PROTOCOL DEVELOPMENT FOR GEL STABILIZATION AND NUTRACEUTICALS IN *Aloe vera* (L.) Burm. f.

MAHESWARI R. S. NAIR

(2015-22-004)

DEPARTMENT OF PLANTATION CROPS AND SPICES

COLLEGE OF AGRICULTURE

VELLAYANI, THIRUVANANTHAPURAM – 695 522

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by

MAHESWARI R.S. NAIR

(2015-22-004)

THESIS

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2021

DECLARATION

I, hereby declare that this thesis entitled "**PROTOCOL DEVELOPMENT FOR GEL STABILIZATION AND NUTRACEUTICALS IN *Aloe vera* (L.) Burm. f.**" 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.

Maheswari RS Nair

Maheswari R. S. Nair

(2015-22-004)

Vellayani

Date: 1 3 2021

CERTIFICATE

Certified that this thesis entitled **"PROTOCOL DEVELOPMENT FOR GEL STABILIZATION AND NUTRACEUTICALS IN *Aloe vera* (L.) Burm. f."** is a record of research work done independently by Ms. Maheswari R.S. Nair under my guidance and supervision and that it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to her.

Place: Vellayani

Date: 1 3. 2021

Dr. Sreekala G. S.

(Major Advisor, Advisory Committee)

Assistant Professor

Department of Plantation Crops and Spices

College of Agriculture, Vellayani

CERTIFICATE

We, the undersigned members of the advisory committee of Ms. Maheswari R.S. Nair a candidate for the degree of **Doctor of Philosophy in Horticulture** with major in Plantation Crops and Spices, agree that the thesis entitled **"PROTOCOL DEVELOPMENT FOR GEL STABILIZATION AND NUTRACEUTICALS IN *Aloe vera* (L.) Burm. f."** may be submitted by Ms. Maheswari R. S. Nair in partial fulfillment of the requirement for the degree.

1/3/2021

Dr. Sreekala G. S.
(Chairperson, Advisory Committee)
Assistant Professor
Department of Plantation Crops and Spices
College of Agriculture, Vellayani

Deepa 1/3/2021

Dr. Deepa. S. Nair
(Member, Advisory Committee)
Assistant Professor and Head
Department of Plantation Crops and Spices
College of Agriculture, Vellayani

Mini 1/3/2021

Dr. Mini. C
(Member, Advisory Committee)
Professor and Head
Department of Post Harvest Technology
College of Agriculture, Vellayani

Meenakumari 1/3/2021

Dr. K. S. Meenakumari
(Member, Advisory Committee)
Professor and Head
Department of Agrl. Microbiology
College of Agriculture, Vellayani

1/3/2021

Dr. Suma Divakar
(Member, Advisory Committee)
Professor and Head
Department of Community Science
College of Agriculture, Vellayani

01/03/21

EXTERNAL EXAMINER

Dr. K. SUNIL KUMAR, ARS
Principal Scientist (Hort.)
ICAR- Central Tuber Crops
Research Institute
Sreekariyam - 695 017

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Maheswari R.S Nair

Dedicated to my dear family and farming community

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LIST OF ABBREVIATIONS

abs	Absorbance
ANOVA	Analysis of variance
AOAC	Association of Official Agricultural Chemists
APG	Angiosperm Phylogeny Group
AVPI – 12	Protease inhibitor protein
BHT	Butylated Hydroxy Toluene
CD (0.05)	Criticl difference at 5% level
Cfu	Colony forming unit
cfu/ml	Colony forming unit/milliliter
cfu/g	Colony forming unit/gram
Cm	Centimeter
Corp.	Corporation
cP	Centipoises
CRD	Completely Randomized Design
^{0}C	Degree celsius
Da	Dalton
DPPH	2,2 – Diphenyl – 1 – picryl hydrazyl

E. coli	*Escherichia coli*
EDTA	Ethylenediaminetetraacetic acid
et al.	Co – workers/ Co – authors
FAO	Food and Agriculture Organization
Fig.	Figure
FL	Florida
g	Gram
GRAS	Government Reciept Accounting System
$HClO_4$	Per chloric acid
HIV	Human Immunodeficiency Virus
HNO_3	Nitric acid
H_2SO_4	Sulfuric acid
i.e.	That is
KAU	Kerala Agricultural University
kcal/100mg	Kilocalorie per 100 milligram
KDa	Kilo Dalton
Kw	Kruskal – wallis
L^{-1}	Per litre
MAS	Months after storage
meq/ml	Milliequivalents per millilitre
Mg	Milligram

