71 (1.646)	1.73 (1.653)	1.76 (1.662)	1.76 (1.662)	1.77 (1.665)	1.79 (1.671)	1.83 (1.681)	1.87 (1.693)	1.89 (1.7)
V <sub>28</sub>	1.67 (1.632)	1.68 (1.636)	1.68 (1.636)	1.69 (1.642)	1.70 (1.645)	1.70 (1.645)	1.72 (1.649)	1.74 (1.655)	1.73 (1.653)	1.75 (1.659)	1.78 (1.668)	1.82 (1.677)
V <sub>29</sub>	1.72 (1.649)	1.71 (1.646)	1.72 (1.649)	1.81 (1.676)	1.84 (1.683)	1.85 (1.686)	1.87 (1.694)	1.87 (1.694)	1.90 (1.704)	1.93 (1.713)	1.96 (1.721)	2.01 (1.734)
V <sub>30</sub>	1.22 (1.49)	1.23 (1.493)	1.24 (1.496)	1.27 (1.509)	1.27 (1.509)	1.28 (1.512)	1.28 (1.512)	1.28 (1.512)	1.32 (1.522)	1.34 (1.528)	1.35 (1.532)	1.36 (1.535)
<b>C.D.</b>	0.058	0.058	0.059	0.062	0.062	0.063	0.064	0.063	0.062	0.063	0.065	0.067
<b>SE(m)±</b>	0.021	0.02	0.021	0.022	0.022	0.022	0.022	0.022	0.022	0.022	0.023	0.024
<b>C.V.</b>	2.256	2.225	2.256	2.346	2.336	2.373	2.405	2.363	2.331	2.362	2.415	2.486

Note: The data in parenthesis indicate square root transformed values

**APPENDIX IX. Internodal length of *Ascocentrum* hybrids/varieties during the period 2016-17, cm**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	1.23 (1.493)	1.30 (1.515)	1.33 (1.526)	1.43 (1.559)	1.50 (1.58)	1.63 (1.622)	1.63 (1.622)	1.63 (1.622)	1.73 (1.652)	1.80 (1.672)	1.90 (1.702)	1.97 (1.722)
V <sub>2</sub>	1.53 (1.59)	1.60 (1.611)	1.70 (1.642)	1.77 (1.663)	1.80 (1.673)	1.90 (1.702)	1.93 (1.712)	1.97 (1.722)	2.10 (1.76)	2.17 (1.779)	2.27 (1.807)	2.37 (1.835)
V <sub>3</sub>	2.53 (1.879)	2.63 (1.905)	2.73 (1.932)	2.80 (1.949)	2.83 (1.957)	2.77 (1.94)	2.80 (1.948)	2.83 (1.956)	2.90 (1.973)	3.00 (1.998)	3.10 (2.023)	3.20 (2.048)
V <sub>4</sub>	1.63 (1.622)	1.70 (1.643)	1.73 (1.652)	1.77 (1.663)	1.87 (1.692)	1.90 (1.702)	1.87 (1.693)	1.90 (1.703)	2.00 (1.732)	2.07 (1.751)	2.17 (1.779)	2.20 (1.789)
V <sub>5</sub>	0.80 (1.341)	0.90 (1.378)	1.07 (1.437)	1.20 (1.483)	1.30 (1.517)	1.43 (1.56)	1.47 (1.57)	1.47 (1.57)	1.57 (1.602)	1.67 (1.633)	1.77 (1.663)	1.80 (1.673)
V <sub>6</sub>	0.70 (1.303)	0.80 (1.341)	0.90 (1.377)	0.97 (1.401)	1.03 (1.424)	1.07 (1.436)	1.07 (1.436)	1.17 (1.471)	1.17 (1.471)	1.30 (1.516)	1.37 (1.537)	1.43 (1.559)
V <sub>7</sub>	0.70 (1.302)	0.83 (1.353)	0.90 (1.377)	1.00 (1.413)	1.10 (1.448)	1.10 (1.448)	1.13 (1.459)	1.23 (1.493)	1.33 (1.527)	1.47 (1.57)	1.57 (1.601)	1.63 (1.622)
V <sub>8</sub>	0.69 (1.301)	0.87 (1.365)	1.00 (1.413)	1.13 (1.46)	1.20 (1.482)	1.23 (1.494)	1.33 (1.527)	1.27 (1.504)	1.33 (1.525)	1.43 (1.558)	1.53 (1.59)	1.60 (1.61)
V <sub>9</sub>	0.27 (1.124)	0.43 (1.195)	0.53 (1.236)	0.66 (1.288)	0.73 (1.314)	0.80 (1.34)	0.97 (1.402)	1.00 (1.414)	1.10 (1.449)	1.20 (1.483)	1.13 (1.46)	1.13 (1.46)
V <sub>10</sub>	0.63 (1.277)	0.76 (1.329)	0.83 (1.354)	0.93 (1.39)	1.00 (1.414)	1.00 (1.414)	1.07 (1.437)	1.10 (1.449)	1.20 (1.483)	1.30 (1.516)	1.40 (1.549)	1.50 (1.58)
V <sub>11</sub>	0.60 (1.264)	0.73 (1.316)	0.97 (1.402)	1.07 (1.437)	1.20 (1.483)	1.23 (1.494)	1.20 (1.483)	1.27 (1.505)	1.33 (1.527)	1.40 (1.549)	1.50 (1.581)	1.57 (1.602)
V <sub>12</sub>	0.93 (1.39)	1.03 (1.426)	1.10 (1.449)	1.20 (1.483)	1.30 (1.516)	1.33 (1.527)	1.37 (1.538)	1.40 (1.549)	1.50 (1.581)	1.57 (1.602)	1.67 (1.633)	1.70 (1.643)
V <sub>13</sub>	0.97 (1.4)	1.13 (1.458)	1.20 (1.48)	1.33 (1.524)	1.47 (1.566)	1.47 (1.564)	1.53 (1.587)	1.60 (1.609)	1.67 (1.629)	1.73 (1.65)	1.80 (1.67)	1.90 (1.7)
V <sub>14</sub>	0.90 (1.376)	1.07 (1.434)	1.13 (1.456)	1.20 (1.479)	1.40 (1.546)	1.40 (1.545)	1.47 (1.568)	1.53 (1.59)	1.60 (1.61)	1.70 (1.641)	1.80 (1.671)	1.83 (1.681)
V <sub>15</sub>	1.00 (1.412)	1.23 (1.493)	1.33 (1.525)	1.47 (1.569)	1.57 (1.6)	1.57 (1.599)	1.63 (1.621)	1.63 (1.621)	1.73 (1.651)	1.83 (1.681)	1.93 (1.711)	1.97 (1.721)
V <sub>16</sub>	1.10 (1.444)	1.30 (1.512)	1.43 (1.556)	1.53 (1.588)	1.63 (1.62)	1.63 (1.619)	1.83 (1.68)	1.87 (1.691)	1.93 (1.711)	2.03 (1.74)	2.13 (1.769)	2.20 (1.788)

**APPENDIX IX. continued**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	0.63 (1.277)	0.77 (1.328)	0.93 (1.39)	1.10 (1.449)	1.20 (1.483)	1.13 (1.459)	1.10 (1.446)	1.07 (1.435)	1.13 (1.458)	1.27 (1.503)	1.37 (1.536)	1.50 (1.58)
V <sub>18</sub>	1.23 (1.494)	1.30 (1.516)	1.37 (1.537)	1.47 (1.57)	1.57 (1.602)	1.63 (1.622)	1.67 (1.633)	1.73 (1.653)	1.77 (1.663)	1.87 (1.693)	1.97 (1.722)	2.07 (1.751)
V <sub>19</sub>	2.80 (1.947)	2.90 (1.973)	2.93 (1.98)	3.03 (2.005)	3.00 (1.997)	3.00 (1.996)	3.07 (2.012)	3.17 (2.037)	3.23 (2.052)	3.30 (2.069)	3.37 (2.085)	3.43 (2.1)
V <sub>20</sub>	2.70 (1.923)	2.70 (1.923)	2.80 (1.949)	2.90 (1.974)	3.00 (1.999)	3.03 (2.007)	3.10 (2.024)	3.17 (2.04)	3.27 (2.065)	3.37 (2.089)	3.47 (2.113)	3.53 (2.128)
V <sub>21</sub>	3.10 (2.025)	3.23 (2.057)	3.43 (2.105)	3.50 (2.12)	3.63 (2.151)	3.63 (2.151)	3.67 (2.158)	3.73 (2.174)	3.80 (2.189)	3.90 (2.212)	4.00 (2.235)	4.03 (2.242)
V <sub>22</sub>	3.10 (2.025)	3.13 (2.033)	3.23 (2.057)	3.33 (2.081)	3.47 (2.113)	3.47 (2.113)	3.50 (2.121)	3.63 (2.152)	3.70 (2.168)	3.80 (2.191)	3.93 (2.221)	4.00 (2.236)
V <sub>23</sub>	3.73 (2.174)	3.83 (2.196)	3.93 (2.219)	4.03 (2.242)	4.10 (2.256)	4.13 (2.264)	4.17 (2.271)	4.27 (2.293)	4.37 (2.315)	4.47 (2.336)	4.57 (2.358)	4.60 (2.365)
V <sub>24</sub>	3.47 (2.065)	3.53 (2.089)	3.60 (2.121)	3.73 (2.145)	3.77 (2.145)	3.80 (2.152)	3.87 (2.168)	3.97 (2.191)	4.07 (2.213)	4.20 (2.243)	4.23 (2.251)	4.30 (2.265)
V <sub>25</sub>	3.47 (2.159)	3.57 (2.175)	3.73 (2.198)	3.77 (2.213)	3.80 (2.228)	3.83 (2.236)	3.93 (2.258)	4.03 (2.28)	4.13 (2.302)	4.23 (2.323)	4.27 (2.33)	4.33 (2.345)
V <sub>26</sub>	2.43 (1.852)	2.53 (1.879)	2.60 (1.896)	2.70 (1.922)	2.80 (1.948)	2.80 (1.948)	2.83 (1.956)	2.93 (1.981)	3.00 (1.999)	3.10 (2.024)	3.17 (2.04)	3.23 (2.057)
V <sub>27</sub>	2.53 (1.878)	2.60 (1.895)	2.60 (1.895)	2.73 (1.929)	2.83 (1.955)	2.87 (1.964)	2.87 (1.964)	2.93 (1.981)	3.00 (1.998)	3.10 (2.023)	3.20 (2.047)	3.30 (2.072)
V <sub>28</sub>	3.10 (2.025)	3.17 (2.041)	3.27 (2.065)	3.40 (2.098)	3.63 (2.152)	3.67 (2.16)	3.77 (2.182)	3.80 (2.19)	3.90 (2.213)	3.97 (2.228)	4.10 (2.258)	4.17 (2.273)
V <sub>29</sub>	2.50 (1.87)	2.60 (1.896)	2.73 (1.932)	2.77 (1.94)	2.90 (1.974)	2.97 (1.991)	3.03 (2.007)	3.07 (2.016)	3.07 (2.016)	3.17 (2.04)	3.17 (2.04)	3.23 (2.056)
V <sub>30</sub>	1.67 (1.632)	1.70 (1.641)	1.80 (1.672)	1.90 (1.702)	1.93 (1.712)	1.97 (1.721)	2.10 (1.76)	2.17 (1.778)	2.23 (1.797)	2.33 (1.824)	2.43 (1.852)	2.47 (1.861)
C.D.	0.123	0.132	0.133	0.13	0.132	0.142	0.144	0.138	0.137	0.134	0.133	0.133
SE(m)±	0.043	0.046	0.047	0.046	0.047	0.05	0.051	0.049	0.048	0.047	0.047	0.047
C.V.	4.61	4.83	4.78	4.592	4.612	4.925	4.967	4.707	4.616	4.439	4.374	4.317

Note: The data in parenthesis indicate square root transformed values

**APPENDIX X. Internodal length of *Ascocentrum* hybrids/varieties during the period 2017-18, cm**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	2.00 (1.731)	2.10 (1.76)	2.13 (1.769)	2.20 (1.787)	2.30 (1.815)	2.30 (1.816)	2.37 (1.834)	2.43 (1.852)	2.50 (1.87)	2.60 (1.896)	2.63 (1.905)	2.70 (1.923)
V <sub>2</sub>	2.43 (1.853)	2.47 (1.862)	2.57 (1.888)	2.67 (1.915)	2.70 (1.923)	2.70 (1.923)	2.77 (1.941)	2.83 (1.958)	2.93 (1.983)	3.03 (2.008)	3.10 (2.025)	3.17 (2.041)
V <sub>3</sub>	3.20 (2.048)	3.23 (2.056)	3.33 (2.08)	3.43 (2.104)	3.53 (2.128)	3.67 (2.158)	3.67 (2.158)	3.67 (2.158)	3.77 (2.182)	3.83 (2.196)	3.90 (2.212)	3.90 (2.212)
V <sub>4</sub>	2.23 (1.798)	2.33 (1.825)	2.40 (1.844)	2.47 (1.862)	2.57 (1.889)	2.60 (1.897)	2.63 (1.906)	2.43 (1.849)	2.47 (1.857)	2.50 (1.866)	2.57 (1.883)	2.60 (1.891)
V <sub>5</sub>	1.80 (1.673)	1.90 (1.702)	1.97 (1.721)	2.03 (1.74)	2.13 (1.769)	2.17 (1.778)	2.23 (1.797)	2.33 (1.825)	2.43 (1.852)	2.50 (1.87)	2.50 (1.87)	2.57 (1.888)
V <sub>6</sub>	1.47 (1.57)	1.57 (1.601)	1.63 (1.622)	1.73 (1.653)	1.83 (1.683)	1.83 (1.683)	1.90 (1.702)	1.97 (1.722)	2.03 (1.741)	2.13 (1.77)	2.20 (1.789)	2.30 (1.816)
V <sub>7</sub>	1.70 (1.642)	1.80 (1.673)	1.93 (1.712)	2.00 (1.731)	2.10 (1.76)	2.13 (1.769)	2.23 (1.797)	2.30 (1.816)	2.40 (1.843)	2.50 (1.87)	2.57 (1.887)	2.67 (1.914)
V <sub>8</sub>	1.60 (1.61)	1.70 (1.641)	1.77 (1.661)	1.83 (1.681)	1.90 (1.7)	1.97 (1.72)	2.03 (1.74)	2.10 (1.759)	2.20 (1.788)	2.30 (1.815)	2.37 (1.834)	2.43 (1.851)
V <sub>9</sub>	1.17 (1.472)	1.27 (1.505)	1.43 (1.559)	1.53 (1.591)	1.63 (1.622)	1.67 (1.633)	1.60 (1.612)	1.73 (1.653)	1.83 (1.683)	1.97 (1.722)	2.03 (1.741)	2.10 (1.76)
V <sub>10</sub>	1.57 (1.6)	1.67 (1.631)	1.83 (1.682)	1.93 (1.712)	2.03 (1.741)	2.07 (1.751)	2.03 (1.741)	2.17 (1.779)	2.27 (1.806)	2.33 (1.825)	2.43 (1.852)	2.53 (1.879)
V <sub>11</sub>	1.63 (1.623)	1.73 (1.653)	1.80 (1.673)	1.87 (1.693)	1.97 (1.722)	1.87 (1.693)	1.97 (1.722)	2.03 (1.742)	2.13 (1.77)	2.23 (1.798)	2.30 (1.817)	2.37 (1.835)
V <sub>12</sub>	1.70 (1.643)	1.77 (1.663)	1.87 (1.693)	1.87 (1.693)	1.90 (1.702)	2.00 (1.732)	2.10 (1.76)	2.20 (1.788)	2.23 (1.797)	2.33 (1.825)	2.37 (1.834)	2.43 (1.852)
V <sub>13</sub>	1.90 (1.7)	1.93 (1.71)	2.00 (1.729)	2.07 (1.748)	2.10 (1.758)	2.07 (1.748)	2.17 (1.776)	2.20 (1.786)	2.27 (1.805)	2.37 (1.833)	2.43 (1.852)	2.53 (1.878)
V <sub>14</sub>	1.83 (1.681)	1.93 (1.711)	1.97 (1.721)	2.00 (1.73)	2.07 (1.749)	2.13 (1.769)	2.20 (1.788)	2.27 (1.806)	2.30 (1.816)	2.40 (1.843)	2.43 (1.852)	2.50 (1.87)
V <sub>15</sub>	2.00 (1.731)	2.07 (1.751)	2.17 (1.779)	2.30 (1.816)	2.40 (1.844)	2.43 (1.853)	2.43 (1.853)	2.53 (1.879)	2.63 (1.906)	2.73 (1.932)	2.83 (1.958)	2.93 (1.983)
V <sub>16</sub>	2.30 (1.816)	2.40 (1.843)	2.50 (1.87)	2.67 (1.912)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)	2.83 (1.957)	2.90 (1.973)	3.00 (1.999)	3.10 (2.024)	3.17 (2.04)

**APPENDIX X. continued**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	1.57 (1.6)	1.63 (1.621)	1.70 (1.642)	1.77 (1.662)	1.87 (1.692)	1.93 (1.711)	2.03 (1.74)	2.07 (1.75)	2.13 (1.769)	2.23 (1.798)	2.30 (1.816)	2.37 (1.834)
V <sub>18</sub>	2.10 (1.761)	2.20 (1.788)	2.33 (1.825)	2.43 (1.853)	2.57 (1.888)	2.57 (1.888)	2.57 (1.888)	2.67 (1.915)	2.77 (1.941)	2.87 (1.966)	2.93 (1.983)	2.97 (1.991)
V <sub>19</sub>	3.47 (2.109)	3.53 (2.124)	3.63 (2.148)	3.70 (2.163)	3.80 (2.186)	3.87 (2.201)	3.97 (2.224)	4.03 (2.239)	4.13 (2.262)	4.20 (2.276)	4.23 (2.283)	4.27 (2.291)
V <sub>20</sub>	3.57 (2.137)	3.60 (2.144)	3.60 (2.144)	3.77 (2.183)	3.83 (2.198)	3.97 (2.228)	4.03 (2.243)	4.10 (2.258)	4.17 (2.273)	4.27 (2.295)	4.33 (2.309)	4.40 (2.324)
V <sub>21</sub>	4.07 (2.25)	4.17 (2.272)	4.23 (2.287)	4.30 (2.302)	4.33 (2.309)	4.43 (2.33)	4.50 (2.345)	4.53 (2.352)	4.63 (2.373)	4.70 (2.387)	4.77 (2.401)	4.77 (2.401)
V <sub>22</sub>	4.07 (2.251)	4.13 (2.265)	4.20 (2.28)	4.30 (2.302)	4.37 (2.316)	4.47 (2.338)	4.57 (2.359)	4.63 (2.373)	4.67 (2.38)	4.70 (2.387)	4.70 (2.387)	4.80 (2.408)
V <sub>23</sub>	4.63 (2.372)	4.70 (2.386)	4.80 (2.407)	4.87 (2.421)	4.93 (2.434)	5.00 (2.448)	5.00 (2.448)	5.03 (2.455)	5.10 (2.468)	5.13 (2.475)	5.13 (2.475)	5.13 (2.475)
V <sub>24</sub>	4.33 (2.273)	4.40 (2.287)	4.50 (2.309)	4.57 (2.331)	4.60 (2.345)	4.67 (2.359)	4.70 (2.366)	4.77 (2.373)	4.80 (2.373)	4.83 (2.38)	4.93 (2.394)	4.97 (2.394)
V <sub>25</sub>	4.40 (2.359)	4.47 (2.373)	4.53 (2.387)	4.60 (2.394)	4.67 (2.401)	4.70 (2.408)	4.73 (2.415)	4.80 (2.436)	4.83 (2.449)	4.90 (2.462)	4.97 (2.483)	5.00 (2.496)
V <sub>26</sub>	3.27 (2.065)	3.33 (2.081)	3.43 (2.105)	3.50 (2.121)	3.60 (2.144)	3.67 (2.159)	3.77 (2.182)	3.90 (2.213)	3.93 (2.22)	4.07 (2.25)	4.13 (2.265)	4.20 (2.28)
V <sub>27</sub>	3.37 (2.088)	3.40 (2.096)	3.53 (2.127)	3.60 (2.143)	3.70 (2.166)	3.70 (2.166)	3.80 (2.189)	3.90 (2.212)	4.00 (2.234)	4.10 (2.257)	4.13 (2.264)	4.17 (2.272)
V <sub>28</sub>	4.20 (2.28)	4.23 (2.287)	4.30 (2.302)	4.40 (2.323)	4.43 (2.33)	4.50 (2.345)	4.50 (2.345)	4.53 (2.352)	4.63 (2.373)	4.67 (2.38)	4.73 (2.394)	4.77 (2.401)
V <sub>29</sub>	3.27 (2.065)	3.33 (2.081)	3.37 (2.089)	3.47 (2.113)	3.53 (2.128)	3.63 (2.152)	3.77 (2.183)	3.87 (2.206)	3.93 (2.221)	4.00 (2.236)	4.07 (2.251)	4.13 (2.266)
V <sub>30</sub>	2.47 (1.861)	2.53 (1.879)	2.80 (1.949)	2.90 (1.975)	3.00 (2)	3.00 (2)	3.00 (2)	3.10 (2.025)	3.17 (2.041)	3.27 (2.065)	3.37 (2.09)	3.43 (2.105)
C.D.	0.129	0.13	0.126	0.129	0.128	0.125	0.121	0.123	0.123	0.124	0.123	0.124
SE(m) <sup>±</sup>	0.046	0.046	0.044	0.046	0.045	0.044	0.043	0.043	0.043	0.044	0.043	0.044
C.V.	4.182	4.17	3.979	4.036	3.968	3.834	3.701	3.735	3.698	3.687	3.624	3.62