MHC I	MHC class I molecule
Min	Minutes
Mm	Millimeter
mmole/kg	Millimole per kilogram
MPa	Millipascal
Ms	Millisiemens
MΩ	Megaohm
NaOH	Sodium hydroxide
Nm	Nanometer
NS	Non significant
OD	Optical density
PBS buffer	Phosphate – buffered saline
PDA	Photo diode array detector
pH	Power of hydrogen ion
Ppm	Parts per million
Rpm	Revolutions per minute
RTS	Ready To Serve
SEm	Standard error of mean
sp.	Species
TCA	Trichloro acetic acid
TSS	Total Soluble Solids

USA	United States of America
USDA	United States Department of Agriculture
µg	Microgram
µg/ml	Microgram/milliliter
µl/l	Microlitre/litre
µL	Microlitre
µm	Micrometer
%	Percentage
χ^2	Chi – square

INTRODUCTION

1. INTRODUCTION

Aloe vera (L.) Burm. f. (*Aloe barbadensis* Miller) known as Ghrit Kumari is a perennial, xerophytic, succulent, pea green colour plant (Surjushe *et al*. 2008). According to APG IV system (2016) the genus is placed in the family Asphodelaceae and subfamily Asphodeloideae. In recent years, *Aloe vera* has assumed an important role in the formulation of natural products both in food and cosmetic industry and has been traditionally employed for the preparation of products having many beneficial properties for human health. Aloe gel, comprising parenchymatic tissue, contains over 98 - 99 per cent water. More than 60 per cent of its dry matter is made up of polysaccharides.

Aloe vera industry is flourishing and the gel is used in many products such as fresh gel, juice and other formulations for health, medicinal and cosmetic purpose (Eshun and He, 2004). Aloe is used as an ingredient of functional foods and in the development of health drinks and beverages like tea (Singh *et al*., 2009).

According to Hamman (2008), the stability of aloe gel is strongly influenced by air, light, temperature and microbes. If the gel is not handled properly for 24-36 hours after extraction, the viscosity of aloe gel decreases drastically close to the viscosity of water (Suriati, 2018). Fresh aloe gel degrades very rapidly at room temperature or when exposed to air. Decomposition of the gel matrix starts just after harvesting due to the action of enzymes and of bacteria normally present in leaves, resulting in loss of biological activity. In order to preserve its components and to ensure the quality, it must be processed soon after harvesting, through a process called "stabilization".

Aloe vera gel stabilization can be brought about by thermal and chemical processing that can produce irreversible changes in its physiological, pharmacological and nutritional characteristics. The degradation of gel on storage can be prevented by drying the gel or by adding suitable preservatives and antioxidants or an addition of algal sulphated polysaccharides (Yaron, 1993). The gel stabilization process in *Aloe vera* involves heated gel or admixing an antioxidant and adjusting the gel pH to a range of 3 - 3.5 followed by cooling (Rezaei *et al.,* 2003). Freeze drying technique for gel stabilization is comparatively expensive and is limited to small throughputs.

Consumers are concerned about the safety of gel containing synthetic preservatives. Synthetic preservatives could be substituted by natural antibacterial compounds such as extracts of spices and herbs for gel preservation. Spices and herbs have been added to foods since ancient times, not only as flavoring agents, but also as folk medicine and food preservatives. In addition to imparting characteristic flavours, certain spices and herbs prolong the storage life of foods through their antioxidant and bacteriostatic or bactericidal activities. Therefore, to preserve the pharmacological and nutritional properties of the *Aloe vera* gel, extracts of medicinal herbs and spices containing high levels of phenolics and strong antibacterial activity can be used. In Kerala, one of the major problems hindering commercial cultivation of aloe is lack of a proper marketing system. However, the fast expanding *Aloe vera* industry urgently needs reliable testing protocols to assess the quality and quantity of bioactive chemicals present in the final products (Bozzi *et al*. 2007). Looking to the importance of biologically active components possessed by the leaves of the *Aloe vera* plant and its wide spread use, it has become imperative that, the leaf should be processed with the aim of preserving essential bioactive components (Chandegara and Varshney, 2013).

Standardization of an appropriate low cost processing technique from this highly perishable commodity may encourage many innovative, progressive growers and entrepreneurs to take up its cultivation. This would enable to evolve new marketing strategies involving the manufacture and distribution of nutraceuticals and cosmetic preparations. In this background, the present study on “Protocol development for gel stabilization and nutraceuticals in *Aloe vera* (L.) Burm. f” has been proposed with the following objectives.