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XI. Number of leaves of *Ascocentrum* hybrids/varieties during the period 2016-17**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	11.40 (3.467)	12.00 (3.562)	11.80 (3.536)	12.60 (3.639)	12.80 (3.678)	13.40 (3.754)	14.40 (3.888)	15.40 (4.017)	16.00 (4.087)	16.60 (4.164)	17.80 (4.306)	18.60 (4.393)
V <sub>2</sub>	9.40 (3.203)	10.20 (3.329)	10.20 (3.333)	11.20 (3.481)	11.20 (3.481)	12.20 (3.623)	13.00 (3.728)	13.60 (3.809)	14.40 (3.91)	15.00 (3.989)	15.60 (4.067)	16.60 (4.189)
V <sub>3</sub>	15.00 (3.996)	15.60 (4.07)	15.60 (4.067)	16.60 (4.188)	17.20 (4.258)	17.60 (4.306)	18.20 (4.375)	19.20 (4.488)	19.80 (4.553)	20.60 (4.64)	21.40 (4.725)	22.00 (4.789)
V <sub>4</sub>	16.40 (4.104)	17.00 (4.187)	17.60 (4.268)	18.20 (4.346)	18.40 (4.365)	19.20 (4.451)	20.00 (4.547)	20.60 (4.618)	21.60 (4.726)	22.60 (4.832)	23.60 (4.936)	24.40 (5.017)
V <sub>5</sub>	14.80 (3.952)	15.40 (4.032)	15.60 (4.062)	16.40 (4.163)	16.40 (4.163)	17.20 (4.256)	18.20 (4.373)	18.80 (4.443)	19.80 (4.555)	20.60 (4.641)	21.20 (4.704)	21.40 (4.726)
V <sub>6</sub>	16.20 (4.137)	17.00 (4.236)	17.60 (4.31)	17.80 (4.334)	18.40 (4.403)	19.20 (4.493)	20.00 (4.581)	20.40 (4.624)	21.00 (4.689)	22.00 (4.794)	22.60 (4.856)	22.80 (4.877)
V <sub>7</sub>	9.40 (3.207)	10.40 (3.361)	11.20 (3.477)	12.20 (3.622)	12.60 (3.674)	13.40 (3.783)	14.40 (3.913)	15.20 (4.012)	16.00 (4.114)	16.20 (4.14)	17.00 (4.23)	18.00 (4.352)
V <sub>8</sub>	22.40 (4.833)	23.20 (4.912)	23.80 (4.971)	23.80 (4.971)	24.60 (5.049)	25.00 (5.091)	26.00 (5.189)	26.40 (5.228)	27.20 (5.303)	27.40 (5.324)	28.20 (5.399)	28.40 (5.417)
V <sub>9</sub>	5.80 (2.598)	6.40 (2.706)	6.80 (2.775)	7.40 (2.886)	7.80 (2.951)	8.80 (3.118)	9.20 (3.187)	10.20 (3.341)	10.80 (3.426)	11.60 (3.541)	12.00 (3.6)	12.80 (3.71)
V <sub>10</sub>	11.60 (3.53)	12.20 (3.612)	13.20 (3.742)	13.40 (3.772)	14.20 (3.875)	14.40 (3.903)	15.20 (4.005)	16.00 (4.108)	16.60 (4.179)	17.00 (4.229)	17.40 (4.273)	18.20 (4.37)
V <sub>11</sub>	8.40 (3.045)	9.20 (3.17)	10.20 (3.326)	10.20 (3.326)	11.20 (3.475)	12.00 (3.585)	12.40 (3.646)	13.00 (3.723)	13.40 (3.781)	14.40 (3.912)	14.40 (3.912)	15.20 (4.015)
V <sub>12</sub>	14.00 (3.863)	14.80 (3.967)	15.60 (4.065)	15.80 (4.092)	16.60 (4.187)	17.20 (4.257)	18.00 (4.349)	18.80 (4.441)	19.40 (4.508)	20.20 (4.596)	20.60 (4.637)	20.60 (4.639)
V <sub>13</sub>	13.60 (3.808)	14.20 (3.889)	15.20 (4.016)	15.20 (4.016)	16.20 (4.139)	16.60 (4.188)	17.20 (4.26)	17.60 (4.306)	18.40 (4.398)	18.20 (4.37)	18.80 (4.439)	19.40 (4.505)
V <sub>14</sub>	11.40 (3.498)	12.40 (3.64)	13.00 (3.724)	13.40 (3.776)	14.20 (3.885)	15.00 (3.988)	15.60 (4.064)	16.20 (4.139)	16.80 (4.208)	17.60 (4.3)	18.20 (4.37)	19.20 (4.484)
V <sub>15</sub>	12.20 (3.608)	13.20 (3.745)	14.00 (3.852)	14.20 (3.878)	15.20 (4.006)	16.00 (4.107)	16.60 (4.178)	17.40 (4.275)	18.00 (4.341)	18.40 (4.385)	19.20 (4.477)	19.80 (4.542)
V <sub>16</sub>	5.40 (2.514)	6.00 (2.626)	6.80 (2.764)	7.20 (2.845)	7.60 (2.912)	8.20 (3.018)	9.00 (3.15)	9.60 (3.247)	10.20 (3.341)	10.60 (3.397)	11.20 (3.488)	11.40 (3.515)
V <sub>17</sub>	9.60 (3.23)	10.40 (3.356)	11.40 (3.504)	11.40 (3.504)	12.40 (3.645)	13.00 (3.72)	13.60 (3.798)	14.40 (3.905)	14.80 (3.957)	15.40 (4.029)	16.00 (4.108)	16.60 (4.178)

**APPENDIX XI. continued**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>18</sub>	11.80 (3.563)	12.80 (3.702)	14.00 (3.864)	14.00 (3.864)	15.00 (3.992)	15.20 (4.014)	16.00 (4.115)	16.60 (4.186)	17.20 (4.258)	17.80 (4.325)	18.60 (4.419)	19.20 (4.484)
V <sub>19</sub>	22.60 (4.857)	23.00 (4.898)	23.60 (4.959)	24.20 (5.019)	25.00 (5.098)	25.60 (5.156)	26.40 (5.234)	27.20 (5.309)	27.80 (5.365)	28.00 (5.384)	28.60 (5.44)	29.00 (5.476)
V <sub>20</sub>	20.60 (4.628)	21.60 (4.735)	22.40 (4.822)	22.60 (4.84)	23.60 (4.943)	24.40 (5.023)	25.00 (5.082)	26.00 (5.18)	26.80 (5.256)	27.20 (5.293)	28.00 (5.368)	28.20 (5.384)
V <sub>21</sub>	31.40 (5.683)	32.00 (5.736)	32.80 (5.807)	33.40 (5.859)	35.20 (6.009)	36.20 (6.09)	37.20 (6.172)	38.00 (6.235)	39.00 (6.315)	39.80 (6.376)	40.60 (6.439)	41.40 (6.499)
V <sub>22</sub>	29.40 (5.487)	30.40 (5.579)	31.00 (5.636)	32.00 (5.721)	33.20 (5.827)	33.80 (5.878)	35.20 (5.996)	36.00 (6.064)	37.20 (6.161)	38.00 (6.227)	39.40 (6.338)	39.80 (6.367)
V <sub>23</sub>	53.80 (7.388)	54.60 (7.44)	55.40 (7.495)	56.00 (7.536)	57.20 (7.615)	58.00 (7.666)	58.80 (7.716)	59.80 (7.781)	60.60 (7.831)	61.60 (7.895)	62.80 (7.968)	63.40 (8.007)
V <sub>24</sub>	46.40 (6.882)	47.80 (6.982)	47.80 (6.982)	49.40 (7.095)	49.80 (7.123)	50.80 (7.193)	51.60 (7.248)	52.60 (7.317)	53.60 (7.385)	54.60 (7.453)	55.60 (7.52)	56.20 (7.559)
V <sub>25</sub>	54.00 (7.412)	54.60 (7.453)	55.60 (7.519)	56.00 (7.546)	57.20 (7.625)	58.20 (7.69)	59.00 (7.743)	60.00 (7.807)	61.00 (7.871)	61.80 (7.922)	62.80 (7.985)	63.80 (8.047)
V <sub>26</sub>	25.60 (5.085)	26.60 (5.185)	27.60 (5.282)	27.60 (5.282)	29.00 (5.418)	29.60 (5.472)	30.20 (5.532)	30.80 (5.584)	31.60 (5.655)	32.20 (5.713)	33.00 (5.782)	33.60 (5.837)
V <sub>27</sub>	24.60 (4.921)	25.80 (5.048)	26.80 (5.151)	27.00 (5.166)	27.80 (5.252)	28.40 (5.302)	29.20 (5.384)	29.60 (5.414)	30.40 (5.494)	31.00 (5.542)	31.80 (5.62)	32.40 (5.667)
V <sub>28</sub>	34.60 (5.884)	35.60 (5.971)	36.20 (6.022)	36.60 (6.057)	37.80 (6.155)	38.60 (6.219)	39.40 (6.286)	40.20 (6.342)	40.80 (6.394)	41.60 (6.455)	42.60 (6.533)	43.20 (6.574)
V <sub>29</sub>	18.40 (4.403)	19.40 (4.515)	20.20 (4.602)	20.60 (4.645)	21.40 (4.732)	21.60 (4.753)	22.40 (4.836)	22.80 (4.877)	23.80 (4.979)	24.00 (4.998)	25.00 (5.097)	25.20 (5.117)
V <sub>30</sub>	26.40 (5.074)	27.40 (5.179)	28.00 (5.235)	28.40 (5.281)	29.40 (5.381)	30.40 (5.479)	31.00 (5.526)	31.80 (5.605)	32.40 (5.651)	33.40 (5.744)	34.00 (5.788)	34.80 (5.864)
C.D.	0.742	0.722	0.712	0.701	0.688	0.694	0.682	0.678	0.681	0.68	0.679	0.68
SE(M)	0.265	0.258	0.254	0.25	0.245	0.248	0.243	0.242	0.243	0.243	0.242	0.243
C.V.	13.467	12.818	12.421	12.081	11.648	11.568	11.174	10.935	10.808	10.661	10.495	10.395

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XII. Number of leaves of *Ascocentrum* hybrids/varieties during the period 2017-18**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	18.80 (4.414)	18.80 (4.414)	19.40 (4.478)	19.80 (4.528)	20.20 (4.57)	21.00 (4.66)	21.40 (4.708)	22.40 (4.814)	23.00 (4.879)	23.00 (4.879)	23.80 (4.957)	24.40 (5.02)
V <sub>2</sub>	16.60 (4.189)	17.00 (4.238)	17.80 (4.328)	18.20 (4.376)	19.00 (4.466)	19.80 (4.556)	20.80 (4.665)	21.20 (4.706)	21.60 (4.748)	22.20 (4.813)	22.80 (4.875)	23.20 (4.916)
V <sub>3</sub>	22.00 (4.789)	22.40 (4.828)	23.40 (4.931)	23.60 (4.953)	24.40 (5.034)	24.60 (5.053)	24.80 (5.071)	25.40 (5.132)	25.80 (5.17)	26.40 (5.229)	27.20 (5.305)	27.40 (5.324)
V <sub>4</sub>	24.40 (5.017)	25.00 (5.076)	25.40 (5.117)	26.40 (5.211)	27.20 (5.285)	28.00 (5.36)	28.80 (5.435)	29.60 (5.508)	30.20 (5.562)	30.40 (5.581)	31.40 (5.671)	31.40 (5.671)
V <sub>5</sub>	22.00 (4.789)	22.40 (4.829)	23.20 (4.911)	23.20 (4.911)	24.20 (5.012)	24.40 (5.033)	25.00 (5.09)	25.60 (5.151)	26.60 (5.248)	26.80 (5.268)	27.60 (5.342)	27.80 (5.361)
V <sub>6</sub>	23.20 (4.918)	23.60 (4.958)	24.40 (5.038)	25.20 (5.117)	25.60 (5.156)	25.80 (5.175)	26.40 (5.233)	26.80 (5.27)	27.00 (5.29)	27.20 (5.307)	27.80 (5.364)	28.60 (5.438)
V <sub>7</sub>	18.00 (4.352)	19.00 (4.465)	19.40 (4.51)	20.00 (4.577)	20.60 (4.642)	20.80 (4.665)	21.20 (4.707)	22.00 (4.791)	22.40 (4.834)	23.00 (4.896)	23.80 (4.976)	24.40 (5.036)
V <sub>8</sub>	28.40 (5.417)	29.40 (5.509)	30.20 (5.582)	31.00 (5.652)	31.40 (5.688)	32.20 (5.757)	33.00 (5.824)	33.80 (5.892)	35.00 (5.991)	36.00 (6.073)	36.80 (6.137)	37.20 (6.171)
V <sub>9</sub>	13.00 (3.732)	14.80 (3.972)	15.60 (4.071)	16.00 (4.118)	16.80 (4.217)	17.20 (4.263)	17.80 (4.334)	18.40 (4.403)	19.20 (4.493)	19.60 (4.537)	20.00 (4.581)	20.60 (4.645)
V <sub>10</sub>	18.40 (4.389)	18.60 (4.413)	19.40 (4.502)	19.80 (4.544)	20.40 (4.608)	20.80 (4.654)	21.00 (4.673)	21.60 (4.739)	22.20 (4.801)	22.40 (4.822)	23.20 (4.904)	23.80 (4.968)
V <sub>11</sub>	15.80 (4.088)	16.20 (4.136)	16.20 (4.137)	16.20 (4.137)	17.00 (4.233)	17.40 (4.282)	17.40 (4.282)	17.60 (4.305)	17.80 (4.325)	18.00 (4.348)	18.20 (4.368)	19.00 (4.458)
V <sub>12</sub>	21.20 (4.704)	22.20 (4.81)	22.80 (4.872)	23.40 (4.931)	23.80 (4.971)	24.20 (5.014)	24.40 (5.027)	25.20 (5.108)	25.80 (5.164)	26.20 (5.202)	27.20 (5.298)	27.60 (5.336)
V <sub>13</sub>	19.40 (4.503)	20.20 (4.59)	21.00 (4.676)	21.00 (4.676)	21.60 (4.744)	22.40 (4.826)	22.00 (4.784)	22.80 (4.866)	23.60 (4.946)	23.60 (4.946)	24.60 (5.047)	25.00 (5.089)
V <sub>14</sub>	19.20 (4.487)	19.80 (4.553)	20.20 (4.598)	20.60 (4.642)	21.00 (4.685)	21.80 (4.77)	22.60 (4.852)	22.60 (4.852)	23.00 (4.894)	24.00 (4.996)	24.40 (5.037)	24.40 (5.038)
V <sub>15</sub>	19.80 (4.535)	20.00 (4.559)	20.80 (4.644)	21.60 (4.728)	22.00 (4.767)	22.80 (4.851)	23.40 (4.911)	24.20 (4.991)	24.40 (5.011)	25.00 (5.072)	25.40 (5.114)	25.60 (5.133)
V <sub>16</sub>	11.80 (3.57)	12.20 (3.628)	13.00 (3.736)	13.60 (3.817)	14.00 (3.87)	15.00 (3.995)	15.40 (4.046)	15.80 (4.095)	16.60 (4.193)	17.00 (4.241)	17.20 (4.264)	17.20 (4.264)
V <sub>17</sub>	17.00 (4.228)	17.40 (4.275)	18.00 (4.348)	18.60 (4.416)	18.80 (4.439)	19.80 (4.547)	20.00 (4.572)	20.60 (4.637)	21.60 (4.743)	22.00 (4.787)	22.60 (4.848)	23.00 (4.89)



**APPENDIX XII. continued**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>18</sub>	19.60 (4.531)	20.20 (4.597)	20.20 (4.591)	20.60 (4.631)	21.00 (4.675)	21.60 (4.74)	21.80 (4.759)	22.40 (4.823)	23.20 (4.908)	23.60 (4.948)	24.40 (5.029)	24.80 (5.07)
V <sub>19</sub>	29.20 (5.493)	30.00 (5.565)	30.60 (5.618)	31.20 (5.671)	32.40 (5.775)	33.40 (5.86)	34.40 (5.944)	35.20 (6.01)	36.20 (6.093)	37.20 (6.173)	38.00 (6.238)	38.80 (6.302)
V <sub>20</sub>	28.60 (5.421)	29.60 (5.513)	30.00 (5.546)	30.60 (5.594)	31.40 (5.669)	31.60 (5.685)	32.60 (5.773)	33.00 (5.808)	33.60 (5.861)	34.00 (5.891)	35.00 (5.976)	35.40 (6.008)
V <sub>21</sub>	42.20 (6.561)	43.00 (6.623)	43.80 (6.684)	44.80 (6.759)	45.60 (6.818)	46.80 (6.906)	47.40 (6.948)	48.40 (7.019)	49.40 (7.091)	50.20 (7.149)	51.00 (7.204)	51.60 (7.244)
V <sub>22</sub>	40.40 (6.418)	41.00 (6.463)	42.20 (6.555)	43.20 (6.629)	44.00 (6.689)	44.60 (6.736)	45.60 (6.811)	46.60 (6.884)	47.20 (6.929)	48.40 (7.015)	49.00 (7.057)	50.00 (7.128)
V <sub>23</sub>	64.20 (8.055)	65.00 (8.106)	66.00 (8.167)	66.80 (8.217)	67.80 (8.278)	68.40 (8.314)	69.00 (8.349)	70.00 (8.409)	70.60 (8.445)	71.40 (8.491)	72.20 (8.539)	72.80 (8.572)
V <sub>24</sub>	57.00 (7.612)	58.00 (7.677)	58.40 (7.703)	59.20 (7.755)	60.00 (7.807)	60.40 (7.833)	61.20 (7.883)	62.00 (7.933)	62.60 (7.972)	63.80 (8.046)	64.80 (8.108)	65.60 (8.158)
V <sub>25</sub>	64.40 (8.084)	65.40 (8.146)	66.40 (8.207)	67.20 (8.255)	68.20 (8.316)	69.20 (8.376)	70.20 (8.435)	70.80 (8.47)	71.80 (8.529)	72.80 (8.588)	73.80 (8.646)	74.80 (8.703)
V <sub>26</sub>	34.60 (5.921)	35.60 (6.006)	36.60 (6.09)	37.20 (6.138)	37.80 (6.185)	39.20 (6.301)	39.80 (6.348)	40.60 (6.41)	41.20 (6.456)	42.20 (6.534)	42.80 (6.579)	43.40 (6.623)
V <sub>27</sub>	33.20 (5.743)	34.00 (5.817)	35.00 (5.906)	35.60 (5.953)	36.00 (5.982)	36.80 (6.053)	37.60 (6.119)	38.40 (6.181)	39.20 (6.246)	39.80 (6.291)	40.60 (6.357)	41.40 (6.425)
V <sub>28</sub>	43.80 (6.62)	44.80 (6.696)	45.80 (6.772)	46.40 (6.816)	47.20 (6.878)	48.00 (6.934)	48.80 (6.99)	49.60 (7.05)	50.60 (7.122)	50.80 (7.139)	51.80 (7.21)	52.20 (7.235)
V <sub>29</sub>	26.20 (5.214)	27.20 (5.309)	27.60 (5.347)	28.40 (5.421)	29.20 (5.494)	29.80 (5.549)	30.60 (5.621)	31.40 (5.692)	32.20 (5.761)	32.60 (5.795)	33.20 (5.847)	34.20 (5.932)
V <sub>30</sub>	35.40 (5.913)	36.00 (5.962)	36.60 (6.016)	37.40 (6.078)	38.20 (6.149)	38.80 (6.196)	39.20 (6.224)	39.80 (6.28)	40.60 (6.348)	41.40 (6.412)	41.80 (6.443)	42.60 (6.51)
C.D.	0.682	0.675	0.673	0.683	0.678	0.673	0.681	0.674	0.668	0.667	0.666	0.658
SE(M)±	0.243	0.241	0.24	0.244	0.242	0.24	0.243	0.24	0.238	0.238	0.238	0.235
C.V.	10.346	10.117	9.967	10.007	9.819	9.645	9.68	9.471	9.296	9.193	9.091	8.906

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XIII. Leaf length of *Asocentrum* hybrids/varieties during the period 2016-17, cm**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	25.06 (5.099)	25.60 (5.151)	26.06 (5.196)	26.54 (5.242)	26.86 (5.272)	27.26 (5.308)	27.46 (5.327)	27.52 (5.332)	27.66 (5.344)	27.66 (5.344)	27.66 (5.344)	27.66 (5.344)
V <sub>2</sub>	23.60 (4.955)	24.52 (5.048)	25.04 (5.099)	25.48 (5.142)	26.16 (5.208)	26.40 (5.23)	26.50 (5.239)	26.50 (5.239)	26.50 (5.239)	26.50 (5.239)	26.50 (5.239)	26.50 (5.239)
V <sub>3</sub>	35.20 (6.014)	35.70 (6.056)	36.14 (6.093)	36.74 (6.142)	37.06 (6.168)	37.40 (6.195)	37.40 (6.195)	37.40 (6.195)	37.40 (6.195)	37.40 (6.195)	37.40 (6.195)	37.40 (6.195)
V <sub>4</sub>	23.10 (4.904)	23.58 (4.953)	24.04 (4.999)	24.58 (5.052)	25.16 (5.11)	25.38 (5.132)	25.64 (5.157)	25.66 (5.16)	25.82 (5.176)	25.82 (5.176)	25.82 (5.176)	25.82 (5.176)
V <sub>5</sub>	24.60 (5.041)	25.10 (5.089)	25.60 (5.138)	24.14 (4.982)	24.58 (5.028)	24.92 (5.065)	24.94 (5.067)	24.94 (5.067)	24.94 (5.067)	24.94 (5.067)	24.94 (5.067)	24.94 (5.067)
V <sub>6</sub>	22.80 (4.86)	23.54 (4.937)	23.98 (4.981)	24.51 (5.033)	24.80 (5.064)	25.22 (5.107)	25.22 (5.107)	25.22 (5.107)	25.22 (5.107)	25.22 (5.107)	25.22 (5.107)	25.22 (5.107)
V <sub>7</sub>	21.20 (4.71)	21.82 (4.776)	22.20 (4.815)	22.76 (4.873)	23.28 (4.926)	23.60 (4.959)	23.74 (4.973)	23.74 (4.973)	23.74 (4.973)	23.74 (4.973)	23.74 (4.973)	23.74 (4.973)
V <sub>8</sub>	23.50 (4.946)	24.10 (5.006)	24.72 (5.068)	25.06 (5.101)	25.80 (5.174)	26.48 (5.239)	26.80 (5.27)	26.94 (5.284)	27.02 (5.291)	27.02 (5.291)	27.02 (5.291)	27.02 (5.291)
V <sub>9</sub>	8.60 (3.084)	9.16 (3.177)	9.64 (3.251)	10.26 (3.345)	10.86 (3.435)	11.52 (3.532)	12.02 (3.603)	12.42 (3.659)	12.62 (3.688)	12.82 (3.716)	13.02 (3.744)	13.02 (3.744)
V <sub>10</sub>	22.68 (4.861)	23.32 (4.927)	23.98 (4.993)	24.72 (5.067)	24.86 (5.081)	25.02 (5.097)	25.18 (5.114)	25.18 (5.114)	25.18 (5.114)	25.18 (5.114)	24.02 (5.002)	24.68 (5.067)
V <sub>11</sub>	10.90 (3.423)	11.48 (3.51)	12.02 (3.587)	12.70 (3.683)	13.36 (3.774)	14.04 (3.864)	14.32 (3.903)	14.62 (3.943)	14.62 (3.943)	14.62 (3.943)	14.62 (3.943)	14.62 (3.943)
V <sub>12</sub>	14.20 (3.829)	14.50 (3.877)	15.02 (3.943)	15.50 (4.006)	16.16 (4.087)	16.86 (4.177)	17.48 (4.251)	17.78 (4.287)	18.16 (4.337)	18.36 (4.365)	18.56 (4.392)	18.56 (4.392)
V <sub>13</sub>	24.10 (5.004)	24.50 (5.044)	24.92 (5.086)	25.44 (5.137)	25.80 (5.173)	26.00 (5.191)	26.00 (5.191)	26.00 (5.191)	26.00 (5.191)	26.00 (5.191)	26.00 (5.191)	26.00 (5.191)
V <sub>14</sub>	24.80 (5.077)	25.24 (5.12)	25.62 (5.157)	26.42 (5.235)	26.94 (5.285)	27.20 (5.309)	27.36 (5.324)	27.36 (5.324)	27.36 (5.324)	27.36 (5.324)	27.36 (5.324)	27.36 (5.324)
V <sub>15</sub>	26.40 (5.234)	26.76 (5.268)	27.12 (5.302)	27.46 (5.335)	27.88 (5.374)	27.92 (5.377)	27.94 (5.379)	27.94 (5.379)	28.08 (5.392)	28.14 (5.398)	28.14 (5.398)	28.14 (5.398)
V <sub>16</sub>	20.50 (4.624)	21.04 (4.682)	21.50 (4.73)	21.92 (4.775)	22.36 (4.821)	22.66 (4.852)	22.66 (4.852)	22.66 (4.852)	22.66 (4.852)	22.66 (4.852)	22.66 (4.852)	22.66 (4.852)

**Appendix XIII. continued**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	15.50 (4.035)	15.96 (4.092)	16.50 (4.159)	17.13 (4.24)	17.72 (4.308)	18.48 (4.397)	18.80 (4.434)	18.94 (4.449)	19.12 (4.469)	19.12 (4.469)	19.12 (4.469)	19.12 (4.469)
V <sub>18</sub>	24.04 (5.002)	24.52 (5.05)	25.22 (5.119)	26.06 (5.201)	26.60 (5.253)	26.82 (5.273)	26.82 (5.273)	26.82 (5.273)	26.82 (5.273)	26.82 (5.273)	26.82 (5.273)	26.82 (5.273)
V <sub>19</sub>	17.10 (4.237)	17.80 (4.32)	18.52 (4.404)	18.94 (4.452)	19.36 (4.498)	19.76 (4.545)	19.90 (4.562)	19.94 (4.567)	20.00 (4.574)	20.06 (4.581)	20.06 (4.581)	20.06 (4.581)
V <sub>20</sub>	18.60 (4.402)	19.10 (4.461)	19.70 (4.531)	20.02 (4.568)	20.38 (4.608)	20.74 (4.647)	20.74 (4.647)	20.74 (4.647)	20.74 (4.647)	20.74 (4.647)	20.74 (4.647)	20.74 (4.647)
V <sub>21</sub>	26.02 (5.194)	26.58 (5.248)	27.18 (5.305)	27.50 (5.336)	27.86 (5.369)	27.86 (5.369)	27.86 (5.369)	27.86 (5.369)	27.86 (5.369)	27.86 (5.369)	27.86 (5.369)	27.86 (5.369)
V <sub>22</sub>	28.50 (5.408)	29.08 (5.463)	29.44 (5.495)	30.12 (5.56)	30.66 (5.609)	30.94 (5.633)	31.14 (5.65)	31.20 (5.655)	31.34 (5.667)	31.34 (5.667)	31.34 (5.667)	31.34 (5.667)
V <sub>23</sub>	29.40 (5.502)	29.92 (5.549)	30.28 (5.581)	30.62 (5.612)	31.04 (5.648)	31.40 (5.681)	31.50 (5.689)	31.72 (5.709)	31.72 (5.709)	31.72 (5.709)	31.72 (5.709)	31.72 (5.709)
V <sub>24</sub>	20.50 (4.626)	20.80 (4.657)	21.20 (4.7)	21.68 (4.751)	22.14 (4.799)	22.52 (4.837)	22.84 (4.871)	22.92 (4.88)	22.96 (4.884)	22.96 (4.884)	22.96 (4.884)	22.96 (4.884)
V <sub>25</sub>	19.50 (4.522)	20.00 (4.576)	20.42 (4.621)	20.78 (4.661)	21.30 (4.717)	21.40 (4.727)	21.60 (4.749)	21.60 (4.749)	21.60 (4.749)	21.60 (4.749)	21.60 (4.749)	21.60 (4.749)
V <sub>26</sub>	19.04 (4.471)	19.50 (4.52)	20.02 (4.578)	20.66 (4.648)	21.28 (4.715)	21.28 (4.715)	21.48 (4.737)	21.48 (4.737)	21.48 (4.737)	21.48 (4.737)	21.48 (4.737)	21.48 (4.737)
V <sub>27</sub>	35.44 (6.036)	35.92 (6.075)	36.26 (6.103)	36.76 (6.144)	37.06 (6.168)	37.70 (6.22)	37.80 (6.228)	37.80 (6.228)	37.80 (6.228)	37.80 (6.228)	37.80 (6.228)	37.80 (6.228)
V <sub>28</sub>	32.80 (5.81)	33.22 (5.846)	33.64 (5.882)	34.20 (5.929)	34.62 (5.965)	34.84 (5.984)	34.94 (5.993)	35.00 (5.998)	35.14 (6.009)	35.14 (6.009)	35.14 (6.009)	35.14 (6.009)
V <sub>29</sub>	27.00 (5.284)	27.50 (5.332)	27.82 (5.362)	28.24 (5.401)	28.88 (5.46)	29.08 (5.478)	29.18 (5.487)	29.40 (5.506)	29.40 (5.506)	29.40 (5.506)	29.40 (5.506)	29.40 (5.506)
V <sub>30</sub>	18.80 (4.416)	19.30 (4.475)	19.76 (4.528)	20.16 (4.573)	20.64 (4.627)	20.68 (4.631)	20.70 (4.633)	20.70 (4.633)	20.84 (4.646)	20.90 (4.651)	20.90 (4.651)	20.90 (4.651)
C.D.	0.469	0.456	0.451	0.444	0.434	0.427	0.421	0.419	0.416	0.412	0.407	0.407
SE(m) <sup>±</sup>	0.167	0.163	0.161	0.158	0.155	0.152	0.15	0.15	0.148	0.147	0.145	0.145
C.V.	7.757	7.462	7.297	7.115	6.894	6.725	6.622	6.579	6.522	6.456	6.375	6.372

Note: The data in parenthesis indicate square root transformed values

**Appendix XIV. Leaf length of *Ascocentrum* hybrids/varieties during the period 2017-18, cm**

Variety	Apr. '17	May. '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	24.50 (5.041)	25.40 (5.132)	25.90 (5.18)	26.26 (5.214)	26.70 (5.257)	26.92 (5.277)	26.98 (5.282)	27.00 (5.284)	27.00 (5.284)	27.00 (5.284)	27.00 (5.284)	27.00 (5.284)
V <sub>2</sub>	23.30 (4.926)	23.02 (4.894)	23.68 (4.961)	24.04 (4.996)	24.80 (5.071)	25.22 (5.112)	25.54 (5.143)	25.68 (5.158)	25.68 (5.158)	25.68 (5.158)	25.68 (5.158)	25.68 (5.158)
V <sub>3</sub>	32.38 (5.768)	32.82 (5.806)	33.22 (5.841)	33.74 (5.886)	34.18 (5.923)	34.34 (5.937)	34.44 (5.945)	34.44 (5.945)	34.44 (5.945)	34.44 (5.945)	34.44 (5.945)	34.44 (5.945)
V <sub>4</sub>	23.50 (4.947)	24.00 (4.997)	24.66 (5.062)	25.08 (5.103)	25.90 (5.182)	26.58 (5.248)	26.96 (5.283)	26.96 (5.283)	26.96 (5.283)	26.96 (5.283)	26.96 (5.283)	26.96 (5.283)
V <sub>5</sub>	25.28 (5.125)	25.76 (5.172)	26.30 (5.224)	26.98 (5.289)	27.58 (5.345)	27.92 (5.376)	27.92 (5.376)	27.92 (5.376)	27.92 (5.376)	27.92 (5.376)	27.92 (5.376)	27.92 (5.376)
V <sub>6</sub>	22.00 (4.786)	22.58 (4.846)	23.16 (4.905)	23.66 (4.955)	23.96 (4.986)	24.44 (5.036)	24.68 (5.061)	24.84 (5.077)	24.84 (5.077)	24.84 (5.077)	24.84 (5.077)	24.84 (5.077)
V <sub>7</sub>	21.96 (4.786)	23.32 (4.927)	24.10 (5.005)	24.54 (5.049)	24.68 (5.064)	24.82 (5.078)	24.82 (5.078)	24.82 (5.078)	24.82 (5.078)	24.82 (5.078)	24.82 (5.078)	24.82 (5.078)
V <sub>8</sub>	23.62 (4.957)	22.98 (4.887)	23.58 (4.949)	23.96 (4.987)	24.64 (5.055)	25.42 (5.133)	25.62 (5.154)	25.90 (5.182)	25.98 (5.19)	25.98 (5.19)	25.98 (5.19)	25.98 (5.19)
V <sub>9</sub>	10.72 (3.376)	11.12 (3.444)	11.70 (3.526)	12.30 (3.611)	12.94 (3.701)	13.52 (3.786)	14.02 (3.854)	14.42 (3.909)	14.62 (3.939)	14.82 (3.967)	15.02 (3.994)	15.02 (3.994)
V <sub>10</sub>	24.48 (5.044)	25.02 (5.098)	24.98 (5.091)	25.18 (5.111)	25.28 (5.122)	25.40 (5.134)	25.48 (5.143)	25.68 (5.163)	25.68 (5.163)	25.68 (5.163)	25.68 (5.163)	25.68 (5.163)
V <sub>11</sub>	12.30 (3.591)	12.74 (3.658)	13.20 (3.72)	13.86 (3.813)	14.32 (3.873)	14.68 (3.928)	14.86 (3.954)	14.96 (3.969)	15.16 (3.999)	15.36 (4.027)	15.56 (4.054)	15.56 (4.054)
V <sub>12</sub>	14.68 (3.917)	15.34 (4.005)	15.62 (4.045)	18.40 (4.345)	19.18 (4.436)	19.48 (4.478)	19.82 (4.523)	20.16 (4.568)	20.22 (4.575)	20.28 (4.582)	20.28 (4.582)	20.28 (4.582)
V <sub>13</sub>	25.28 (5.122)	25.60 (5.154)	26.02 (5.194)	26.58 (5.248)	26.84 (5.273)	27.22 (5.309)	27.22 (5.309)	27.22 (5.309)	27.22 (5.309)	27.22 (5.309)	27.22 (5.309)	27.22 (5.309)
V <sub>14</sub>	23.10 (4.877)	23.60 (4.929)	23.98 (4.969)	24.56 (5.028)	25.30 (5.1)	25.40 (5.113)	25.54 (5.129)	25.58 (5.134)	25.64 (5.141)	25.70 (5.148)	25.70 (5.148)	25.70 (5.148)
V <sub>15</sub>	23.32 (4.923)	23.80 (4.972)	24.36 (5.028)	24.86 (5.079)	25.24 (5.117)	25.66 (5.158)	25.72 (5.165)	25.74 (5.167)	25.90 (5.183)	25.90 (5.183)	25.90 (5.183)	25.90 (5.183)
V <sub>16</sub>	19.34 (4.504)	19.90 (4.564)	20.42 (4.621)	21.10 (4.696)	21.68 (4.757)	21.78 (4.768)	21.98 (4.79)	21.98 (4.79)	21.98 (4.79)	21.98 (4.79)	21.98 (4.79)	21.98 (4.79)

**Appendix XIV. continued**

Variety	Apr. '17	May. '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	14.36 (3.861)	14.80 (3.918)	15.36 (3.989)	15.64 (4.027)	16.06 (4.084)	16.40 (4.128)	16.54 (4.144)	16.58 (4.149)	16.64 (4.156)	16.70 (4.163)	16.70 (4.163)	16.70 (4.163)
V <sub>18</sub>	21.08 (4.638)	21.46 (4.682)	21.94 (4.734)	22.72 (4.825)	23.14 (4.868)	23.48 (4.908)	23.56 (4.919)	23.56 (4.919)	23.56 (4.919)	23.56 (4.919)	23.56 (4.919)	23.56 (4.919)
V <sub>19</sub>	17.54 (4.298)	18.06 (4.358)	18.42 (4.399)	20.04 (4.573)	20.62 (4.635)	20.72 (4.647)	20.86 (4.664)	20.90 (4.668)	20.96 (4.676)	21.02 (4.683)	21.02 (4.683)	21.02 (4.683)
V <sub>20</sub>	21.26 (4.707)	21.68 (4.751)	22.30 (4.814)	22.68 (4.854)	23.06 (4.895)	23.06 (4.895)	23.06 (4.895)	23.06 (4.895)	23.06 (4.895)	23.06 (4.895)	23.06 (4.895)	23.06 (4.895)
V <sub>21</sub>	25.24 (5.121)	25.20 (5.115)	26.02 (5.195)	26.02 (5.191)	26.48 (5.237)	26.68 (5.257)	27.08 (5.297)	27.28 (5.317)	27.28 (5.317)	27.28 (5.317)	27.28 (5.317)	27.28 (5.317)
V <sub>22</sub>	28.56 (5.402)	28.98 (5.442)	29.56 (5.496)	30.22 (5.554)	30.70 (5.599)	30.82 (5.61)	30.88 (5.615)	30.90 (5.617)	30.90 (5.617)	30.90 (5.617)	30.90 (5.617)	30.90 (5.617)
V <sub>23</sub>	28.20 (5.363)	28.70 (5.412)	29.12 (5.45)	30.04 (5.536)	30.70 (5.6)	30.84 (5.615)	30.84 (5.615)	30.84 (5.615)	30.84 (5.615)	30.84 (5.615)	30.84 (5.615)	30.84 (5.615)
V <sub>24</sub>	20.12 (4.582)	20.74 (4.65)	21.26 (4.706)	22.32 (4.817)	22.76 (4.863)	23.16 (4.905)	23.36 (4.927)	23.60 (4.953)	23.64 (4.957)	23.64 (4.957)	23.64 (4.957)	23.64 (4.957)
V <sub>25</sub>	18.90 (4.446)	19.22 (4.482)	19.78 (4.544)	20.34 (4.604)	20.92 (4.667)	21.42 (4.719)	21.42 (4.719)	21.42 (4.719)	21.42 (4.719)	21.42 (4.719)	21.42 (4.719)	21.42 (4.719)
V <sub>26</sub>	18.30 (4.391)	18.72 (4.438)	19.14 (4.486)	19.52 (4.528)	20.04 (4.585)	20.18 (4.6)	20.18 (4.6)	20.18 (4.6)	20.18 (4.6)	20.18 (4.6)	20.18 (4.6)	20.18 (4.6)
V <sub>27</sub>	36.60 (6.131)	37.04 (6.167)	37.92 (6.238)	38.38 (6.275)	38.66 (6.297)	38.74 (6.303)	38.74 (6.303)	38.74 (6.303)	38.74 (6.303)	38.74 (6.303)	38.74 (6.303)	38.74 (6.303)
V <sub>28</sub>	30.58 (5.619)	31.10 (5.665)	31.52 (5.702)	32.02 (5.745)	32.52 (5.789)	32.72 (5.806)	32.88 (5.82)	32.90 (5.822)	32.90 (5.822)	32.90 (5.822)	32.90 (5.822)	32.90 (5.822)
V <sub>29</sub>	25.00 (5.087)	25.68 (5.154)	26.20 (5.206)	26.92 (5.274)	27.36 (5.315)	27.56 (5.333)	27.96 (5.367)	27.96 (5.367)	27.96 (5.367)	27.96 (5.367)	27.96 (5.367)	27.96 (5.367)
V <sub>30</sub>	19.13 (4.445)	19.76 (4.517)	19.73 (4.512)	20.08 (4.55)	20.60 (4.609)	20.84 (4.633)	20.90 (4.638)	20.80 (4.626)	20.80 (4.626)	20.80 (4.626)	20.80 (4.626)	20.80 (4.626)
C.D.	0.565	0.556	0.554	0.564	0.551	0.529	0.516	0.503	0.496	0.492	0.489	0.489
SE(M)	0.202	0.198	0.198	0.201	0.196	0.189	0.184	0.179	0.177	0.175	0.174	0.174
C.V.	9.402	9.159	9.033	9.075	8.765	8.377	8.133	7.92	7.809	7.729	7.688	7.688

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XV. Leaf breadth of *Ascocentrum* hybrids/varieties during the period 2016-17, cm**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	2.46 (1.86)	2.50 (1.871)	2.56 (1.886)	2.64 (1.907)	2.76 (1.939)	2.86 (1.964)	2.96 (1.989)	2.96 (1.989)	2.98 (1.994)	3.00 (2)	3.00 (2)	3.00 (2)
V <sub>2</sub>	2.64 (1.908)	2.76 (1.939)	2.78 (1.944)	2.84 (1.959)	2.92 (1.979)	3.02 (2.004)	3.06 (2.014)	3.06 (2.014)	3.08 (2.019)	3.10 (2.024)	3.10 (2.024)	3.10 (2.024)
V <sub>3</sub>	3.40 (2.097)	3.44 (2.106)	3.46 (2.111)	3.48 (2.115)	3.56 (2.134)	3.68 (2.162)	3.70 (2.167)	3.74 (2.176)	3.74 (2.176)	3.76 (2.181)	3.76 (2.181)	3.76 (2.181)
V <sub>4</sub>	2.54 (1.881)	2.64 (1.906)	2.70 (1.922)	2.78 (1.942)	2.78 (1.943)	2.88 (1.969)	2.90 (1.974)	2.92 (1.98)	2.94 (1.985)	2.94 (1.985)	2.94 (1.985)	2.94 (1.985)
V <sub>5</sub>	2.72 (1.928)	2.82 (1.953)	2.92 (1.979)	2.98 (1.994)	3.04 (2.009)	3.08 (2.019)	3.10 (2.024)	3.10 (2.024)	3.10 (2.024)	3.10 (2.024)	3.10 (2.024)	3.10 (2.024)
V <sub>6</sub>	1.70 (1.643)	1.76 (1.661)	1.88 (1.697)	1.94 (1.714)	1.94 (1.714)	2.04 (1.743)	2.06 (1.749)	2.16 (1.777)	2.22 (1.794)	2.28 (1.811)	2.34 (1.827)	2.38 (1.838)
V <sub>7</sub>	2.65 (1.911)	2.81 (1.952)	2.91 (1.977)	2.99 (1.996)	2.98 (1.994)	3.03 (2.006)	3.05 (2.011)	3.08 (2.019)	3.08 (2.019)	3.08 (2.019)	3.08 (2.019)	3.08 (2.019)
V <sub>8</sub>	2.32 (1.822)	2.42 (1.849)	2.48 (1.865)	2.52 (1.875)	2.58 (1.891)	2.64 (1.907)	2.67 (1.915)	2.70 (1.922)	2.72 (1.927)	2.74 (1.932)	2.74 (1.932)	2.74 (1.932)
V <sub>9</sub>	0.52 (1.232)	0.56 (1.248)	0.62 (1.272)	0.66 (1.287)	0.74 (1.319)	0.78 (1.333)	0.82 (1.349)	0.82 (1.349)	0.82 (1.349)	0.82 (1.349)	0.82 (1.349)	0.82 (1.349)
V <sub>10</sub>	2.30 (1.816)	2.40 (1.843)	2.46 (1.86)	2.50 (1.87)	2.54 (1.88)	2.62 (1.902)	2.62 (1.902)	2.62 (1.902)	2.62 (1.902)	2.62 (1.902)	2.62 (1.902)	2.62 (1.902)
V <sub>11</sub>	1.44 (1.561)	1.54 (1.593)	1.76 (1.66)	1.84 (1.684)	1.96 (1.719)	2.02 (1.737)	2.10 (1.76)	2.14 (1.771)	2.16 (1.777)	2.16 (1.777)	2.16 (1.777)	2.16 (1.777)
V <sub>12</sub>	2.26 (1.804)	2.42 (1.848)	2.52 (1.875)	2.60 (1.896)	2.54 (1.881)	2.60 (1.897)	2.60 (1.897)	2.62 (1.902)	2.64 (1.908)	2.66 (1.913)	2.66 (1.913)	2.66 (1.913)
V <sub>13</sub>	2.36 (1.832)	2.46 (1.859)	2.54 (1.88)	2.58 (1.891)	2.66 (1.912)	2.72 (1.927)	2.72 (1.927)	2.76 (1.938)	2.76 (1.938)	2.76 (1.938)	2.76 (1.938)	2.76 (1.938)
V <sub>14</sub>	2.22 (1.794)	2.34 (1.827)	2.48 (1.865)	2.58 (1.892)	2.68 (1.918)	2.74 (1.934)	2.74 (1.934)	2.74 (1.934)	2.74 (1.934)	2.74 (1.934)	2.74 (1.934)	2.74 (1.934)
V <sub>15</sub>	2.00 (1.732)	2.12 (1.766)	2.16 (1.777)	2.24 (1.8)	2.20 (1.789)	2.30 (1.816)	2.34 (1.827)	2.44 (1.854)	2.46 (1.859)	2.46 (1.858)	2.52 (1.875)	2.56 (1.886)
V <sub>16</sub>	2.34 (1.826)	2.54 (1.881)	2.60 (1.897)	2.68 (1.918)	2.78 (1.944)	2.90 (1.974)	2.92 (1.979)	2.92 (1.979)	2.92 (1.979)	2.94 (1.985)	2.94 (1.985)	2.96 (1.99)

**APPENDIX XV. continued**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	2.12 (1.766)	2.26 (1.805)	2.32 (1.821)	2.34 (1.827)	2.36 (1.832)	2.42 (1.848)	2.42 (1.848)	2.44 (1.854)	2.44 (1.854)	2.46 (1.859)	2.46 (1.859)	2.40 (1.842)
V <sub>18</sub>	2.52 (1.876)	2.62 (1.902)	2.64 (1.908)	2.70 (1.923)	2.78 (1.944)	2.87 (1.968)	2.92 (1.98)	2.94 (1.985)	2.98 (1.995)	3.00 (2)	3.00 (2)	3.00 (2)
V <sub>19</sub>	2.98 (1.994)	3.06 (2.014)	3.14 (2.034)	3.18 (2.044)	3.26 (2.064)	3.38 (2.093)	3.44 (2.107)	3.42 (2.102)	3.42 (2.102)	3.42 (2.102)	3.42 (2.102)	3.42 (2.102)
V <sub>20</sub>	2.78 (1.944)	2.88 (1.97)	2.98 (1.995)	3.04 (2.01)	3.06 (2.015)	3.18 (2.044)	3.26 (2.064)	3.30 (2.074)	3.30 (2.074)	3.30 (2.074)	3.30 (2.074)	3.30 (2.074)
V <sub>21</sub>	2.52 (1.876)	2.62 (1.902)	2.68 (1.917)	2.70 (1.922)	2.74 (1.933)	2.86 (1.964)	2.90 (1.974)	2.90 (1.974)	2.92 (1.979)	2.94 (1.984)	2.94 (1.984)	2.94 (1.984)
V <sub>22</sub>	2.48 (1.865)	2.66 (1.913)	2.78 (1.944)	2.86 (1.964)	2.94 (1.984)	3.06 (2.014)	3.16 (2.039)	3.16 (2.039)	3.20 (2.049)	3.22 (2.054)	3.24 (2.059)	3.26 (2.063)
V <sub>23</sub>	3.68 (2.163)	3.72 (2.172)	3.82 (2.195)	3.86 (2.204)	3.90 (2.213)	4.00 (2.235)	4.00 (2.235)	4.02 (2.24)	4.02 (2.24)	4.04 (2.244)	4.04 (2.244)	4.04 (2.244)
V <sub>24</sub>	3.54 (2.129)	3.64 (2.153)	3.68 (2.162)	3.70 (2.167)	3.78 (2.185)	3.88 (2.208)	3.88 (2.208)	3.90 (2.213)	3.90 (2.213)	3.92 (2.217)	3.92 (2.217)	3.92 (2.217)
V <sub>25</sub>	3.24 (2.057)	3.34 (2.082)	3.38 (2.092)	3.40 (2.096)	3.46 (2.11)	3.56 (2.134)	3.60 (2.144)	3.64 (2.153)	3.66 (2.158)	3.66 (2.158)	3.66 (2.158)	3.66 (2.158)
V <sub>26</sub>	3.24 (2.059)	3.29 (2.07)	3.33 (2.081)	3.36 (2.088)	3.44 (2.107)	3.54 (2.131)	3.58 (2.14)	3.60 (2.145)	3.60 (2.145)	3.60 (2.145)	3.60 (2.145)	3.60 (2.145)
V <sub>27</sub>	3.66 (2.158)	3.66 (2.158)	3.74 (2.176)	3.76 (2.18)	3.82 (2.194)	3.94 (2.221)	3.96 (2.226)	3.98 (2.231)	3.98 (2.231)	3.98 (2.231)	3.98 (2.231)	3.98 (2.231)
V <sub>28</sub>	2.69 (1.92)	2.81 (1.951)	2.88 (1.968)	2.98 (1.993)	3.06 (2.014)	3.16 (2.039)	3.22 (2.053)	3.18 (2.044)	3.20 (2.049)	3.20 (2.049)	3.20 (2.049)	3.20 (2.049)
V <sub>29</sub>	2.65 (1.91)	2.76 (1.939)	2.82 (1.955)	2.91 (1.977)	2.99 (1.996)	3.11 (2.026)	3.18 (2.044)	3.20 (2.048)	3.22 (2.053)	3.24 (2.058)	3.24 (2.058)	3.24 (2.058)
V <sub>30</sub>	2.48 (1.865)	2.70 (1.923)	2.80 (1.949)	2.92 (1.98)	2.98 (1.995)	3.09 (2.023)	3.16 (2.039)	3.22 (2.054)	3.24 (2.059)	3.24 (2.059)	3.24 (2.059)	3.24 (2.059)
C.D.	0.071	0.072	0.074	0.078	0.079	0.076	0.076	0.073	0.072	0.073	0.071	0.072
SE(M)	0.025	0.026	0.026	0.028	0.028	0.027	0.027	0.026	0.026	0.026	0.025	0.026
C.V.	3.028	3.003	3.069	3.191	3.242	3.076	3.038	2.929	2.897	2.9	2.853	2.867