- To formulate a low cost stabilization technique for aloe gel using herbal extracts and aromatic oils.
- Development of protocols for the preparation of dried latex and marketable nutraceuticals using aloe gel.

REVIEW OF LITERATURE

2. REVIEW OF LITERATURE

Aloe vera, widely distributed in southern parts of North America, Europe and Asia (Waller *et al*. 1978) is native to the North African and the Arabian Peninsula (Ferreira *et al*., 2007).

Aloe vera is more popular all over the world because it propagates itself faster than any other known species of *Aloe* (Anselm, 2004). *Aloe vera*, a miraculous plant was used by mankind from centuries (Park and Lee, 2006). *Aloe vera* plant is the source of two products, gel and latex, both obtained from fleshy leaves. *Aloe vera* leaves are formed by a thick epidermis covered with cuticle surrounding the mesophyll, which are differentiated into chlorenchyma cells and thinner walled cells of parenchyma (fillet). The parenchyma cells contain a transparent mucilaginous jelly which is referred to as *Aloe vera* gel (Ramachandra and Rao, 2008). The latex, bitter yellow exudate originating from the bundle sheath cells is used for its purgative effects. The name *Aloe vera* was derived from the Arabic word "Alloeh" meaning "shining bitter substance," while "vera" in Latin meaning "true" or "genuine". The Egyptians called it as "the plant of immortality and the Greek scientists regarded it as the universal panacea (Himesh *et al*., 2011).

The investigation on "Protocol development for gel stabilization and nutraceuticals in *Aloe vera* (L.) Burm. f." was taken up with the objective to formulate a low cost stabilization technique for aloe gel using herbal extracts and aromatic oils, protocol development for preparation of dried latex and marketable nutraceuticals using aloe gel. The following literature provides a brief review of the research carried out in this area and the related aspects. Wherever information is lacking pertinent literature on other crops has been reviewed.

2.1 COMPONENTS OF *Aloe vera*

Waller *et al*. (1978) reported higher proportion of mannose (0.0394 mmole/kg) sugar in aloe gel which was the main component for biological activity. Femenia *et al*. (1999) reported that *Aloe vera* plant contains major portion of water and seventy five different ingredients including vitamins, minerals, enzymes, sugars, anthroquinones or phenolic compounds, lignin, tannic acids, polysaccharide, glycoprotein, saponins, sterols, aminoacids and salicylic acids.

The aloe gel contains many vitamins including vitamin A, C, E, B_1, B_2, niacin, choline and folic acid (Lawless and Allen, 2000). The aloe gel, the parenchymatic tissue of aloe leaves, comprising 98 – 99 per cent water and more than 60 percent of its dry matter is made up of polysaccharides (Segovia *et al*., 2009). According to Lanjhiyana *et al*. (2011), the aloe gel consists of a range of compounds which includes water soluble and fat soluble vitamins, minerals, enzymes, mono and polysaccharides, sugar, lignin, phenolic compounds and organic acids. Bhatnagar (2016) reported that *Aloe vera* contains 75 potentially active constituents including vitamins, enzymes, minerals, sugars, lignin, saponin, salicylic acids and amino acids.

The bright green gelatinous delicate leaves of *Aloe vera* contain a very small quantity of viscous yellow fluid known as aloe latex, which is embedded in the pericyclic cells of the vascular bundles of *Aloe vera* leaves. The main constituents of the latex are anthraquinones including the hydroxyathracene derivatives, aloin A and B, barbaloin, isobarbaloin and aloe amedin (Bradley, 1992). Chandegara *et al*. (2015) reported that, aloin, an anthraquinone compound is present in the bitter yellow sap of the middle leaf layer of *Aloe vera*, which exerts a marked laxative effect.

2.2 CHARACTERISTICS OF ALOE GEL

The viscous, pseudoplastic nature of aloe gel is mainly due to the presence of polysaccharides and is composed of a mixture of acetylated glucomannans (Gowda *et al*., 1979) which is lost shortly after extraction apparently due to enzymatic degradation (Yaron, *et al*., 1992). Fresh aloe gel degrades very rapidly at room temperature or when exposed to air and this degradation can be prevented by drying the gel or by adding suitable preservatives and antioxidants or by addition of algal sulphated polysaccharides (Yaron, 1993). Saeed *et al*. (2004)

reported that like most juices of both fruits and vegetables, aloe gel is unstable and subject to discolouration and spoilage from contamination by microorganisms. According to Ni and Tizard (2004) the pH, malic acid and conductivity are the parameters that are routinely used for the identification and evaluation of *Aloe vera* gels. Fresh aloe gel is reported to possess a pH nearing 4.7 and conductivity 2.4 mS (Waller *et al*., 2004).