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XVI. Leaf breadth of *Ascocentrum* hybrids/varieties during the period 2017-18, cm**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	2.54 (1.881)	2.65 (1.91)	2.77 (1.942)	2.91 (1.976)	2.95 (1.986)	3.04 (2.009)	3.10 (2.024)	3.10 (2.024)	3.10 (2.024)	3.10 (2.024)	3.10 (2.024)	3.10 (2.024)
V <sub>2</sub>	2.62 (1.902)	2.75 (1.936)	2.83 (1.957)	2.95 (1.986)	3.03 (2.006)	3.10 (2.024)	3.14 (2.034)	3.14 (2.034)	3.14 (2.034)	3.14 (2.034)	3.14 (2.034)	3.14 (2.034)
V <sub>3</sub>	3.64 (2.154)	3.68 (2.163)	3.80 (2.19)	3.80 (2.19)	3.84 (2.199)	3.90 (2.213)	3.90 (2.213)	3.90 (2.213)	3.90 (2.213)	3.90 (2.213)	3.90 (2.213)	3.90 (2.213)
V <sub>4</sub>	2.61 (1.9)	2.75 (1.936)	2.85 (1.962)	2.91 (1.976)	3.00 (1.999)	3.07 (2.016)	3.10 (2.025)	3.12 (2.029)	3.14 (2.034)	3.16 (2.039)	3.16 (2.039)	3.16 (2.039)
V <sub>5</sub>	2.76 (1.936)	2.84 (1.957)	2.90 (1.972)	2.92 (1.977)	2.90 (1.972)	3.04 (2.008)	3.08 (2.018)	3.10 (2.022)	3.10 (2.022)	3.10 (2.022)	3.10 (2.022)	3.25 (1.848)
V <sub>6</sub>	1.78 (1.661)	1.88 (1.691)	2.00 (1.727)	2.06 (1.746)	2.18 (1.78)	2.24 (1.797)	2.30 (1.814)	2.32 (1.82)	2.32 (1.82)	2.32 (1.82)	2.32 (1.82)	2.32 (1.82)
V <sub>7</sub>	2.54 (1.881)	2.64 (1.906)	2.72 (1.927)	2.82 (1.952)	2.74 (1.932)	2.82 (1.954)	2.84 (1.959)	2.88 (1.969)	2.90 (1.975)	2.90 (1.975)	2.90 (1.975)	2.90 (1.975)
V <sub>8</sub>	2.28 (1.81)	2.42 (1.849)	2.50 (1.87)	2.56 (1.886)	2.62 (1.902)	2.70 (1.922)	2.72 (1.927)	2.76 (1.938)	2.76 (1.938)	2.76 (1.938)	2.78 (1.943)	2.65 (1.728)
V <sub>9</sub>	0.64 (1.277)	0.70 (1.3)	0.76 (1.323)	0.83 (1.347)	0.92 (1.38)	0.96 (1.394)	1.02 (1.416)	1.04 (1.424)	1.04 (1.424)	1.04 (1.424)	1.04 (1.424)	1.04 (1.424)
V <sub>10</sub>	2.22 (1.793)	2.30 (1.815)	2.38 (1.837)	2.50 (1.869)	2.56 (1.885)	2.68 (1.917)	2.70 (1.922)	2.72 (1.928)	2.74 (1.933)	2.74 (1.933)	2.74 (1.933)	2.74 (1.933)
V <sub>11</sub>	1.46 (1.568)	1.60 (1.611)	1.86 (1.69)	1.96 (1.719)	2.06 (1.748)	2.12 (1.766)	2.18 (1.782)	2.20 (1.788)	2.20 (1.788)	2.20 (1.788)	2.20 (1.788)	2.20 (1.788)
V <sub>12</sub>	2.38 (1.837)	2.50 (1.87)	2.58 (1.891)	2.62 (1.902)	2.70 (1.922)	2.76 (1.938)	2.76 (1.938)	2.76 (1.938)	2.78 (1.943)	2.80 (1.949)	2.80 (1.949)	2.80 (1.949)
V <sub>13</sub>	2.30 (1.815)	2.48 (1.864)	2.58 (1.891)	2.66 (1.912)	2.62 (1.902)	2.66 (1.912)	2.66 (1.912)	2.68 (1.918)	2.70 (1.923)	2.72 (1.928)	2.72 (1.928)	2.72 (1.928)
V <sub>14</sub>	2.16 (1.777)	2.30 (1.816)	2.38 (1.838)	2.42 (1.849)	2.44 (1.855)	2.56 (1.886)	2.56 (1.886)	2.60 (1.897)	2.62 (1.902)	2.64 (1.908)	2.64 (1.908)	2.64 (1.908)
V <sub>15</sub>	1.86 (1.689)	1.98 (1.724)	2.18 (1.782)	2.24 (1.799)	2.28 (1.81)	2.34 (1.827)	2.38 (1.838)	2.44 (1.853)	2.48 (1.864)	2.50 (1.869)	2.50 (1.869)	2.52 (1.874)
V <sub>16</sub>	2.38 (1.837)	2.52 (1.875)	2.60 (1.896)	2.68 (1.917)	2.64 (1.908)	2.68 (1.918)	2.68 (1.918)	2.72 (1.928)	2.72 (1.928)	2.74 (1.934)	2.74 (1.934)	2.76 (1.939)



**APPENDIX XVI. continued**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	2.32 (1.821)	2.16 (1.777)	2.18 (1.783)	2.24 (1.8)	2.34 (1.827)	2.32 (1.822)	2.38 (1.838)	2.42 (1.848)	2.50 (1.87)	2.50 (1.869)	2.48 (1.864)	2.54 (1.881)
V <sub>18</sub>	2.44 (1.854)	2.58 (1.892)	2.66 (1.913)	2.72 (1.928)	2.82 (1.954)	2.88 (1.969)	2.88 (1.969)	2.88 (1.969)	2.88 (1.969)	2.88 (1.969)	2.88 (1.969)	2.88 (1.969)
V <sub>19</sub>	2.76 (1.937)	2.88 (1.967)	2.96 (1.987)	3.00 (1.998)	3.10 (2.022)	3.12 (2.028)	3.12 (2.028)	3.12 (2.028)	3.12 (2.028)	3.12 (2.028)	3.12 (2.028)	3.12 (2.028)
V <sub>20</sub>	2.70 (1.923)	2.84 (1.959)	2.94 (1.984)	3.00 (1.999)	3.08 (2.019)	3.20 (2.049)	3.26 (2.063)	3.22 (2.054)	3.22 (2.054)	3.22 (2.054)	3.22 (2.054)	3.22 (2.054)
V <sub>21</sub>	2.48 (1.865)	2.58 (1.892)	2.60 (1.897)	2.66 (1.912)	2.74 (1.933)	2.81 (1.952)	2.84 (1.959)	2.86 (1.964)	2.90 (1.974)	2.92 (1.979)	2.92 (1.979)	2.92 (1.979)
V <sub>22</sub>	2.86 (1.962)	2.96 (1.987)	3.04 (2.007)	3.10 (2.022)	3.14 (2.032)	3.21 (2.05)	3.28 (2.067)	3.34 (2.081)	3.36 (2.086)	3.36 (2.086)	3.36 (2.086)	3.36 (2.086)
V <sub>23</sub>	3.64 (2.154)	3.72 (2.172)	3.84 (2.2)	3.86 (2.204)	3.92 (2.218)	3.94 (2.222)	3.94 (2.222)	3.94 (2.222)	3.94 (2.222)	3.94 (2.222)	3.94 (2.222)	3.94 (2.222)
V <sub>24</sub>	3.66 (2.158)	3.68 (2.163)	3.76 (2.181)	3.76 (2.181)	3.80 (2.191)	3.88 (2.208)	3.88 (2.208)	3.88 (2.208)	3.88 (2.208)	3.88 (2.208)	3.88 (2.208)	3.88 (2.208)
V <sub>25</sub>	2.98 (1.994)	3.10 (2.024)	3.20 (2.049)	3.26 (2.064)	3.38 (2.093)	3.38 (2.092)	3.44 (2.107)	3.46 (2.111)	3.46 (2.111)	3.48 (2.116)	3.48 (2.116)	3.48 (2.116)
V <sub>26</sub>	3.28 (2.066)	3.30 (2.071)	3.36 (2.086)	3.44 (2.105)	3.48 (2.114)	3.54 (2.129)	3.56 (2.134)	3.58 (2.138)	3.60 (2.143)	3.62 (2.148)	3.62 (2.148)	3.62 (2.148)
V <sub>27</sub>	3.28 (2.064)	3.36 (2.085)	3.48 (2.113)	3.52 (2.123)	3.58 (2.138)	3.72 (2.17)	3.76 (2.18)	3.78 (2.184)	3.78 (2.184)	3.78 (2.184)	3.80 (2.189)	3.80 (2.189)
V <sub>28</sub>	2.54 (1.88)	2.78 (1.943)	2.90 (1.974)	3.00 (2)	3.08 (2.019)	3.18 (2.044)	3.30 (2.073)	3.30 (2.073)	3.32 (2.078)	3.32 (2.078)	3.34 (2.083)	3.38 (2.092)
V <sub>29</sub>	2.60 (1.897)	2.70 (1.923)	2.72 (1.928)	2.80 (1.948)	2.90 (1.974)	2.99 (1.998)	3.06 (2.014)	3.08 (2.019)	3.12 (2.029)	3.14 (2.034)	3.14 (2.034)	3.14 (2.034)
V <sub>30</sub>	2.52 (1.875)	2.75 (1.937)	2.87 (1.967)	2.97 (1.991)	3.08 (2.02)	3.22 (2.054)	3.30 (2.073)	3.32 (2.078)	3.32 (2.078)	3.32 (2.078)	3.34 (2.083)	3.36 (2.088)
C.D.	0.102	0.101	0.098	0.1	0.099	0.097	0.093	0.093	0.092	0.092	0.093	0.168
SE(M)	0.037	0.036	0.035	0.036	0.035	0.034	0.033	0.033	0.033	0.033	0.033	0.06
C.V.	4.364	4.223	4.08	4.125	4.042	3.898	3.739	3.732	3.664	3.693	3.706	6.758

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XVII. Leaf area of *Ascocentrum* hybrids/varieties during the period 2016-17, cm<sup>2</sup>**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	50.47 (7.171)	51.18 (7.22)	51.80 (7.262)	52.45 (7.307)	52.90 (7.337)	53.45 (7.374)	53.74 (7.394)	53.82 (7.398)	54.01 (7.41)	54.01 (7.411)	54.01 (7.411)	54.01 (7.411)
V <sub>2</sub>	48.64 (7.043)	49.87 (7.13)	50.55 (7.177)	51.14 (7.218)	52.04 (7.281)	52.39 (7.304)	52.53 (7.314)	52.53 (7.314)	52.54 (7.314)	52.54 (7.315)	52.54 (7.315)	52.54 (7.315)
V <sub>3</sub>	63.91 (8.056)	64.58 (8.097)	65.15 (8.132)	65.94 (8.18)	66.38 (8.207)	66.86 (8.236)	66.86 (8.237)	66.87 (8.238)	66.87 (8.238)	66.88 (8.238)	66.88 (8.238)	66.88 (8.238)
V <sub>4</sub>	47.96 (6.994)	48.61 (7.04)	49.23 (7.084)	49.95 (7.135)	50.70 (7.187)	51.02 (7.21)	51.36 (7.234)	51.40 (7.236)	51.61 (7.252)	51.61 (7.252)	51.61 (7.252)	51.61 (7.252)
V <sub>5</sub>	49.96 (7.127)	50.64 (7.174)	51.32 (7.221)	49.45 (7.083)	50.04 (7.126)	50.49 (7.159)	50.52 (7.162)	50.52 (7.162)	50.52 (7.162)	50.52 (7.162)	50.52 (7.162)	50.52 (7.162)
V <sub>6</sub>	47.30 (6.94)	48.28 (7.01)	48.89 (7.053)	49.59 (7.103)	49.97 (7.13)	50.54 (7.171)	50.55 (7.172)	50.58 (7.174)	50.60 (7.175)	50.62 (7.177)	50.64 (7.178)	50.65 (7.179)
V <sub>7</sub>	45.53 (6.82)	46.39 (6.883)	46.91 (6.921)	47.66 (6.975)	48.33 (7.023)	48.76 (7.053)	48.95 (7.067)	48.96 (7.068)	48.96 (7.068)	48.96 (7.068)	48.96 (7.068)	48.96 (7.068)
V <sub>8</sub>	48.40 (7.027)	49.21 (7.084)	50.04 (7.142)	50.49 (7.174)	51.47 (7.242)	52.37 (7.304)	52.80 (7.333)	52.98 (7.346)	53.10 (7.354)	53.10 (7.354)	53.10 (7.354)	53.10 (7.354)
V <sub>9</sub>	28.52 (5.429)	29.26 (5.497)	29.90 (5.555)	30.71 (5.628)	31.52 (5.699)	32.38 (5.776)	33.05 (5.833)	33.56 (5.877)	33.82 (5.9)	34.08 (5.922)	34.34 (5.945)	34.34 (5.945)
V <sub>10</sub>	47.34 (6.949)	48.20 (7.011)	49.07 (7.073)	50.04 (7.142)	50.24 (7.156)	50.47 (7.172)	50.68 (7.187)	50.68 (7.187)	50.68 (7.187)	50.68 (7.187)	49.18 (7.083)	50.03 (7.144)
V <sub>11</sub>	31.79 (5.717)	32.58 (5.786)	33.35 (5.852)	34.25 (5.93)	35.15 (6.006)	36.05 (6.081)	36.44 (6.114)	36.84 (6.147)	36.85 (6.148)	36.85 (6.148)	36.85 (6.148)	36.85 (6.148)
V <sub>12</sub>	36.33 (6.082)	36.77 (6.121)	37.48 (6.178)	38.13 (6.231)	38.96 (6.297)	39.89 (6.372)	40.69 (6.435)	41.09 (6.466)	41.59 (6.506)	41.85 (6.529)	42.11 (6.551)	42.11 (6.551)
V <sub>13</sub>	49.20 (7.081)	49.75 (7.12)	50.32 (7.16)	51.00 (7.208)	51.49 (7.243)	51.77 (7.261)	51.77 (7.261)	51.79 (7.262)	51.79 (7.262)	51.79 (7.262)	51.79 (7.262)	51.79 (7.262)
V <sub>14</sub>	50.06 (7.144)	50.67 (7.186)	51.20 (7.224)	52.27 (7.298)	52.98 (7.346)	53.33 (7.371)	53.54 (7.385)	53.54 (7.385)	53.54 (7.385)	53.54 (7.385)	53.54 (7.385)	53.54 (7.385)
V <sub>15</sub>	52.06 (7.284)	52.57 (7.319)	53.04 (7.351)	53.51 (7.383)	54.04 (7.419)	54.13 (7.425)	54.17 (7.427)	54.20 (7.429)	54.39 (7.442)	54.46 (7.447)	54.48 (7.448)	54.50 (7.449)
V <sub>16</sub>	44.52 (6.74)	45.29 (6.796)	45.90 (6.841)	46.47 (6.883)	47.08 (6.927)	47.50 (6.958)	47.51 (6.958)	47.51 (6.958)	47.51 (6.958)	47.52 (6.958)	47.52 (6.958)	47.52 (6.959)

**APPENDIX XVII. continued**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	37.97 (6.231)	38.61 (6.283)	39.33 (6.34)	40.16 (6.407)	40.93 (6.467)	41.93 (6.544)	42.35 (6.576)	42.53 (6.59)	42.77 (6.608)	42.77 (6.608)	42.77 (6.608)	42.75 (6.607)
V <sub>18</sub>	49.17 (7.082)	49.82 (7.128)	50.74 (7.192)	51.84 (7.269)	52.57 (7.319)	52.89 (7.34)	52.90 (7.341)	52.91 (7.341)	52.92 (7.342)	52.93 (7.343)	52.93 (7.343)	52.93 (7.343)
V <sub>19</sub>	40.32 (6.42)	41.26 (6.493)	42.21 (6.567)	42.77 (6.609)	43.34 (6.652)	43.90 (6.695)	44.10 (6.711)	44.14 (6.714)	44.22 (6.72)	44.30 (6.726)	44.30 (6.726)	44.30 (6.726)
V <sub>20</sub>	42.20 (6.56)	42.88 (6.613)	43.69 (6.675)	44.13 (6.709)	44.60 (6.744)	45.10 (6.781)	45.13 (6.783)	45.14 (6.784)	45.14 (6.784)	45.14 (6.784)	45.14 (6.784)	45.14 (6.784)
V <sub>21</sub>	51.73 (7.259)	52.49 (7.312)	53.29 (7.366)	53.71 (7.395)	54.19 (7.427)	54.23 (7.43)	54.24 (7.43)	54.24 (7.43)	54.25 (7.431)	54.25 (7.431)	54.25 (7.431)	54.25 (7.431)
V <sub>22</sub>	54.94 (7.464)	55.75 (7.519)	56.25 (7.552)	57.16 (7.614)	57.88 (7.662)	58.28 (7.688)	58.58 (7.706)	58.65 (7.711)	58.85 (7.723)	58.85 (7.723)	58.86 (7.724)	58.87 (7.724)
V <sub>23</sub>	56.49 (7.574)	57.17 (7.619)	57.67 (7.652)	58.13 (7.682)	58.68 (7.717)	59.18 (7.75)	59.31 (7.758)	59.60 (7.777)	59.60 (7.777)	59.61 (7.778)	59.61 (7.778)	59.61 (7.778)
V <sub>24</sub>	44.91 (6.77)	45.33 (6.8)	45.86 (6.839)	46.49 (6.885)	47.11 (6.93)	47.64 (6.967)	48.05 (6.997)	48.16 (7.005)	48.21 (7.009)	48.22 (7.009)	48.22 (7.009)	48.22 (7.009)
V <sub>25</sub>	43.52 (6.67)	44.20 (6.719)	44.75 (6.761)	45.23 (6.796)	45.92 (6.847)	46.08 (6.859)	46.35 (6.879)	46.37 (6.88)	46.37 (6.88)	46.37 (6.88)	46.37 (6.88)	46.37 (6.88)
V <sub>26</sub>	42.92 (6.624)	43.53 (6.669)	44.22 (6.721)	45.06 (6.784)	45.89 (6.844)	45.92 (6.847)	46.19 (6.867)	46.20 (6.868)	46.20 (6.868)	46.20 (6.868)	46.20 (6.868)	46.20 (6.868)
V <sub>27</sub>	64.31 (8.081)	64.93 (8.119)	65.40 (8.148)	66.05 (8.188)	66.46 (8.213)	67.33 (8.265)	67.46 (8.274)	67.47 (8.274)	67.47 (8.274)	67.47 (8.274)	67.47 (8.274)	67.47 (8.274)
V <sub>28</sub>	60.58 (7.844)	61.16 (7.881)	61.73 (7.917)	62.48 (7.965)	63.05 (8.001)	63.37 (8.021)	63.52 (8.031)	63.58 (8.035)	63.77 (8.047)	63.77 (8.047)	63.77 (8.047)	63.77 (8.047)
V <sub>29</sub>	53.05 (7.347)	53.73 (7.394)	54.17 (7.423)	54.74 (7.462)	55.59 (7.519)	55.89 (7.538)	56.04 (7.548)	56.33 (7.567)	56.34 (7.567)	56.35 (7.568)	56.35 (7.568)	56.35 (7.568)
V <sub>30</sub>	42.37 (6.568)	43.08 (6.624)	43.71 (6.672)	44.27 (6.714)	44.91 (6.762)	45.00 (6.769)	45.05 (6.772)	45.07 (6.773)	45.25 (6.786)	45.33 (6.791)	45.33 (6.791)	45.33 (6.791)
C.D.	0.408	0.401	0.398	0.394	0.387	0.383	0.38	0.379	0.377	0.374	0.37	0.37
SE(M)	0.146	0.143	0.142	0.141	0.138	0.137	0.135	0.135	0.135	0.134	0.132	0.132
C.V.	4.694	4.574	4.51	4.439	4.335	4.269	4.222	4.206	4.186	4.155	4.106	4.105

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XVIII. Leaf area (cm<sup>2</sup>) of *Ascoctrinum* hybrids / varieties during the period 2017-18, cm<sup>2</sup>**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	49.77 (7.12)	50.97 (7.205)	51.66 (7.253)	52.17 (7.288)	52.75 (7.328)	53.07 (7.349)	53.17 (7.355)	53.19 (7.357)	53.19 (7.357)	53.19 (7.357)	53.19 (7.357)	53.19 (7.357)
V <sub>2</sub>	48.24 (7.015)	47.92 (6.99)	48.80 (7.053)	49.31 (7.088)	50.32 (7.159)	50.88 (7.198)	51.31 (7.227)	51.49 (7.24)	51.49 (7.24)	51.49 (7.24)	51.49 (7.24)	51.49 (7.24)
V <sub>3</sub>	60.34 (7.826)	60.92 (7.863)	61.48 (7.898)	62.15 (7.941)	62.73 (7.978)	62.96 (7.992)	63.09 (8.001)	63.09 (8.001)	63.09 (8.001)	63.09 (8.001)	63.09 (8.001)	63.09 (8.001)
V <sub>4</sub>	48.50 (7.034)	49.19 (7.083)	50.08 (7.145)	50.64 (7.184)	51.73 (7.259)	52.64 (7.321)	53.14 (7.355)	53.15 (7.355)	53.15 (7.356)	53.16 (7.356)	53.16 (7.356)	53.16 (7.356)
V <sub>5</sub>	50.85 (7.2)	51.50 (7.245)	52.22 (7.295)	53.11 (7.355)	53.88 (7.407)	54.36 (7.439)	54.38 (7.44)	54.38 (7.441)	54.38 (7.441)	54.38 (7.441)	54.38 (7.441)	54.22 (7.43)
V <sub>6</sub>	46.29 (6.871)	47.07 (6.928)	47.86 (6.984)	48.53 (7.032)	48.96 (7.062)	49.60 (7.108)	49.93 (7.133)	50.14 (7.148)	50.14 (7.148)	50.14 (7.148)	50.14 (7.148)	50.14 (7.148)
V <sub>7</sub>	46.48 (6.887)	48.27 (7.017)	49.31 (7.09)	49.91 (7.133)	50.07 (7.144)	50.28 (7.159)	50.28 (7.159)	50.30 (7.16)	50.30 (7.161)	50.30 (7.161)	50.30 (7.161)	50.30 (7.161)
V <sub>8</sub>	48.55 (7.036)	47.76 (6.977)	48.57 (7.035)	49.08 (7.072)	49.98 (7.135)	51.02 (7.208)	51.28 (7.227)	51.66 (7.254)	51.76 (7.261)	51.76 (7.261)	51.77 (7.262)	51.55 (7.246)
V <sub>9</sub>	31.30 (5.667)	31.84 (5.717)	32.61 (5.784)	33.41 (5.852)	34.27 (5.926)	35.03 (5.993)	35.70 (6.049)	36.23 (6.094)	36.49 (6.117)	36.74 (6.139)	37.00 (6.161)	37.00 (6.161)
V <sub>10</sub>	49.64 (7.114)	50.37 (7.165)	50.34 (7.161)	50.64 (7.183)	50.79 (7.193)	50.98 (7.207)	51.09 (7.215)	51.36 (7.234)	51.36 (7.235)	51.36 (7.235)	51.36 (7.235)	51.36 (7.235)
V <sub>11</sub>	33.61 (5.862)	34.23 (5.916)	34.91 (5.973)	35.80 (6.049)	36.42 (6.1)	36.91 (6.143)	37.16 (6.165)	37.30 (6.177)	37.56 (6.2)	37.82 (6.222)	38.08 (6.244)	38.08 (6.244)
V <sub>12</sub>	36.99 (6.147)	37.89 (6.22)	38.28 (6.253)	41.89 (6.52)	42.93 (6.6)	43.34 (6.634)	43.78 (6.671)	44.22 (6.707)	44.30 (6.714)	44.39 (6.721)	44.39 (6.721)	44.39 (6.721)
V <sub>13</sub>	50.71 (7.188)	51.18 (7.221)	51.75 (7.261)	52.51 (7.313)	52.83 (7.335)	53.33 (7.369)	53.33 (7.369)	53.34 (7.369)	53.35 (7.37)	53.35 (7.37)	53.35 (7.37)	53.35 (7.37)
V <sub>14</sub>	47.83 (6.97)	48.53 (7.02)	49.05 (7.058)	49.81 (7.113)	50.78 (7.18)	50.94 (7.193)	51.13 (7.207)	51.19 (7.212)	51.27 (7.218)	51.36 (7.225)	51.36 (7.225)	51.36 (7.225)
V <sub>15</sub>	48.02 (6.997)	48.68 (7.044)	49.47 (7.1)	50.14 (7.147)	50.65 (7.183)	51.21 (7.223)	51.30 (7.229)	51.35 (7.232)	51.57 (7.248)	51.57 (7.249)	51.57 (7.249)	51.58 (7.249)
V <sub>16</sub>	43.03 (6.633)	43.80 (6.69)	44.50 (6.742)	45.41 (6.81)	46.15 (6.864)	46.29 (6.874)	46.55 (6.894)	46.56 (6.895)	46.56 (6.895)	46.57 (6.895)	46.57 (6.895)	46.58 (6.896)

**APPENDIX XVIII.continued**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	36.56 (6.101)	37.08 (6.144)	37.81 (6.203)	38.19 (6.235)	38.77 (6.283)	39.20 (6.319)	39.40 (6.334)	39.47 (6.34)	39.57 (6.348)	39.65 (6.354)	39.64 (6.354)	39.66 (6.355)
V <sub>18</sub>	45.31 (6.775)	45.84 (6.816)	46.49 (6.864)	47.52 (6.942)	48.10 (6.983)	48.56 (7.018)	48.66 (7.027)	48.66 (7.027)	48.66 (7.027)	48.66 (7.027)	48.66 (7.027)	48.66 (7.027)
V <sub>19</sub>	40.82 (6.463)	41.53 (6.518)	42.03 (6.556)	44.14 (6.711)	44.92 (6.769)	45.06 (6.78)	45.24 (6.794)	45.29 (6.798)	45.37 (6.804)	45.45 (6.81)	45.45 (6.81)	45.45 (6.81)
V <sub>20</sub>	45.62 (6.822)	46.21 (6.865)	47.05 (6.924)	47.56 (6.962)	48.08 (7)	48.12 (7.003)	48.14 (7.004)	48.12 (7.003)	48.12 (7.003)	48.12 (7.003)	48.12 (7.003)	48.12 (7.003)
V <sub>21</sub>	50.71 (7.19)	50.69 (7.188)	51.76 (7.262)	51.78 (7.261)	52.40 (7.304)	52.68 (7.325)	53.21 (7.362)	53.48 (7.38)	53.49 (7.381)	53.50 (7.382)	53.50 (7.382)	53.50 (7.382)
V <sub>22</sub>	55.14 (7.47)	55.71 (7.509)	56.49 (7.562)	57.36 (7.618)	58.00 (7.661)	58.18 (7.672)	58.28 (7.679)	58.32 (7.682)	58.33 (7.682)	58.33 (7.682)	58.33 (7.682)	58.33 (7.682)
V <sub>23</sub>	54.92 (7.453)	55.59 (7.499)	56.18 (7.537)	57.38 (7.618)	58.25 (7.678)	58.44 (7.691)	58.44 (7.691)	58.44 (7.691)	58.44 (7.691)	58.44 (7.691)	58.44 (7.691)	58.44 (7.691)
V <sub>24</sub>	44.46 (6.735)	45.27 (6.795)	45.96 (6.847)	47.34 (6.946)	47.92 (6.988)	48.47 (7.027)	48.72 (7.046)	49.04 (7.07)	49.09 (7.073)	49.09 (7.073)	49.09 (7.073)	49.09 (7.073)
V <sub>25</sub>	42.66 (6.599)	43.11 (6.634)	43.87 (6.69)	44.61 (6.745)	45.40 (6.804)	46.05 (6.851)	46.07 (6.852)	46.08 (6.852)	46.08 (6.852)	46.08 (6.853)	46.08 (6.853)	46.08 (6.853)
V <sub>26</sub>	41.97 (6.554)	42.53 (6.596)	43.09 (6.639)	43.61 (6.678)	44.29 (6.729)	44.49 (6.744)	44.50 (6.744)	44.51 (6.745)	44.51 (6.745)	44.52 (6.745)	44.52 (6.745)	44.52 (6.745)
V <sub>27</sub>	65.69 (8.166)	66.29 (8.202)	67.46 (8.274)	68.07 (8.311)	68.46 (8.334)	68.60 (8.343)	68.62 (8.343)	68.62 (8.344)	68.62 (8.344)	68.62 (8.344)	68.63 (8.344)	68.63 (8.344)
V <sub>28</sub>	57.65 (7.658)	58.40 (7.707)	58.98 (7.744)	59.66 (7.788)	60.34 (7.831)	60.63 (7.85)	60.88 (7.866)	60.90 (7.867)	60.91 (7.868)	60.91 (7.868)	60.91 (7.868)	60.93 (7.869)
V <sub>29</sub>	50.44 (7.165)	51.35 (7.229)	52.03 (7.277)	52.99 (7.341)	53.59 (7.382)	53.88 (7.401)	54.42 (7.435)	54.43 (7.436)	54.44 (7.437)	54.45 (7.437)	54.45 (7.437)	54.45 (7.437)
V <sub>30</sub>	42.80 (6.597)	43.70 (6.665)	43.70 (6.663)	44.18 (6.699)	44.89 (6.753)	45.25 (6.778)	45.35 (6.785)	45.23 (6.775)	45.23 (6.775)	45.23 (6.775)	45.23 (6.776)	45.24 (6.776)
C.D.	0.491	0.487	0.489	0.5	0.49	0.474	0.464	0.454	0.448	0.445	0.443	0.443
SE(M)	0.175	0.174	0.174	0.178	0.175	0.169	0.166	0.162	0.16	0.159	0.158	0.158
C.V.	5.667	5.586	5.563	5.645	5.5	5.297	5.172	5.053	4.991	4.947	4.925	4.931

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XIX. Number of roots of *Ascocentrum* hybrids/varieties during the period 2016-17**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	13.00 (3.736)	13.00 (3.736)	13.20 (3.764)	13.80 (3.844)	14.60 (3.946)	14.80 (3.971)	15.60 (4.071)	15.60 (4.071)	16.60 (4.192)	16.80 (4.214)	17.60 (4.307)	18.40 (4.399)
V <sub>2</sub>	8.40 (3.044)	9.80 (3.234)	10.20 (3.303)	10.20 (3.316)	10.60 (3.379)	11.00 (3.442)	11.20 (3.465)	12.00 (3.585)	12.40 (3.643)	12.80 (3.695)	13.60 (3.802)	14.00 (3.845)
V <sub>3</sub>	6.40 (2.714)	7.00 (2.82)	7.00 (2.828)	7.80 (2.964)	8.40 (3.061)	8.40 (3.061)	8.60 (3.096)	8.80 (3.128)	9.20 (3.19)	9.20 (3.19)	9.40 (3.218)	10.00 (3.311)
V <sub>4</sub>	18.80 (4.444)	18.80 (4.444)	19.20 (4.489)	19.40 (4.511)	19.80 (4.557)	20.20 (4.6)	20.80 (4.665)	21.00 (4.686)	22.00 (4.792)	22.60 (4.854)	23.60 (4.955)	24.60 (5.055)
V <sub>5</sub>	9.00 (3.143)	9.00 (3.143)	9.40 (3.213)	10.00 (3.3)	10.20 (3.334)	10.80 (3.421)	11.60 (3.54)	12.00 (3.592)	13.00 (3.73)	13.40 (3.782)	14.00 (3.856)	14.60 (3.933)
V <sub>6</sub>	12.20 (3.63)	12.20 (3.63)	12.80 (3.713)	13.20 (3.767)	14.20 (3.898)	14.40 (3.924)	14.60 (3.949)	15.20 (4.024)	16.20 (4.146)	16.80 (4.217)	17.80 (4.334)	18.60 (4.424)
V <sub>7</sub>	7.60 (2.927)	8.40 (3.06)	8.60 (3.094)	9.22 (3.192)	9.42 (3.224)	9.42 (3.224)	9.64 (3.256)	10.10 (3.323)	10.30 (3.351)	10.18 (3.331)	10.44 (3.367)	10.64 (3.399)
V <sub>8</sub>	12.20 (3.587)	12.40 (3.61)	12.80 (3.675)	13.00 (3.703)	13.40 (3.757)	13.60 (3.789)	14.60 (3.912)	14.60 (3.912)	15.20 (3.992)	15.40 (4.014)	16.40 (4.138)	17.20 (4.234)
V <sub>9</sub>	1.40 (1.522)	1.80 (1.639)	2.40 (1.794)	2.80 (1.924)	3.20 (2.034)	3.80 (2.17)	4.40 (2.313)	4.40 (2.313)	4.80 (2.393)	5.20 (2.474)	5.80 (2.577)	6.60 (2.71)
V <sub>10</sub>	7.80 (2.945)	7.80 (2.945)	8.00 (2.982)	8.40 (3.041)	8.40 (3.041)	8.60 (3.072)	9.40 (3.199)	9.40 (3.199)	9.80 (3.252)	10.00 (3.287)	10.40 (3.339)	10.80 (3.399)
V <sub>11</sub>	8.40 (3.047)	8.60 (3.083)	8.80 (3.112)	9.60 (3.234)	9.80 (3.262)	10.20 (3.318)	10.40 (3.346)	11.00 (3.442)	11.40 (3.495)	11.60 (3.521)	11.80 (3.547)	12.20 (3.597)
V <sub>12</sub>	7.00 (2.802)	7.40 (2.88)	7.80 (2.945)	8.00 (2.978)	8.00 (2.978)	8.60 (3.078)	8.80 (3.112)	8.80 (3.112)	9.00 (3.138)	9.00 (3.138)	9.60 (3.231)	10.00 (3.29)
V <sub>13</sub>	7.40 (2.893)	7.60 (2.927)	8.00 (2.991)	8.40 (3.054)	8.80 (3.123)	9.40 (3.215)	9.60 (3.246)	9.60 (3.246)	10.40 (3.363)	10.80 (3.42)	11.40 (3.504)	12.40 (3.645)
V <sub>14</sub>	6.00 (2.643)	6.20 (2.677)	6.20 (2.677)	7.00 (2.822)	7.20 (2.853)	7.40 (2.89)	7.60 (2.919)	7.80 (2.956)	8.00 (2.99)	8.20 (3.018)	8.20 (3.018)	8.20 (3.009)
V <sub>15</sub>	6.60 (2.75)	7.40 (2.893)	7.60 (2.927)	8.40 (3.063)	8.80 (3.128)	9.40 (3.222)	10.00 (3.314)	10.00 (3.314)	11.00 (3.462)	11.20 (3.489)	12.20 (3.63)	13.20 (3.764)
V <sub>16</sub>	5.60 (2.531)	6.40 (2.695)	7.00 (2.794)	7.00 (2.794)	7.60 (2.909)	8.00 (2.972)	8.00 (2.972)	8.80 (3.105)	9.40 (3.197)	10.20 (3.32)	10.40 (3.346)	11.20 (3.458)

**APPENDIX XIX. continued**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	6.20 (2.645)	6.40 (2.68)	7.00 (2.802)	7.40 (2.865)	7.60 (2.895)	8.40 (3.028)	8.40 (3.028)	8.60 (3.067)	9.20 (3.153)	9.40 (3.181)	10.20 (3.3)	10.80 (3.386)
V <sub>18</sub>	4.60 (2.356)	4.80 (2.395)	5.60 (2.55)	5.40 (2.511)	6.00 (2.619)	6.00 (2.619)	6.20 (2.652)	6.20 (2.652)	7.00 (2.801)	7.20 (2.831)	7.40 (2.859)	7.60 (2.886)
V <sub>19</sub>	6.00 (2.632)	7.00 (2.819)	7.60 (2.931)	8.40 (3.065)	8.80 (3.128)	9.00 (3.161)	9.60 (3.252)	9.60 (3.252)	10.00 (3.31)	10.40 (3.369)	11.20 (3.482)	11.60 (3.534)
V <sub>20</sub>	5.80 (2.582)	5.80 (2.582)	6.80 (2.772)	7.00 (2.82)	7.20 (2.856)	8.00 (2.996)	8.20 (3.027)	8.40 (3.061)	9.20 (3.189)	9.20 (3.189)	10.20 (3.342)	10.60 (3.4)
V <sub>21</sub>	6.20 (2.664)	6.40 (2.701)	7.00 (2.809)	7.80 (2.951)	8.00 (2.979)	9.00 (3.148)	9.20 (3.175)	9.40 (3.21)	10.20 (3.331)	10.40 (3.363)	11.20 (3.484)	12.20 (3.626)
V <sub>22</sub>	5.20 (2.482)	5.40 (2.525)	6.80 (2.787)	7.40 (2.893)	7.80 (2.962)	8.60 (3.096)	9.20 (3.19)	9.40 (3.223)	10.00 (3.313)	10.40 (3.373)	11.20 (3.488)	11.80 (3.576)
V <sub>23</sub>	16.00 (4.118)	16.20 (4.141)	16.20 (4.141)	16.80 (4.214)	17.00 (4.238)	17.40 (4.287)	18.40 (4.401)	18.60 (4.423)	19.40 (4.514)	19.60 (4.535)	20.60 (4.644)	21.00 (4.686)
V <sub>24</sub>	8.20 (3.005)	8.40 (3.041)	9.00 (3.146)	9.80 (3.27)	10.00 (3.303)	10.40 (3.361)	11.20 (3.477)	11.20 (3.477)	12.20 (3.619)	12.60 (3.675)	13.20 (3.753)	13.80 (3.836)
V <sub>25</sub>	12.80 (3.685)	14.20 (3.863)	14.40 (3.893)	14.20 (3.869)	14.60 (3.924)	14.80 (3.952)	15.60 (4.043)	16.00 (4.1)	16.40 (4.15)	17.00 (4.217)	18.00 (4.335)	18.80 (4.42)
V <sub>26</sub>	11.40 (3.485)	12.00 (3.572)	12.20 (3.6)	12.60 (3.657)	13.00 (3.717)	13.00 (3.717)	13.60 (3.787)	14.00 (3.845)	14.60 (3.928)	15.40 (4.029)	16.00 (4.1)	16.40 (4.146)
V <sub>27</sub>	8.60 (3.067)	9.00 (3.136)	9.20 (3.164)	9.60 (3.221)	9.80 (3.249)	10.00 (3.285)	10.20 (3.312)	10.80 (3.403)	11.00 (3.428)	11.20 (3.454)	11.40 (3.479)	11.80 (3.531)
V <sub>28</sub>	7.80 (2.929)	8.00 (2.966)	8.40 (3.036)	9.40 (3.2)	9.40 (3.2)	10.00 (3.294)	10.60 (3.379)	10.60 (3.379)	11.60 (3.526)	11.80 (3.554)	12.60 (3.666)	13.20 (3.753)
V <sub>29</sub>	8.40 (2.968)	8.60 (3.011)	9.20 (3.126)	10.00 (3.248)	10.20 (3.288)	10.80 (3.377)	11.20 (3.441)	11.40 (3.463)	12.00 (3.548)	12.20 (3.569)	13.00 (3.68)	14.00 (3.817)
V <sub>30</sub>	11.20 (3.447)	11.20 (3.447)	11.40 (3.487)	12.20 (3.593)	12.60 (3.658)	13.00 (3.707)	13.60 (3.792)	14.00 (3.842)	15.00 (3.977)	15.20 (4.003)	15.80 (4.072)	16.40 (4.156)
<b>C.D.</b>	0.52	0.527	0.5	0.497	0.482	0.479	0.499	0.486	0.495	0.513	0.541	0.559
<b>SE(m)±</b>	0.185	0.188	0.178	0.177	0.172	0.171	0.178	0.173	0.176	0.183	0.193	0.199
<b>C.V.</b>	13.75	13.67	12.663	12.272	11.703	11.409	11.671	11.243	11.156	11.432	11.778	11.922

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XX. Number of roots of *Ascoctrum* hybrids/varieties during the period 2017-18**

Variety	Apr. '17	May. '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	18.60 (4.422)	18.80 (4.444)	19.20 (4.489)	19.20 (4.489)	19.40 (4.511)	19.80 (4.553)	20.00 (4.576)	20.40 (4.621)	20.80 (4.662)	21.00 (4.682)	21.60 (4.746)	22.00 (4.789)
V <sub>2</sub>	14.40 (3.899)	14.40 (3.899)	14.60 (3.92)	14.80 (3.946)	15.00 (3.972)	15.20 (3.997)	15.80 (4.073)	16.00 (4.097)	16.00 (4.097)	16.60 (4.175)	17.20 (4.243)	17.40 (4.27)
V <sub>3</sub>	10.40 (3.368)	11.00 (3.458)	11.80 (3.569)	12.20 (3.623)	12.20 (3.623)	12.40 (3.654)	13.20 (3.759)	13.40 (3.785)	13.60 (3.815)	14.80 (3.969)	15.60 (4.069)	16.20 (4.141)
V <sub>4</sub>	24.60 (5.055)	24.60 (5.055)	25.60 (5.154)	25.80 (5.173)	25.80 (5.173)	26.40 (5.23)	26.80 (5.269)	27.00 (5.289)	27.20 (5.307)	27.60 (5.344)	28.20 (5.401)	28.80 (5.454)
V <sub>5</sub>	15.00 (3.985)	15.00 (3.985)	15.60 (4.055)	15.80 (4.081)	15.80 (4.081)	16.40 (4.154)	16.60 (4.177)	16.80 (4.203)	17.20 (4.246)	17.60 (4.296)	17.80 (4.321)	18.40 (4.388)
V <sub>6</sub>	18.80 (4.448)	19.00 (4.469)	19.20 (4.49)	19.60 (4.534)	20.00 (4.58)	20.20 (4.601)	20.20 (4.601)	21.00 (4.688)	21.20 (4.708)	21.40 (4.73)	21.60 (4.751)	22.40 (4.835)
V <sub>7</sub>	11.18 (3.479)	11.38 (3.508)	11.58 (3.534)	11.98 (3.592)	12.80 (3.707)	13.60 (3.811)	14.40 (3.917)	14.60 (3.945)	15.00 (3.998)	15.80 (4.096)	16.00 (4.121)	16.60 (4.194)
V <sub>8</sub>	17.20 (4.234)	17.20 (4.234)	17.40 (4.258)	17.80 (4.305)	18.40 (4.376)	18.40 (4.376)	18.40 (4.376)	18.80 (4.416)	19.40 (4.483)	19.80 (4.533)	20.00 (4.553)	20.40 (4.595)
V <sub>9</sub>	6.80 (2.753)	6.80 (2.753)	7.20 (2.83)	7.60 (2.899)	8.00 (2.978)	8.20 (3.005)	8.60 (3.072)	8.80 (3.106)	9.20 (3.177)	9.60 (3.244)	10.00 (3.306)	10.20 (3.332)
V <sub>10</sub>	11.00 (3.432)	11.40 (3.488)	12.00 (3.575)	12.20 (3.604)	12.40 (3.63)	12.60 (3.654)	13.00 (3.701)	13.60 (3.791)	13.80 (3.818)	14.20 (3.866)	14.20 (3.866)	14.40 (3.896)
V <sub>11</sub>	12.60 (3.664)	13.20 (3.748)	13.40 (3.777)	13.60 (3.808)	14.00 (3.862)	14.00 (3.862)	14.80 (3.966)	15.60 (4.068)	15.60 (4.068)	16.00 (4.117)	16.20 (4.142)	16.40 (4.167)
V <sub>12</sub>	10.00 (3.29)	10.40 (3.351)	11.00 (3.44)	11.60 (3.53)	11.80 (3.561)	12.20 (3.619)	12.80 (3.697)	12.80 (3.697)	13.00 (3.727)	13.60 (3.804)	13.80 (3.829)	14.60 (3.935)
V <sub>13</sub>	12.60 (3.67)	12.60 (3.67)	13.40 (3.781)	13.60 (3.805)	14.00 (3.861)	14.40 (3.917)	14.60 (3.941)	14.80 (3.965)	15.20 (4.014)	15.20 (4.014)	15.40 (4.043)	16.20 (4.138)
V <sub>14</sub>	8.60 (3.077)	8.80 (3.11)	9.20 (3.181)	9.60 (3.24)	9.80 (3.265)	10.40 (3.351)	10.80 (3.415)	11.60 (3.53)	11.60 (3.53)	12.00 (3.581)	12.60 (3.67)	12.80 (3.697)
V <sub>15</sub>	13.20 (3.764)	13.20 (3.764)	13.20 (3.764)	13.40 (3.792)	13.40 (3.792)	13.60 (3.819)	14.20 (3.897)	14.40 (3.922)	14.80 (3.972)	15.20 (4.024)	15.40 (4.048)	15.60 (4.072)
V <sub>16</sub>	11.40 (3.495)	11.80 (3.56)	12.80 (3.699)	13.40 (3.784)	13.40 (3.784)	13.80 (3.839)	14.20 (3.89)	14.60 (3.941)	14.80 (3.964)	15.40 (4.041)	15.80 (4.093)	16.00 (4.117)



**APPENDIX XX. continued**

Variety	Apr. '17	May. '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	11.00 (3.425)	11.00 (3.425)	11.60 (3.51)	12.20 (3.597)	12.20 (3.597)	12.20 (3.597)	12.40 (3.622)	13.20 (3.736)	13.60 (3.789)	13.60 (3.789)	14.00 (3.846)	14.20 (3.87)
V <sub>18</sub>	7.60 (2.886)	7.80 (2.917)	8.20 (2.996)	9.20 (3.151)	9.40 (3.177)	10.00 (3.268)	10.40 (3.331)	11.00 (3.432)	11.20 (3.464)	11.60 (3.52)	12.00 (3.569)	12.00 (3.569)
V <sub>19</sub>	11.80 (3.565)	12.00 (3.595)	12.40 (3.649)	12.80 (3.707)	13.20 (3.759)	13.40 (3.785)	13.40 (3.785)	14.00 (3.864)	14.60 (3.944)	15.00 (3.996)	15.00 (3.996)	15.40 (4.045)
V <sub>20</sub>	10.80 (3.429)	11.20 (3.488)	11.60 (3.547)	12.20 (3.63)	12.60 (3.686)	12.60 (3.686)	12.80 (3.713)	13.20 (3.765)	13.40 (3.792)	13.60 (3.82)	14.00 (3.871)	14.40 (3.922)
V <sub>21</sub>	12.20 (3.626)	12.20 (3.626)	12.40 (3.655)	12.60 (3.683)	13.20 (3.761)	13.40 (3.785)	13.60 (3.813)	13.80 (3.838)	14.20 (3.893)	14.20 (3.893)	14.40 (3.919)	14.60 (3.943)
V <sub>22</sub>	11.80 (3.576)	11.80 (3.576)	11.80 (3.576)	11.60 (3.547)	12.20 (3.631)	12.40 (3.658)	13.00 (3.74)	13.00 (3.74)	13.00 (3.74)	13.80 (3.846)	14.00 (3.872)	14.40 (3.923)
V <sub>23</sub>	21.00 (4.686)	21.20 (4.707)	21.40 (4.728)	21.60 (4.749)	22.00 (4.792)	22.40 (4.833)	22.40 (4.833)	22.80 (4.873)	23.60 (4.955)	23.60 (4.955)	23.80 (4.975)	24.60 (5.055)
V <sub>24</sub>	13.80 (3.836)	13.80 (3.836)	14.00 (3.864)	14.60 (3.944)	15.00 (3.993)	15.40 (4.044)	15.40 (4.044)	15.60 (4.069)	16.00 (4.116)	16.40 (4.166)	16.40 (4.166)	16.80 (4.212)
V <sub>25</sub>	19.00 (4.446)	19.00 (4.446)	19.40 (4.488)	19.40 (4.488)	19.80 (4.533)	20.20 (4.579)	20.40 (4.6)	20.80 (4.644)	21.20 (4.683)	21.60 (4.727)	22.00 (4.771)	22.20 (4.789)
V <sub>26</sub>	16.60 (4.173)	16.80 (4.197)	17.00 (4.22)	17.20 (4.246)	17.80 (4.315)	17.80 (4.315)	18.20 (4.363)	18.80 (4.431)	19.00 (4.45)	19.40 (4.496)	19.60 (4.52)	20.00 (4.562)
V <sub>27</sub>	12.20 (3.597)	12.60 (3.656)	13.00 (3.704)	13.20 (3.735)	13.60 (3.789)	13.60 (3.789)	14.00 (3.84)	14.80 (3.948)	14.80 (3.948)	15.20 (3.997)	15.40 (4.028)	15.60 (4.057)
V <sub>28</sub>	13.20 (3.753)	13.20 (3.753)	13.40 (3.782)	13.80 (3.835)	14.20 (3.885)	14.40 (3.91)	14.40 (3.91)	14.60 (3.936)	15.20 (4.012)	15.40 (4.037)	15.40 (4.037)	15.80 (4.083)
V <sub>29</sub>	14.00 (3.817)	14.00 (3.817)	15.00 (3.949)	15.40 (4.003)	15.60 (4.027)	15.60 (4.027)	15.80 (4.057)	16.00 (4.076)	16.60 (4.152)	16.60 (4.152)	17.00 (4.2)	17.60 (4.273)
V <sub>30</sub>	16.40 (4.156)	16.60 (4.18)	17.00 (4.224)	17.00 (4.224)	17.60 (4.298)	18.00 (4.345)	18.20 (4.368)	18.40 (4.392)	18.80 (4.434)	19.00 (4.456)	19.00 (4.456)	19.40 (4.498)
<b>C.D.</b>	0.533	0.525	0.521	0.509	0.503	0.507	0.507	0.491	0.49	0.482	0.472	0.479
<b>SE(m)±</b>	0.19	0.187	0.186	0.182	0.18	0.181	0.181	0.175	0.175	0.172	0.168	0.171
<b>C.V.</b>	11.278	11.053	10.801	10.434	10.209	10.191	10.072	9.647	9.534	9.281	9.006	9.028

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XXI. Length of roots of *Ascocentrum* hybrids/varieties during the period 2016-17, cm**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	127.67 (11.339)	128.67 (11.383)	130.67 (11.47)	131.00 (11.485)	131.33 (11.5)	132.00 (11.529)	133.67 (11.601)	134.67 (11.645)	135.50 (11.68)	136.67 (11.73)	137.83 (11.78)	139.67 (11.857)
V <sub>2</sub>	130.00 (11.441)	133.33 (11.584)	135.67 (11.684)	137.00 (11.742)	137.67 (11.771)	140.00 (11.869)	143.33 (12.005)	144.67 (12.061)	146.17 (12.122)	147.83 (12.191)	150.03 (12.28)	151.33 (12.333)
V <sub>3</sub>	122.67 (11.099)	127.00 (11.289)	129.87 (11.414)	131.23 (11.474)	132.67 (11.536)	134.67 (11.626)	137.67 (11.754)	139.33 (11.825)	141.00 (11.898)	143.17 (11.99)	145.00 (12.068)	146.33 (12.124)
V <sub>4</sub>	151.00 (12.324)	153.67 (12.433)	155.67 (12.514)	157.67 (12.593)	158.67 (12.632)	160.00 (12.685)	162.67 (12.79)	164.67 (12.868)	166.67 (12.946)	169.33 (13.049)	170.00 (13.074)	172.00 (13.151)
V <sub>5</sub>	119.67 (10.979)	122.67 (11.115)	129.00 (11.394)	131.00 (11.481)	133.00 (11.568)	136.67 (11.728)	138.33 (11.799)	141.00 (11.911)	143.33 (12.009)	146.33 (12.132)	149.00 (12.241)	151.33 (12.335)
V <sub>6</sub>	132.33 (11.518)	135.67 (11.663)	138.67 (11.793)	140.33 (11.861)	141.67 (11.917)	143.33 (11.988)	171.67 (13.115)	174.00 (13.205)	176.00 (13.279)	178.00 (13.353)	180.33 (13.441)	181.67 (13.491)
V <sub>7</sub>	154.00 (12.447)	159.33 (12.661)	161.00 (12.727)	162.67 (12.792)	165.67 (12.91)	168.33 (13.013)	169.83 (13.07)	170.67 (13.102)	171.67 (13.14)	172.67 (13.177)	174.00 (13.227)	175.20 (13.272)
V <sub>8</sub>	151.00 (12.309)	153.33 (12.401)	155.67 (12.496)	157.00 (12.551)	158.33 (12.603)	159.60 (12.656)	160.90 (12.708)	162.67 (12.779)	164.00 (12.833)	165.67 (12.898)	167.00 (12.948)	168.33 (13.002)
V <sub>9</sub>	25.73 (5.13)	27.37 (5.297)	28.73 (5.438)	30.03 (5.557)	30.23 (5.569)	31.00 (5.633)	31.70 (5.697)	32.17 (5.741)	32.77 (5.795)	33.47 (5.859)	33.83 (5.891)	37.97 (6.221)
V <sub>10</sub>	109.33 (10.5)	110.33 (10.548)	111.00 (10.58)	113.00 (10.673)	114.43 (10.739)	115.50 (10.789)	116.67 (10.843)	119.00 (10.951)	121.33 (11.056)	123.67 (11.161)	126.00 (11.265)	128.50 (11.374)
V <sub>11</sub>	86.00 (9.322)	87.50 (9.4)	89.67 (9.513)	91.73 (9.62)	93.33 (9.702)	95.33 (9.802)	97.00 (9.885)	99.33 (9.998)	100.67 (10.065)	101.83 (10.121)	104.00 (10.224)	106.83 (10.359)
V <sub>12</sub>	92.67 (9.663)	95.50 (9.811)	98.00 (9.935)	99.43 (10.008)	100.80 (10.077)	101.53 (10.115)	102.93 (10.187)	103.33 (10.209)	105.00 (10.291)	106.50 (10.364)	108.10 (10.44)	110.00 (10.53)
V <sub>13</sub>	155.33 (12.501)	158.00 (12.606)	160.67 (12.711)	162.67 (12.79)	164.67 (12.869)	166.50 (12.94)	169.00 (13.036)	171.67 (13.138)	175.00 (13.264)	176.67 (13.327)	177.67 (13.364)	178.67 (13.402)
V <sub>14</sub>	154.00 (12.447)	159.33 (12.661)	161.00 (12.727)	162.67 (12.792)	165.67 (12.91)	168.33 (13.013)	170.17 (13.083)	172.00 (13.152)	173.00 (13.19)	174.33 (13.24)	175.67 (13.289)	176.33 (13.314)
V <sub>15</sub>	165.00 (12.868)	170.33 (13.077)	173.33 (13.189)	174.67 (13.24)	177.33 (13.343)	179.20 (13.414)	181.00 (13.483)	183.33 (13.569)	184.67 (13.619)	186.33 (13.68)	187.67 (13.729)	188.67 (13.765)
V <sub>16</sub>	171.67 (13.12)	177.00 (13.327)	180.00 (13.438)	181.00 (13.476)	184.00 (13.591)	186.03 (13.667)	187.67 (13.729)	189.47 (13.796)	191.33 (13.864)	193.00 (13.925)	194.17 (13.967)	194.33 (13.973)

**APPENDIX XXI. continued**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	111.00 (10.579)	113.33 (10.687)	117.00 (10.852)	119.00 (10.942)	120.43 (11.005)	121.90 (11.071)	123.00 (11.122)	125.33 (11.228)	127.67 (11.332)	130.00 (11.434)	132.67 (11.55)	135.17 (11.657)
V <sub>18</sub>	169.33 (13.044)	172.50 (13.165)	174.33 (13.234)	177.00 (13.335)	179.33 (13.422)	180.67 (13.472)	181.67 (13.51)	184.67 (13.622)	185.67 (13.659)	186.00 (13.671)	187.00 (13.707)	187.67 (13.731)
V <sub>19</sub>	48.83 (7.053)	49.33 (7.088)	49.83 (7.122)	50.67 (7.18)	51.50 (7.236)	52.27 (7.291)	52.77 (7.326)	53.30 (7.362)	54.87 (7.471)	55.20 (7.493)	55.60 (7.52)	56.00 (7.546)
V <sub>20</sub>	49.83 (7.11)	50.33 (7.148)	52.00 (7.268)	53.33 (7.361)	55.43 (7.506)	56.67 (7.588)	57.35 (7.633)	57.87 (7.666)	58.67 (7.718)	59.51 (7.774)	60.27 (7.823)	61.37 (7.894)
V <sub>21</sub>	45.23 (6.777)	46.20 (6.85)	47.33 (6.938)	49.33 (7.085)	50.10 (7.14)	52.27 (7.292)	53.00 (7.342)	53.70 (7.389)	55.30 (7.497)	55.87 (7.534)	56.67 (7.586)	57.40 (7.634)
V <sub>22</sub>	74.20 (8.648)	75.00 (8.692)	76.00 (8.75)	77.00 (8.807)	77.90 (8.858)	78.47 (8.892)	78.99 (8.922)	81.33 (9.047)	82.07 (9.089)	82.47 (9.111)	83.57 (9.169)	84.83 (9.236)
V <sub>23</sub>	85.80 (9.316)	86.67 (9.362)	87.33 (9.398)	88.47 (9.458)	90.00 (9.538)	90.33 (9.556)	90.80 (9.58)	91.53 (9.618)	92.33 (9.66)	93.20 (9.704)	94.00 (9.746)	94.93 (9.793)
V <sub>24</sub>	86.60 (9.197)	87.67 (9.255)	88.33 (9.291)	89.63 (9.361)	91.00 (9.433)	91.40 (9.454)	92.20 (9.479)	94.00 (9.566)	94.87 (9.601)	95.67 (9.642)	98.13 (9.701)	100.00 (9.763)
V <sub>25</sub>	89.00 (9.643)	90.33 (9.712)	91.00 (9.747)	92.67 (9.832)	93.67 (9.882)	94.00 (9.899)	94.87 (9.959)	95.87 (10.015)	96.33 (10.048)	97.23 (10.093)	98.47 (10.219)	99.77 (10.314)
V <sub>26</sub>	41.33 (6.505)	42.17 (6.569)	42.67 (6.606)	43.27 (6.651)	44.07 (6.711)	45.00 (6.78)	45.57 (6.822)	46.33 (6.878)	47.00 (6.926)	47.67 (6.974)	48.67 (7.044)	49.33 (7.09)
V <sub>27</sub>	39.23 (6.341)	40.37 (6.43)	41.33 (6.506)	43.00 (6.632)	43.83 (6.694)	46.10 (6.857)	46.90 (6.916)	47.43 (6.955)	48.87 (7.054)	49.20 (7.078)	50.17 (7.145)	50.40 (7.161)
V <sub>28</sub>	47.67 (6.934)	47.67 (6.934)	51.67 (7.208)	52.83 (7.291)	54.17 (7.383)	55.67 (7.489)	57.23 (7.594)	58.43 (7.675)	59.73 (7.761)	61.10 (7.851)	62.43 (7.938)	63.97 (8.036)
V <sub>29</sub>	80.83 (9.004)	82.50 (9.098)	84.33 (9.201)	86.73 (9.326)	89.33 (9.463)	93.00 (9.65)	94.67 (9.737)	98.33 (9.919)	100.00 (10.005)	101.67 (10.087)	104.67 (10.232)	107.63 (10.376)
V <sub>30</sub>	103.33 (10.203)	104.00 (10.235)	105.33 (10.303)	108.00 (10.433)	109.43 (10.501)	110.57 (10.555)	112.00 (10.624)	114.00 (10.716)	116.00 (10.808)	118.00 (10.898)	120.00 (10.988)	122.17 (11.085)
C.D.	0.976	0.953	0.955	0.946	0.944	0.92	0.907	0.903	0.89	0.893	0.909	0.929
SE(m)	0.344	0.336	0.337	0.334	0.333	0.324	0.32	0.318	0.314	0.315	0.321	0.328
C.V.	5.974	5.773	5.728	5.633	5.578	5.398	5.272	5.21	5.102	5.091	5.149	5.224

Note: The data in parenthesis indicate square root transformed value

**APPENDIX XXII. Length of roots of *Ascocentrum* hybrids/varieties during the 2017-18, cm**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	141.67 (11.94)	143.00 (11.996)	144.67 (12.064)	145.67 (12.105)	146.67 (12.146)	148.00 (12.201)	149.33 (12.255)	150.33 (12.296)	151.33 (12.336)	152.00 (12.363)	153.00 (12.404)	153.67 (12.431)
V <sub>2</sub>	151.33 (12.333)	152.67 (12.387)	153.67 (12.427)	154.33 (12.455)	156.33 (12.536)	157.67 (12.59)	159.33 (12.657)	161.00 (12.723)	162.67 (12.789)	164.33 (12.854)	166.33 (12.932)	166.67 (12.946)
V <sub>3</sub>	147.33 (12.167)	149.17 (12.241)	151.33 (12.329)	152.33 (12.371)	153.67 (12.424)	155.17 (12.485)	156.37 (12.533)	157.00 (12.558)	157.67 (12.584)	159.00 (12.638)	160.00 (12.677)	161.00 (12.717)
V <sub>4</sub>	172.50 (13.17)	174.17 (13.232)	175.00 (13.263)	176.00 (13.301)	177.00 (13.338)	178.17 (13.382)	179.33 (13.425)	180.67 (13.474)	181.67 (13.511)	182.67 (13.548)	183.67 (13.585)	184.67 (13.621)
V <sub>5</sub>	152.83 (12.397)	154.00 (12.444)	154.67 (12.471)	155.67 (12.511)	156.67 (12.551)	157.50 (12.584)	158.67 (12.63)	159.67 (12.671)	161.00 (12.723)	162.63 (12.788)	164.60 (12.865)	165.60 (12.904)
V <sub>6</sub>	183.67 (13.569)	185.83 (13.652)	187.83 (13.728)	190.00 (13.809)	191.33 (13.857)	193.33 (13.931)	194.33 (13.967)	195.33 (14.002)	196.33 (14.038)	188.33 (13.734)	192.33 (13.886)	194.33 (13.96)
V <sub>7</sub>	175.67 (13.29)	177.00 (13.339)	177.83 (13.371)	179.00 (13.415)	180.67 (13.478)	182.33 (13.54)	184.00 (13.601)	185.33 (13.65)	187.33 (13.723)	188.67 (13.772)	190.33 (13.832)	191.67 (13.88)
V <sub>8</sub>	169.00 (13.028)	170.50 (13.086)	172.33 (13.157)	174.67 (13.245)	177.00 (13.333)	179.33 (13.42)	181.00 (13.483)	183.33 (13.569)	184.93 (13.629)	186.67 (13.692)	188.40 (13.756)	189.80 (13.807)
V <sub>9</sub>	38.39 (6.251)	38.99 (6.298)	39.65 (6.349)	39.99 (6.378)	40.71 (6.434)	41.30 (6.483)	41.63 (6.507)	42.70 (6.59)	43.39 (6.643)	43.91 (6.681)	44.60 (6.732)	45.23 (6.779)
V <sub>10</sub>	129.57 (11.422)	131.33 (11.499)	133.20 (11.579)	135.00 (11.657)	136.87 (11.737)	138.50 (11.806)	140.37 (11.885)	141.97 (11.951)	143.67 (12.023)	145.47 (12.098)	147.10 (12.165)	149.00 (12.242)
V <sub>11</sub>	107.80 (10.406)	108.50 (10.438)	109.50 (10.483)	111.13 (10.562)	112.23 (10.614)	113.00 (10.646)	113.87 (10.686)	114.97 (10.738)	116.30 (10.802)	117.63 (10.866)	119.30 (10.945)	121.30 (11.041)
V <sub>12</sub>	111.67 (10.609)	113.37 (10.688)	114.33 (10.733)	115.77 (10.799)	117.17 (10.863)	118.30 (10.915)	119.63 (10.976)	121.00 (11.038)	122.63 (11.111)	124.13 (11.178)	125.33 (11.231)	127.00 (11.305)
V <sub>13</sub>	180.00 (13.451)	181.33 (13.5)	182.67 (13.549)	183.67 (13.586)	184.67 (13.623)	185.67 (13.659)	186.67 (13.696)	187.67 (13.733)	189.00 (13.781)	190.00 (13.817)	191.00 (13.853)	192.00 (13.889)
V <sub>14</sub>	177.33 (13.352)	178.50 (13.395)	180.00 (13.452)	181.67 (13.514)	183.33 (13.576)	185.00 (13.638)	186.33 (13.687)	188.33 (13.76)	189.67 (13.808)	191.33 (13.868)	192.67 (13.916)	193.33 (13.94)
V <sub>15</sub>	189.67 (13.802)	181.67 (13.497)	185.67 (13.651)	187.53 (13.72)	188.53 (13.756)	189.87 (13.805)	190.87 (13.842)	191.00 (13.846)	191.67 (13.87)	192.33 (13.893)	193.00 (13.917)	193.67 (13.94)
V <sub>16</sub>	195.00 (13.996)	196.00 (14.032)	197.00 (14.068)	198.20 (14.11)	199.00 (14.138)	200.20 (14.181)	201.20 (14.216)	202.20 (14.251)	202.87 (14.274)	203.87 (14.309)	204.63 (14.336)	205.30 (14.36)

**APPENDIX XXII. continued**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	136.00 (11.693)	137.67 (11.765)	139.20 (11.83)	140.67 (11.892)	142.20 (11.957)	143.80 (12.024)	145.63 (12.1)	147.23 (12.165)	148.60 (12.222)	150.20 (12.288)	151.70 (12.349)	153.53 (12.423)
V <sub>18</sub>	189.67 (13.804)	191.33 (13.865)	192.33 (13.901)	193.00 (13.924)	185.67 (13.644)	190.03 (13.81)	191.53 (13.865)	192.20 (13.889)	193.87 (13.95)	195.20 (13.998)	195.70 (14.015)	196.20 (14.033)
V <sub>19</sub>	56.93 (7.608)	58.37 (7.7)	59.40 (7.765)	60.80 (7.857)	61.03 (7.872)	61.53 (7.903)	62.87 (7.989)	63.93 (8.057)	65.10 (8.129)	66.10 (8.191)	66.67 (8.224)	68.17 (8.315)
V <sub>20</sub>	62.83 (7.986)	64.03 (8.062)	64.80 (8.11)	65.60 (8.159)	66.37 (8.205)	67.33 (8.264)	68.07 (8.308)	69.00 (8.364)	69.87 (8.415)	71.37 (8.504)	72.27 (8.557)	73.07 (8.603)
V <sub>21</sub>	57.94 (7.673)	58.60 (7.716)	59.13 (7.751)	59.70 (7.788)	63.03 (7.996)	63.56 (8.029)	64.33 (8.076)	65.33 (8.136)	66.27 (8.193)	67.23 (8.251)	66.60 (8.212)	68.40 (8.321)
V <sub>22</sub>	86.33 (9.318)	87.03 (9.355)	87.50 (9.379)	88.67 (9.442)	91.00 (9.561)	92.67 (9.647)	94.33 (9.731)	96.00 (9.815)	97.87 (9.909)	99.98 (10.013)	101.05 (10.065)	101.53 (10.092)
V <sub>23</sub>	95.67 (9.831)	97.37 (9.917)	98.63 (9.98)	100.17 (10.056)	101.63 (10.129)	103.00 (10.196)	105.00 (10.293)	107.00 (10.389)	108.93 (10.482)	110.63 (10.563)	112.37 (10.645)	114.17 (10.729)
V <sub>24</sub>	101.17 (9.806)	102.77 (9.878)	104.33 (9.932)	105.97 (9.998)	107.97 (10.096)	109.67 (10.163)	111.80 (10.26)	113.60 (10.357)	115.33 (10.454)	117.52 (10.558)	118.91 (10.625)	120.00 (10.675)
V <sub>25</sub>	97.89 (10.23)	98.98 (10.29)	99.67 (10.348)	100.43 (10.398)	101.27 (10.439)	102.50 (10.512)	103.74 (10.577)	105.29 (10.639)	106.74 (10.693)	108.39 (10.768)	109.59 (10.818)	111.66 (10.917)
V <sub>26</sub>	50.30 (7.159)	51.36 (7.234)	52.37 (7.303)	51.65 (7.253)	51.81 (7.263)	52.14 (7.286)	53.19 (7.357)	53.66 (7.388)	54.10 (7.417)	55.13 (7.484)	56.32 (7.562)	57.82 (7.658)
V <sub>27</sub>	51.48 (7.237)	52.62 (7.318)	53.57 (7.382)	52.78 (7.326)	55.57 (7.5)	55.99 (7.527)	56.96 (7.59)	57.66 (7.633)	58.27 (7.671)	58.97 (7.715)	59.99 (7.78)	61.09 (7.85)
V <sub>28</sub>	65.94 (8.163)	67.10 (8.234)	67.89 (8.281)	68.50 (8.317)	71.93 (8.528)	72.69 (8.572)	74.20 (8.657)	75.23 (8.716)	76.50 (8.789)	77.83 (8.863)	78.67 (8.909)	80.27 (8.998)
V <sub>29</sub>	108.47 (10.416)	109.88 (10.483)	111.06 (10.536)	112.41 (10.6)	114.12 (10.679)	116.49 (10.792)	118.06 (10.863)	119.22 (10.916)	120.66 (10.985)	121.66 (11.031)	122.99 (11.094)	124.16 (11.147)
V <sub>30</sub>	123.00 (11.123)	124.67 (11.197)	126.20 (11.264)	127.67 (11.328)	129.20 (11.396)	130.73 (11.463)	132.33 (11.532)	133.63 (11.588)	135.00 (11.647)	136.93 (11.73)	139.10 (11.823)	140.27 (11.87)
C.D.	0.919	0.938	0.931	0.924	0.966	0.955	0.965	0.971	0.964	0.999	0.983	0.973
SE(m)	0.324	0.331	0.328	0.326	0.341	0.337	0.34	0.342	0.34	0.352	0.347	0.343
C.V.	5.145	5.228	5.163	5.102	5.303	5.216	5.244	5.249	5.186	5.35	5.243	5.161

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XXIII. Girth of roots of *Ascocentrum* hybrids/varieties during the period 2016-17, cm**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>1</sub>	1.63 (1.622)	1.59 (1.609)	1.73 (1.652)	1.73 (1.652)	1.83 (1.682)	1.90 (1.702)	1.90 (1.702)	1.90 (1.702)	1.97 (1.722)	2.03 (1.742)	2.13 (1.77)	2.17 (1.779)
V <sub>2</sub>	1.53 (1.59)	1.56 (1.596)	1.70 (1.642)	1.77 (1.663)	1.80 (1.673)	1.90 (1.702)	1.93 (1.712)	1.97 (1.722)	2.10 (1.76)	2.17 (1.779)	2.27 (1.807)	2.37 (1.835)
V <sub>3</sub>	2.27 (1.807)	2.19 (1.786)	2.27 (1.807)	2.33 (1.826)	2.40 (1.844)	2.33 (1.826)	2.33 (1.826)	2.43 (1.853)	2.47 (1.862)	2.57 (1.888)	2.67 (1.915)	2.70 (1.923)
V <sub>4</sub>	1.60 (1.612)	1.60 (1.612)	1.73 (1.653)	1.80 (1.673)	1.87 (1.692)	1.90 (1.702)	2.00 (1.731)	2.03 (1.741)	2.10 (1.76)	2.17 (1.779)	2.27 (1.807)	2.30 (1.816)
V <sub>5</sub>	2.13 (1.769)	2.13 (1.769)	2.17 (1.779)	2.20 (1.788)	2.23 (1.798)	2.27 (1.807)	2.27 (1.807)	2.33 (1.825)	2.40 (1.843)	2.47 (1.861)	2.57 (1.888)	2.63 (1.905)
V <sub>6</sub>	1.73 (1.653)	1.73 (1.653)	1.87 (1.693)	1.93 (1.712)	1.97 (1.722)	2.00 (1.732)	2.10 (1.76)	2.13 (1.77)	2.20 (1.789)	2.23 (1.798)	2.33 (1.825)	2.40 (1.844)
V <sub>7</sub>	1.93 (1.712)	1.93 (1.712)	2.00 (1.731)	2.10 (1.76)	2.13 (1.769)	2.17 (1.778)	2.23 (1.797)	2.33 (1.825)	2.43 (1.852)	2.50 (1.869)	2.60 (1.896)	2.67 (1.914)
V <sub>8</sub>	1.77 (1.659)	1.83 (1.678)	1.93 (1.711)	1.93 (1.708)	2.03 (1.738)	1.97 (1.719)	2.07 (1.749)	2.10 (1.758)	2.10 (1.758)	2.20 (1.786)	2.30 (1.814)	2.33 (1.824)
V <sub>9</sub>	0.77 (1.329)	0.77 (1.329)	0.97 (1.402)	1.07 (1.437)	1.17 (1.472)	1.17 (1.472)	1.27 (1.505)	1.37 (1.538)	1.40 (1.549)	1.50 (1.581)	1.57 (1.602)	1.63 (1.623)
V <sub>10</sub>	1.83 (1.68)	1.83 (1.68)	1.97 (1.721)	1.97 (1.718)	2.03 (1.738)	2.07 (1.747)	2.13 (1.767)	2.20 (1.785)	2.27 (1.802)	2.37 (1.83)	2.47 (1.857)	2.53 (1.876)
V <sub>11</sub>	1.23 (1.494)	1.30 (1.516)	1.37 (1.537)	1.47 (1.57)	1.57 (1.602)	1.63 (1.622)	1.63 (1.622)	1.73 (1.653)	1.77 (1.663)	1.83 (1.682)	1.93 (1.712)	2.00 (1.731)
V <sub>12</sub>	2.30 (1.816)	2.33 (1.826)	2.33 (1.825)	2.43 (1.853)	2.53 (1.879)	2.47 (1.861)	2.50 (1.87)	2.57 (1.888)	2.63 (1.905)	2.73 (1.931)	2.83 (1.957)	2.90 (1.974)
V <sub>13</sub>	1.83 (1.679)	1.90 (1.699)	1.93 (1.71)	1.97 (1.72)	2.00 (1.731)	2.07 (1.75)	2.10 (1.76)	2.17 (1.779)	2.27 (1.807)	2.33 (1.825)	2.43 (1.853)	2.50 (1.871)
V <sub>14</sub>	1.73 (1.653)	1.83 (1.683)	1.90 (1.702)	2.00 (1.731)	2.03 (1.741)	2.07 (1.75)	2.20 (1.788)	2.27 (1.806)	2.33 (1.825)	2.40 (1.843)	2.50 (1.87)	2.53 (1.879)
V <sub>15</sub>	1.60 (1.612)	1.70 (1.643)	1.73 (1.653)	1.80 (1.673)	1.87 (1.692)	1.90 (1.702)	2.00 (1.731)	2.03 (1.741)	2.10 (1.76)	2.17 (1.779)	2.27 (1.807)	2.30 (1.816)
V <sub>16</sub>	1.43 (1.559)	1.50 (1.58)	1.53 (1.591)	1.53 (1.591)	1.60 (1.612)	1.63 (1.622)	1.70 (1.643)	1.80 (1.673)	1.83 (1.682)	1.90 (1.702)	2.00 (1.731)	2.10 (1.76)

**APPENDIX XXIII. continued**

Variety	Apr. '16	May '16	Jun. '16	Jul. '16	Aug. '16	Sept. '16	Oct. '16	Nov. '16	Dec. '16	Jan. '17	Feb. '17	Mar. '17
V <sub>17</sub>	1.40 (1.548)	1.53 (1.59)	1.60 (1.611)	1.60 (1.61)	1.70 (1.641)	1.73 (1.651)	1.83 (1.68)	1.90 (1.701)	1.90 (1.701)	2.00 (1.73)	2.07 (1.75)	2.13 (1.769)
V <sub>18</sub>	2.10 (1.76)	2.13 (1.769)	2.10 (1.76)	2.20 (1.788)	2.30 (1.816)	2.27 (1.807)	2.27 (1.807)	2.37 (1.835)	2.43 (1.853)	2.50 (1.87)	2.60 (1.897)	2.80 (1.949)
V <sub>19</sub>	2.33 (1.823)	2.37 (1.832)	2.40 (1.841)	2.47 (1.859)	2.53 (1.876)	2.53 (1.876)	2.53 (1.876)	2.57 (1.885)	2.67 (1.91)	2.73 (1.927)	2.83 (1.953)	2.90 (1.97)
V <sub>20</sub>	2.47 (1.862)	2.60 (1.897)	2.70 (1.923)	2.77 (1.94)	2.87 (1.965)	2.80 (1.948)	2.80 (1.948)	2.83 (1.956)	2.90 (1.973)	3.00 (1.998)	3.10 (2.023)	3.17 (2.04)
V <sub>21</sub>	2.47 (1.861)	2.47 (1.861)	2.53 (1.878)	2.63 (1.905)	2.70 (1.922)	2.77 (1.939)	2.80 (1.948)	2.87 (1.965)	2.93 (1.982)	3.03 (2.007)	3.10 (2.024)	3.20 (2.049)
V <sub>22</sub>	2.60 (1.897)	2.63 (1.905)	2.73 (1.931)	2.83 (1.957)	2.90 (1.974)	2.90 (1.974)	3.00 (1.999)	3.03 (2.007)	3.13 (2.032)	3.23 (2.057)	3.27 (2.064)	3.33 (2.081)
V <sub>23</sub>	2.73 (1.931)	2.83 (1.957)	2.90 (1.974)	3.00 (1.999)	3.00 (1.999)	3.07 (2.016)	3.10 (2.024)	3.17 (2.041)	3.23 (2.057)	3.27 (2.065)	3.30 (2.073)	3.33 (2.081)
V <sub>24</sub>	2.70 (1.914)	2.77 (1.932)	2.83 (1.958)	2.93 (1.983)	2.97 (2.008)	3.03 (2.016)	3.10 (2.041)	3.13 (2.049)	3.23 (2.065)	3.30 (2.089)	3.33 (2.097)	3.40 (2.113)
V <sub>25</sub>	2.70 (1.932)	2.77 (1.949)	2.87 (1.966)	2.97 (1.992)	3.13 (2.016)	3.20 (2.041)	3.30 (2.057)	3.33 (2.065)	3.37 (2.081)	3.47 (2.097)	3.50 (2.105)	3.57 (2.121)
V <sub>26</sub>	2.37 (1.834)	2.47 (1.861)	2.47 (1.86)	2.57 (1.887)	2.70 (1.921)	2.63 (1.903)	2.63 (1.903)	2.73 (1.929)	2.77 (1.938)	2.87 (1.964)	2.97 (1.989)	3.00 (1.998)
V <sub>27</sub>	2.20 (1.788)	2.20 (1.788)	2.27 (1.806)	2.33 (1.824)	2.37 (1.833)	2.40 (1.843)	2.43 (1.851)	2.47 (1.86)	2.57 (1.887)	2.63 (1.904)	2.73 (1.93)	2.83 (1.956)
V <sub>28</sub>	2.33 (1.823)	2.40 (1.84)	2.43 (1.849)	2.53 (1.876)	2.57 (1.885)	2.67 (1.912)	2.70 (1.92)	2.73 (1.93)	2.83 (1.955)	2.87 (1.964)	2.90 (1.973)	3.13 (2.033)
V <sub>29</sub>	2.20 (1.788)	2.27 (1.807)	2.30 (1.816)	2.40 (1.843)	2.47 (1.861)	2.43 (1.852)	2.50 (1.87)	2.57 (1.888)	2.63 (1.905)	2.73 (1.931)	2.83 (1.957)	2.87 (1.965)
V <sub>30</sub>	1.73 (1.653)	1.80 (1.673)	1.87 (1.693)	1.93 (1.712)	1.93 (1.712)	2.00 (1.732)	2.10 (1.76)	2.13 (1.77)	2.20 (1.789)	2.23 (1.798)	2.30 (1.816)	2.33 (1.826)
C.D.	0.126	0.128	0.124	0.132	0.132	0.131	0.135	0.133	0.132	0.137	0.134	0.124
SE(m)	0.044	0.045	0.044	0.047	0.046	0.046	0.047	0.047	0.047	0.048	0.047	0.044
C.V.	4.463	4.495	4.308	4.544	4.489	4.428	4.528	4.452	4.367	4.477	4.316	3.965

Note: The data in parenthesis indicate square root transformed values

**APPENDIX XXIV. Girth of roots of *Ascocentrum* hybrids/varieties during the period 2017-18, cm**

Variety	Apr. '17	May '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>1</sub>	2.20 (1.789)	2.27 (1.807)	2.37 (1.835)	2.40 (1.844)	2.43 (1.853)	2.43 (1.853)	2.47 (1.862)	2.47 (1.862)	2.47 (1.862)	2.47 (1.862)	2.53 (1.879)	2.57 (1.887)
V <sub>2</sub>	2.43 (1.853)	2.47 (1.862)	2.53 (1.88)	2.60 (1.897)	2.63 (1.906)	2.63 (1.906)	2.67 (1.915)	2.70 (1.923)	2.73 (1.932)	2.77 (1.94)	2.77 (1.94)	2.77 (1.94)
V <sub>3</sub>	2.73 (1.932)	2.80 (1.949)	2.87 (1.966)	2.93 (1.983)	3.00 (1.999)	3.03 (2.008)	3.10 (2.024)	3.13 (2.033)	3.17 (2.041)	3.17 (2.041)	3.17 (2.041)	3.17 (2.041)
V <sub>4</sub>	2.33 (1.825)	2.43 (1.852)	2.67 (1.914)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)
V <sub>5</sub>	2.73 (1.931)	2.77 (1.94)	2.83 (1.957)	2.87 (1.965)	2.93 (1.982)	2.97 (1.99)	3.00 (1.999)	3.03 (2.007)	3.03 (2.007)	3.03 (2.007)	3.03 (2.007)	3.03 (2.007)
V <sub>6</sub>	2.47 (1.862)	2.57 (1.888)	2.77 (1.941)	2.80 (1.949)	2.80 (1.949)	2.80 (1.949)	2.83 (1.957)	2.83 (1.957)	2.83 (1.957)	2.83 (1.957)	2.83 (1.957)	2.83 (1.957)
V <sub>7</sub>	2.67 (1.914)	2.67 (1.914)	2.87 (1.966)	2.90 (1.975)	2.93 (1.983)	2.93 (1.983)	2.93 (1.983)	2.97 (1.991)	3.00 (1.999)	3.03 (2.007)	3.07 (2.015)	3.10 (2.023)
V <sub>8</sub>	2.43 (1.851)	2.47 (1.86)	2.60 (1.896)	2.70 (1.922)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)	2.77 (1.94)	2.77 (1.94)	2.77 (1.94)	2.77 (1.94)	2.77 (1.94)
V <sub>9</sub>	1.63 (1.623)	1.70 (1.643)	1.77 (1.663)	1.77 (1.663)	1.77 (1.663)	1.83 (1.683)	1.87 (1.693)	1.90 (1.703)	1.93 (1.713)	1.93 (1.713)	1.93 (1.713)	1.93 (1.713)
V <sub>10</sub>	2.57 (1.885)	2.53 (1.877)	2.63 (1.904)	2.67 (1.913)	2.70 (1.922)	2.70 (1.922)	2.70 (1.922)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)	2.73 (1.931)
V <sub>11</sub>	2.00 (1.731)	2.03 (1.74)	2.07 (1.75)	2.13 (1.769)	2.13 (1.769)	2.13 (1.769)	2.13 (1.769)	2.17 (1.779)	2.20 (1.788)	2.23 (1.797)	2.23 (1.797)	2.23 (1.797)
V <sub>12</sub>	3.00 (1.999)	3.00 (1.999)	3.07 (2.016)	3.10 (2.024)	3.17 (2.041)	3.20 (2.049)	3.23 (2.057)	3.27 (2.066)	3.30 (2.074)	3.30 (2.074)	3.30 (2.074)	3.33 (2.082)
V <sub>13</sub>	2.57 (1.888)	2.60 (1.897)	2.67 (1.915)	2.67 (1.915)	2.70 (1.923)	2.70 (1.923)	2.70 (1.923)	2.70 (1.923)	2.70 (1.923)	2.70 (1.923)	2.70 (1.923)	2.70 (1.923)
V <sub>14</sub>	2.57 (1.888)	2.60 (1.897)	2.80 (1.949)	2.90 (1.975)	2.93 (1.983)	2.93 (1.983)	2.93 (1.983)	2.97 (1.991)	3.00 (1.999)	3.03 (2.007)	3.07 (2.015)	3.10 (2.023)
V <sub>15</sub>	2.33 (1.825)	2.43 (1.852)	2.67 (1.914)	2.87 (1.964)	2.90 (1.974)	2.90 (1.974)	2.90 (1.974)	2.97 (1.991)	3.03 (2.008)	3.07 (2.016)	3.10 (2.025)	3.10 (2.025)
V <sub>16</sub>	2.13 (1.77)	2.20 (1.788)	2.47 (1.86)	2.53 (1.878)	2.63 (1.905)	2.63 (1.905)	2.63 (1.905)	2.70 (1.923)	2.77 (1.941)	2.80 (1.949)	2.83 (1.957)	2.87 (1.966)



**APPENDIX XXIV. continued**

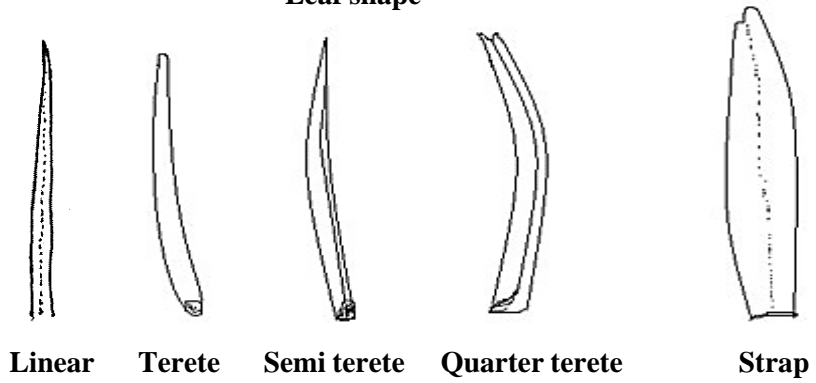
Variety	Apr. '17	May. '17	Jun. '17	Jul. '17	Aug. '17	Sept. '17	Oct. '17	Nov. '17	Dec. '17	Jan. '18	Feb. '18	Mar. '18
V <sub>17</sub>	2.17 (1.779)	2.20 (1.788)	2.30 (1.816)	2.37 (1.835)	2.47 (1.862)	2.47 (1.862)	2.50 (1.871)	2.53 (1.88)	2.57 (1.889)	2.57 (1.889)	2.57 (1.889)	2.57 (1.889)
V <sub>18</sub>	2.90 (1.975)	2.90 (1.975)	2.97 (1.991)	3.00 (2)	3.07 (2.016)	3.10 (2.025)	3.13 (2.033)	3.17 (2.041)	3.20 (2.049)	3.20 (2.049)	3.20 (2.049)	3.23 (2.057)
V <sub>19</sub>	2.97 (1.987)	3.00 (1.996)	3.03 (2.005)	3.03 (2.005)	3.07 (2.013)	3.10 (2.022)	3.13 (2.03)	3.17 (2.038)	3.20 (2.046)	3.20 (2.046)	3.20 (2.046)	3.23 (2.054)
V <sub>20</sub>	3.17 (2.04)	3.20 (2.048)	3.30 (2.072)	3.40 (2.096)	3.50 (2.12)	3.60 (2.144)	3.67 (2.158)	3.67 (2.158)	3.73 (2.174)	3.80 (2.189)	3.90 (2.212)	3.93 (2.219)
V <sub>21</sub>	3.23 (2.057)	3.23 (2.057)	3.30 (2.073)	3.43 (2.105)	3.47 (2.113)	3.47 (2.113)	3.50 (2.121)	3.53 (2.129)	3.57 (2.137)	3.63 (2.152)	3.67 (2.16)	3.67 (2.16)
V <sub>22</sub>	3.33 (2.081)	3.40 (2.096)	3.43 (2.105)	3.50 (2.12)	3.53 (2.128)	3.53 (2.128)	3.57 (2.136)	3.60 (2.144)	3.60 (2.144)	3.60 (2.144)	3.60 (2.144)	3.60 (2.144)
V <sub>23</sub>	3.37 (2.089)	3.37 (2.089)	3.43 (2.105)	3.43 (2.105)	3.43 (2.105)	3.43 (2.105)	3.50 (2.121)	3.53 (2.129)	3.53 (2.129)	3.53 (2.129)	3.53 (2.129)	3.53 (2.129)
V <sub>24</sub>	3.40 (2.113)	3.47 (2.136)	3.50 (2.136)	3.57 (2.16)	3.60 (2.167)	3.60 (2.167)	3.60 (2.175)	3.67 (2.183)	3.67 (2.183)	3.67 (2.183)	3.67 (2.183)	3.67 (2.183)
V <sub>25</sub>	3.57 (2.121)	3.67 (2.136)	3.67 (2.144)	3.77 (2.16)	3.80 (2.167)	3.80 (2.167)	3.87 (2.175)	3.87 (2.183)	3.87 (2.183)	3.87 (2.183)	3.87 (2.183)	3.87 (2.183)
V <sub>26</sub>	3.07 (2.015)	3.10 (2.023)	3.20 (2.047)	3.27 (2.063)	3.37 (2.087)	3.43 (2.103)	3.53 (2.126)	3.57 (2.134)	3.60 (2.143)	3.63 (2.15)	3.67 (2.157)	3.73 (2.172)
V <sub>27</sub>	2.90 (1.973)	2.93 (1.982)	3.00 (1.999)	3.03 (2.007)	3.10 (2.023)	3.13 (2.031)	3.13 (2.031)	3.13 (2.031)	3.17 (2.039)	3.20 (2.046)	3.23 (2.054)	3.23 (2.054)
V <sub>28</sub>	3.17 (2.041)	3.20 (2.049)	3.23 (2.057)	3.27 (2.065)	3.27 (2.065)	3.27 (2.065)	3.27 (2.065)	3.33 (2.081)	3.33 (2.081)	3.33 (2.081)	3.33 (2.081)	3.33 (2.081)
V <sub>29</sub>	2.93 (1.982)	2.90 (1.974)	2.93 (1.982)	2.93 (1.982)	2.97 (1.991)	3.00 (1.999)	3.03 (2.007)	3.07 (2.016)	3.10 (2.024)	3.10 (2.024)	3.10 (2.024)	3.13 (2.032)
V <sub>30</sub>	2.37 (1.835)	2.40 (1.844)	2.60 (1.897)	2.60 (1.897)	2.60 (1.897)	2.67 (1.914)	2.70 (1.923)	2.70 (1.923)	2.73 (1.932)	2.73 (1.932)	2.73 (1.932)	2.73 (1.932)
C.D.	0.12	0.119	0.118	0.12	0.118	0.117	0.119	0.115	0.116	0.121	0.126	0.131
SE(m)	0.042	0.042	0.042	0.042	0.042	0.041	0.042	0.04	0.041	0.043	0.044	0.046
C.V.	3.806	3.754	3.675	3.724	3.637	3.588	3.655	3.503	3.533	3.669	3.813	3.978

Note: The data in parenthesis indicate square root transformed values

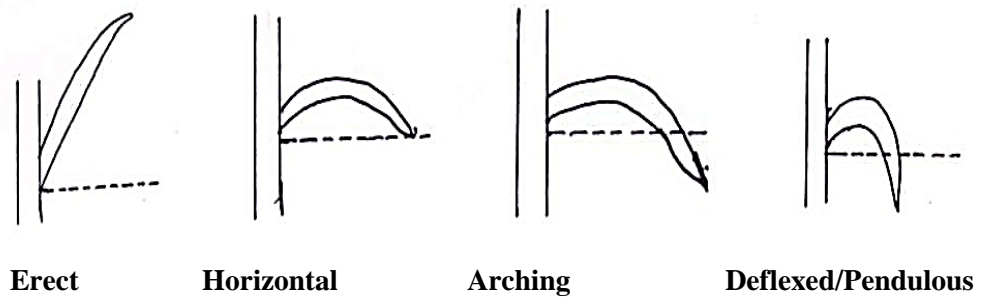
## APPENDIX XXV

### Description of leaf characters and floral morphological characters

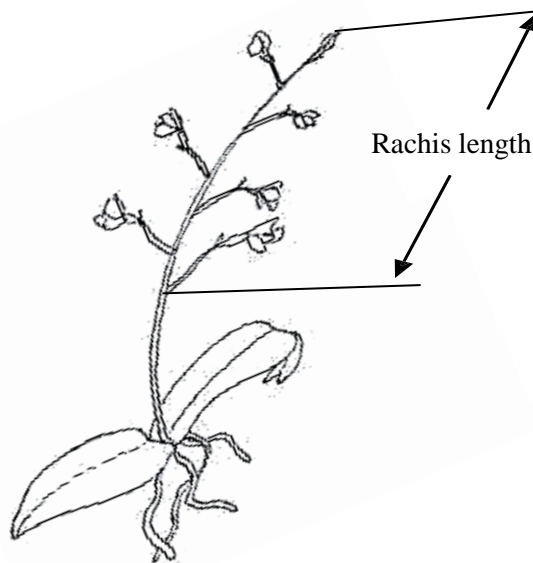
#### Leaf shape



#### Leaf orientation

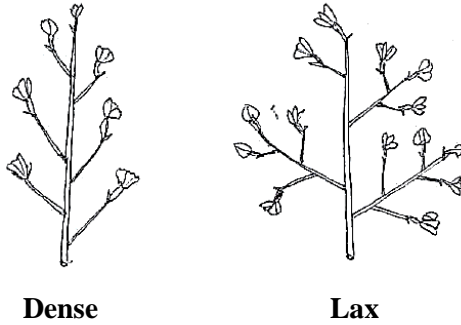


#### Rachis length

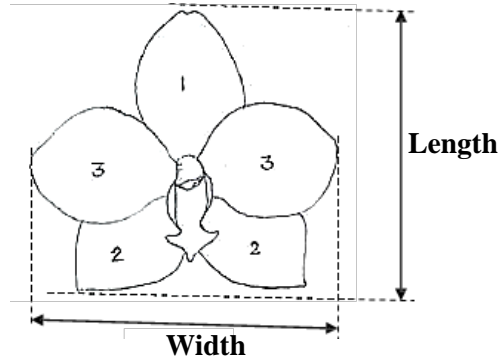


**APPENDIX XXV. continued**

**Nature of inflorescence**

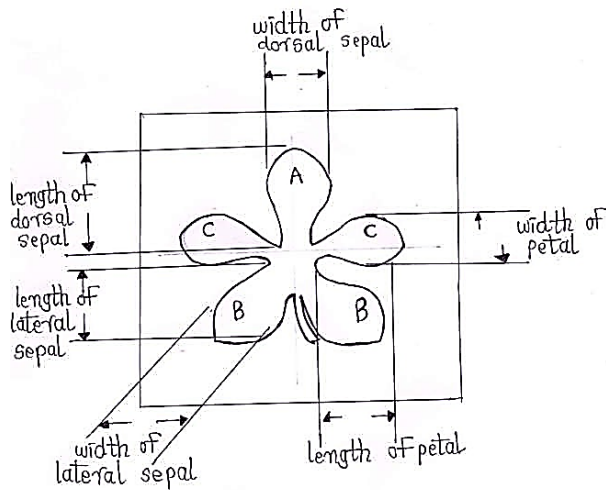


**Flower length and width in front view**



**Dorsal sepal (1), Lateral sepal (2) and Petal (3)**

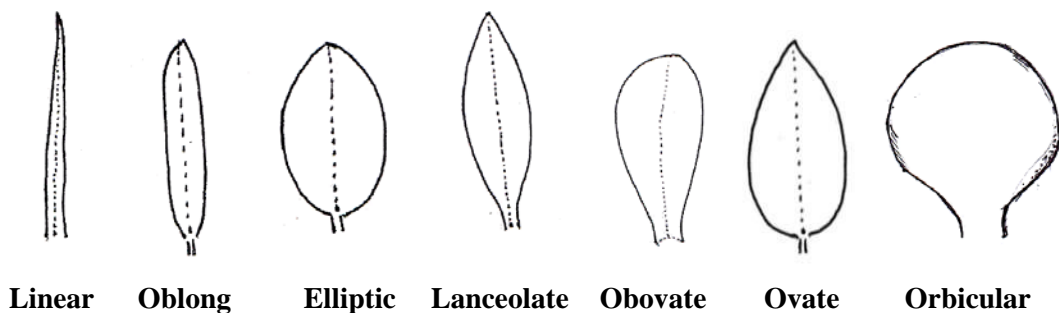
**Sepals and petals (tepals) in back view**



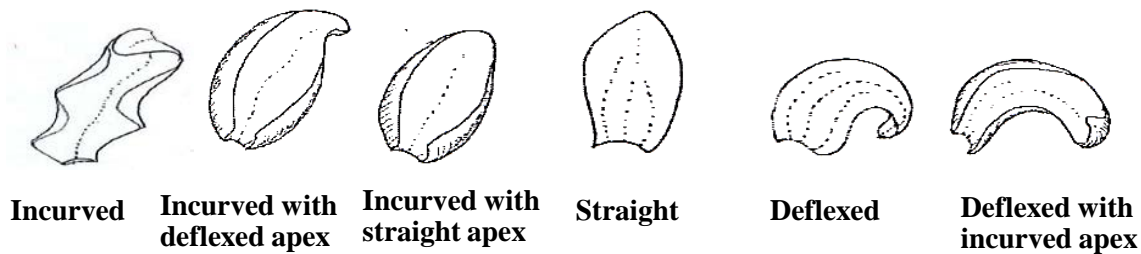
**Dorsal sepal (A), Lateral sepal (B) and Petal (C)**

APPENDIX XXV. continued

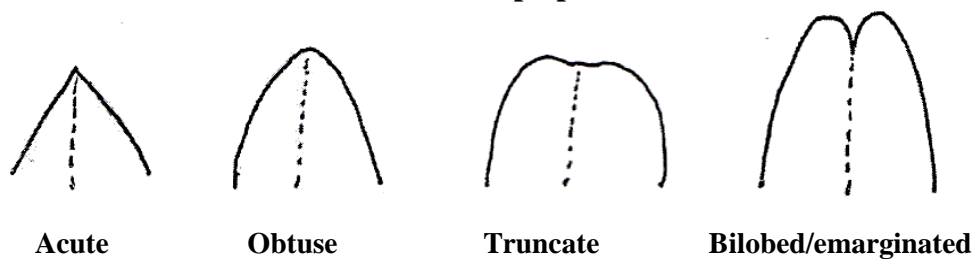
Petal/sepal shape



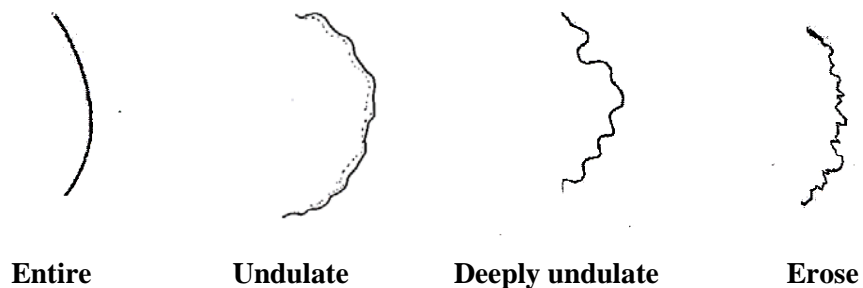
Petal/apical lip curvature



Petal/leaf/lip apex

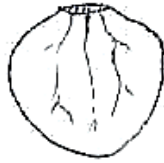


Petal/leaf margin



**APPENDIX XXV. continued**

**Lip shape at mid lobe**



**Ovate**



**Elliptic**



**Obovate**



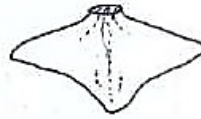
**Semi circular**



**Deltoid**



**Obdeltoid**

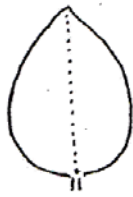


**Rhombic**



**Orbicular**

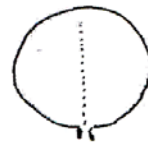
**Lip shape at apical lobe**



**Ovate**

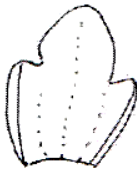


**Lanceolate**

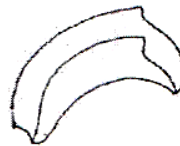


**Orbicular**

**Nature of lip at apical lobe**



**Straight**



**Deflexed with straight apex**



**Deflexed Incurved with apex**

**APPENDIX XXV. continued**

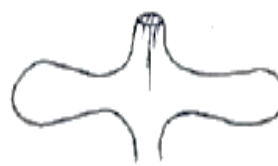
**Lip shape at lateral lobe**



**Semi circular**



**Oblong**



**Ob lanceolate**



**Obtriangular**



**Sub orbicular**

**Nature of lip**



**Slightly incurved**

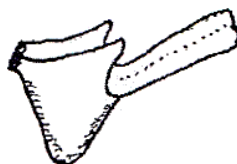


**Strongly incurved**

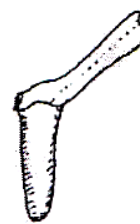
**Spur type**



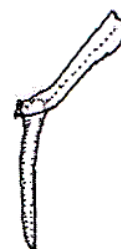
**Saccate**



**Conical**



**Cylindrical**



**Tubular**

## APPENDIX XXVI

### Monthly distribution of weather parameters during the experiment April 2016 - October 2019

Sl. No.	Month	Mean max. (°C)	Mean min. (°C)	RH I (%)	RH II (%)	Mean RH (%)	Wind speed (kmph)	Mean SS (hrs.)	Total rain (mm)	Rainy days	Evp. (mm)
1	Apr '16	35.83	26.25	85.77	55.70	70.73	2.11	7.94	0.86	0.07	4.66
2	May '16	33.97	24.20	89.68	66.35	78.02	1.85	5.88	8.69	0.29	3.77
3	Jun '16	29.81	21.64	95.17	82.97	89.07	1.27	1.64	21.82	0.73	2.13
4	Jul '16	29.95	21.62	96.13	75.39	85.76	1.45	2.30	12.68	0.65	2.50
5	Aug '16	30.38	23.25	95.48	70.55	83.02	1.87	4.92	5.92	0.61	2.89
6	Sept '16	30.30	23.56	94.90	68.73	81.82	1.78	4.82	2.87	0.33	2.93
7	Oct '16	31.48	22.69	93.48	67.77	80.63	1.02	5.49	1.20	0.13	2.79
8	Nov '16	33.03	22.17	82.63	53.70	68.17	1.89	5.82	0.46	0.03	3.03
9	Dec '16	32.37	22.32	84.58	52.19	68.39	2.94	6.47	1.71	0.10	3.33
10	Jan '17	34.09	22.89	68.74	37.45	53.10	5.34	7.59	0.00	0.00	4.69
11	Feb '17	35.99	23.19	70.14	30.75	50.45	5.02	8.68	0.00	0.00	5.65
12	Mar '17	36.13	24.68	84.81	48.48	66.65	2.23	7.42	0.43	0.03	4.48
13	Apr '17	35.72	26.04	85.10	55.23	70.17	2.07	6.48	0.64	0.03	3.87
14	May '17	34.59	24.89	87.35	60.13	73.74	1.76	5.48	5.40	0.35	3.60
15	Jun '17	30.61	23.65	94.63	77.60	86.12	1.08	1.96	21.01	0.83	2.51
16	Jul '17	30.80	22.79	94.52	74.16	84.34	1.12	2.88	12.44	0.74	2.71
17	Aug '17	30.05	23.34	95.90	78.03	86.97	1.02	3.06	15.42	0.55	2.61
18	Sept '17	31.52	22.94	94.47	73.60	84.03	0.68	4.21	13.80	0.57	2.75
19	Oct '17	31.74	22.35	92.77	70.29	81.53	0.22	4.91	5.92	0.32	2.35
20	Nov '17	33.04	21.79	86.93	58.20	72.55	1.93	6.43	1.92	0.17	2.98
21	Dec '17	32.45	21.11	78.06	48.61	63.34	5.16	7.29	0.37	0.06	3.85
22	Jan '18	33.50	20.91	68.48	37.32	52.90	5.43	8.20	0.00	0.00	4.37
23	Feb '18	35.65	22.48	62.82	30.32	46.57	5.69	9.47	0.19	0.04	5.64

**APPENDIX XXV. continued**

<b>Sl. No.</b>	<b>Month</b>	<b>Mean max. (°C)</b>	<b>Mean min. (°C).</b>	<b>RH I (%)</b>	<b>RH II (%)</b>	<b>Mean RH (%)</b>	<b>Wind speed (kmph)</b>	<b>Mean SS (hrs.)</b>	<b>Total rain (mm)</b>	<b>Rainy days</b>	<b>Evp. (mm)</b>
24	Mar '18	36.68	23.98	78.94	38.52	58.73	3.28	7.99	1.07	0.06	5.01
25	Apr '18	36.16	24.80	85.53	54.40	69.97	2.04	7.26	0.96	0.07	4.25
25	May '18	33.25	22.62	90.61	65.65	78.13	1.77	4.81	15.60	0.45	3.33
26	Jun '18	29.79	23.22	95.40	82.93	89.17	1.54	1.71	24.34	0.73	2.12
27	Jul '18	29.62	22.56	96.06	80.87	88.47	1.69	1.87	25.59	0.71	2.57
28	Aug '18	29.20	22.26	96.03	78.23	87.13	1.77	2.21	29.94	0.74	2.28
29	Sept '18	32.22	22.46	91.43	60.07	75.75	1.72	7.21	0.97	0.03	3.32
30	Oct '18	32.81	22.91	90.00	62.26	76.13	2.00	5.68	12.68	0.42	3.08
31	Nov '18	32.73	23.37	82.23	54.17	68.20	4.34	6.92	2.22	0.17	3.41
32	Dec '18	33.03	22.50	78.16	47.26	62.71	4.71	6.96	0.00	0.00	3.53
33	Jan '19	32.92	20.41	71.26	37.97	53.97	6.52	8.43	0.00	0.00	4.70
34	Feb '19	35.29	23.38	77.21	40.50	58.75	5.35	8.73	0.00	0.00	5.12
35	Mar '19	36.75	24.84	84.71	44.52	64.45	2.94	8.58	0.00	0.00	4.80
36	Apr '19	36.12	25.53	86.40	54.20	70.20	2.25	8.02	2.55	0.10	4.72
37	May '19	34.64	24.87	88.58	59.42	74.00	2.01	6.81	1.57	0.13	3.96
38	Jun '19	32.19	23.48	93.03	73.17	83.10	1.72	3.72	10.81	0.50	2.81
39	Jul '19	30.38	22.75	95.16	75.84	85.50	1.68	2.63	21.11	0.68	2.37
40	Aug '19	29.48	21.86	96.19	82.39	89.29	1.55	1.48	31.53	0.77	1.90
41	Sept '19	31.24	21.95	95.43	74.57	85.00	1.43	3.28	13.97	0.63	2.51
42	Oct '19	32.41	21.43	91.32	68.13	79.73	1.75	5.49	13.50	0.52	2.71



## APPENDIX XXVII

### APPENDIX XXVII. List of laboratory equipment used for the study

Sl No.	Equipment	Stage used	Company
1	Vortexer	DNA isolation	GeNei <sup>TM</sup>
2	High speed refrigerated centrifuge	DNA isolation	Eppendorf 5804 R
3	Nanodrop <sup>R</sup> spectopotometer ND-1000	Qualitative assessment of nucleic acids	Jenway- Genova Nano
4	Laminar Air Flow Cabinet	Preparation of PCR reaction mixture	Rotek, B&C
5	Eppendorf Master Gradient PCR	Polymerase chain reaction	Applied Biosystems
6	Electrophoresis unit	Agarose Gel Electrophoresis (AGE)	GeNei
7	Gel Doc <sup>TM</sup> XR+	Gel documentation	BIO-RAD
8	Ultra low temperature freezer	Storage of DNA samples	Eppendorf 5804 R
9	Plastic wares	Micro Tips & tubes for pipetting and sample collection	Tarson India Ltd.

**MORPHO-MOLECULAR CHARACTERISATION OF  
INTERGENERIC HYBRIDS OF *Ascocentrum***

*by*  
**KATARE RENUKA SHAMRAO**  
**(2014-22-106)**

**ABSTRACT OF THE THESIS**  
**Submitted in partial fulfilment of the**  
**requirements for the degree of**

**Doctor of Philosophy in Horticulture**

**Faculty of Agriculture**  
**Kerala Agricultural University**



**DEPARTMENT OF FLORICULTURE AND LANDSCAPING**  
**COLLEGE OF HORTICULTURE**  
**VELLANIKKARA, THRISSUR – 680 656**  
**KERALA, INDIA**

**2019**

## ABSTRACT

Morpho-molecular characterisation intergeneric hybrids of *Ascocentrum* was conducted at the Department of Floriculture and landscaping, College of Horticulture, Vellanikkara, during 2016-19 with the objective of characterising based on morphological and molecular analysis for commercial exploitation and compatibility assessment.

Thirty varieties selected for the morphological characterisation. In quantitative characters *Mok. Omayai* Yellow showed highest plant height, internodal length and leaf breadth throughout the study period. Plant spread, leaf length and leaf area were highest in *Kag. Youthong Beauty. Mok. Sayan × Ascda. Doung Porn* was observed with highest shoot girth, shoot diameter and number of leaves. *Ascda. Yip Sum Wah × V. JVB* showed maximum number of roots, *Mok. Khaw Phiak Suan × Ascda. Jiraprapa* was observed with the highest root length and *Mok. Chao Praya Sunset Yellow Spot* with the highest root girth.

*Vasco. Aroonsri Beauty* had the least values in all vegetative characters throughout the study period including plant height, spread, internodal length, shoot, leaf and root characters except the number of leaves which was the least in *Mok. Khaw Phiak Suan × Ascda. Jiraprapa. Variety Vasco. Aroonsri Beauty* also produced the maximum number of florets/spike and had the shortest internodal length.

Cluster analysis with 14 different floral characters revealed 12 clusters at 75 per cent similarity. The highest inter-cluster distance was observed in cluster 6 and cluster 10. Cluster 6, which included *Ascda. Sirichi Fragrance* and *Vasco Blue Bay White* was found to have the lowest internodal length with the highest value

for number of florets per spike, also observed to have lower flower length and flower width. Cluster 10 was found to have the high mean values for spike length, flower length and flower width.

In qualitative characters, based on growth habit, two types were found among the varieties *viz.*, hanging and prostrate nature of growth. Leaf texture was found smooth and rigid with entire leaf margin. Leaf apex was acute in *Vasco*. Aroonsri Beauty and *Ascda*. Yip Sum Wah  $\times$  V. JVB and was emarginated in rest of the hybrids. Wide variation was found among the flower colour, colour pattern, nature of petals and lip.

Regarding post harvest traits, variety *Mok*. Omayaiy Yellow recorded highest fresh weight of spike and physiological loss in weight. Whereas, variety *Kag*. Youthong Beauty took maximum days to start wilting of a floret. *Mok*. Chark Kuan Pink was observed to have longest vase life, spike longevity and highest water uptake.

In visual evaluation the highest total mean score for the spike to use as a cut flower was observed in *Mok*. Omayaiy Yellow (54.6 out of 60), while the lowest was observed in *Vasco*. Blue Bay White. The highest mean score was obtained in *Vasco*. Pine River Pink (53.83) for plants for the indoor display.

In pollen studies *Ascda*. Sirichi Fragrance and *Mok*. Sayan  $\times$  *Ascda*. Doung Porn showed the highest pollen fertility and germination and these were selected as two male parents for further cross compatibility check. *Vasco*. Aroonsri Beauty and *Kag*. Youthong Beauty were found self-incompatible as well as cross incompatible with both the male parents and *Vasco*. Pine River Blue was found cross incompatible with *Mok*. Sayan.  $\times$  *Ascda*. Doung Porn whereas, the rest of the varieties were found cross-compatible with both the male parents.

Among the 21 SSR primers, ten generated polymorphic patterns. The number of amplicons detected varied from two to seven. The highest number of alleles was found in FJ539054, FJ539061 and JN375718. Primers DQ494847 (3) observed to have less number of amplicons. The PIC value ranged from 0.095 to 0.800.. One unique band was produced by JN375713 and FJ539050 primers in *Kag. Samrong* and *Vasco. Aroonsri Beauty*, respectively. The least Jaccard's similarity value (0.05) was observed between *Kag. Samrong* and *Ascda. Suksamran Sunlight*, *Ascda. Yip Sum Wah* × *V. Josephine Van Brero*, *Vasco. Aroonsri Beauty*. The UPGMA clustering algorithm grouped the varieties into two main clusters. The variety *Kag. Samrong* clustered separately from all other members, whereas, other members were grouped in one cluster. At 50 per cent level of similarity, the hybrids grouped into 13 clusters.

Out of 29 ISSR primers used, 20 showed amplification in all hybrids with polymorphic bands. ISSR primer (GACAC) 4 generated 11 amplicons, whereas, ISSR 901 generated 31 amplicons. ISSR primer (GACAC) 4 had lowest PIC value, and UBC810, the highest PIC value (0.926). The least Jaccard's similarity value (0.03) was observed between *Vasco. Blue Bay White* and *Mok. Khaw Phiak Suan* × *Ascda. Bicentennial Yellow Spot*, which indicates that these hybrids are dissimilar to each other. The highest Jaccard's similarity value was observed between *Mok. Khaw Phiak Suan* × *Ascda. Bicentennial Yellow Spot* and *Mok. Chao Praya Sunset Yellow Spot*. A UPGMA-based dendrogram separated the 20 hybrids of *Ascocentrum* orchids into two main clusters, each with 10 members. At 30 per cent level of similarity, all the 20 hybrids grouped into 14 different clusters. Six clusters were observed with two members each.