Ramachandra and Rao (2008) reported that when exposed to air, the aloe gel rapidly oxidizes, decomposes and loses its biological activities. Chandegara and Varshney (2013) suggested that the aloe leaves in which base is not intact and sealed will greatly increase the microbial count. The increased microbial count reduces the biological activity.

2.3 ALOE GEL STABILIZATION

The gel stabilization process of *Aloe vera* included admixing heated gel with an antioxidant and adjusting the pH to a range of 3-3.5 followed by cooling (Rezaei *et al*., 2003). Ramachandra and Rao (2008) reported that proper filtration influences the stability of aloe juice. The unpasteurized aloe juice fortified with vitamin C and citric acid, avoided browning reactions, improved flavor and stabilized the juice. The biological activity of aloe gel remains intact when the gel is heated at 65 ^{0}C for periods less than 15 min. However higher temperatures resulted in reduced biological activity. So best method suggested for pasteurization is HTST (High Temperature Shot Time), followed by flash cooling to 5°C or below (Aloecorp, 2012). Gel stabilization and sterilization are the different processing techniques suggested to reduce the degradation (Ramachandra and Rao, 2008).

2.4 ROLE OF *Aloe vera* AS FOOD, MEDICINE AND COSMETIC

Aloe vera is the natural plant which can be used both externally and internally. The two parts of *Aloe vera* that are commonly used are bitter latex containing aloin which is a strong laxative and aloe gel. Aloe gel is used in skin care, acts as an astringent, moisturizer, humidifier and cleanser; it is ideal for sunburns, fragile skin, and for removal and repair of dead skin and cells. It cures gum diseases, relieves itching, aids healing, relieves joint and muscle pain and it is used internally as analgesic. It has anti inflammatory, wound healing, immune modulating and anti tumor activities as well as antiviral, antibacterial and antifungal properties (Manvitha and Bidya, 2014).

Rowe *et al*. (1941) reported utilization of *Aloe vera* in the treatment of X-ray burns. Benzoin tincture a compound present in *Aloe vera* was utilized for healing wounds, skin burns and constipation (Robbers *et al*., 1996). Choi and Chung (2003) reported that ethanolic extracts of *Aloe vera* showed advanced antibacterial property than aqueous extract.

The pharmaceutical properties recently discovered for both the *Aloe vera* gel and whole leaf extract is its ability to improve the bioavailability of coadministered vitamins in human subjects (Azam *et al*., 2003). The *Aloe vera* juice is widely used in production of ready to serve drink, health drink, soft drink, laxative drink, *Aloe vera* lemon juice, sherbet, aloe sports drink with electrolyte, diet drink with soluble fibers, hangover drink with B vitamin, healthy vegetable juice mix, tropical fruit juice mix *Aloe vera* yoghurts and *Aloe vera* mix for whiskey and white bread (Eshun and He, 2004). The heterogeneous composition of the *Aloe vera* pulp may contribute to the diverse pharmacological and therapeutic activities (Talmadge *et al*., 2004). The interest and use of gel has increased dramatically in the field of health care and cosmetics (Devi and Rao, 2005). Due to its absorption enhancing effects, the *Aloe vera* drug can be easily employed to deliver poorly absorbable drugs through the oral route of drug administration. The dried powder obtained from gel was successfully used to manufacture directly compressible matrix-type tablets (Loots *et al*., 2006).

Aloe vera gel is utilized in functional foods especially for the preparation of health drinks and also used in other food products including milk, ice cream, confectionery and as a flavoring component and preservative in some foods (Christaki and Paneri, 2010).

Eshun and He (2004) reported that the aloe gel has been utilized as a valuable ingredient for food application due to its biological activities and functional properties. The *Aloe vera* has been used as base material for the cosmetic and toiletry products like powders, capsule, creams, lotions, soaps, shampoos, facial cleansers, oils and other products for both external and internal uses (Hamman 2008; Haque *et al*. 2014). A health beverage was prepared from fresh *Aloe vera* leaves (Shailaja *et al*., 2018).

The dried latex from aloe leaves known as aloin is primarily used as a laxative or cathartic agents in traditional medicines (Grindlay and Reynolds, 1986). Considering the large percentage of aloin found in *Aloe vera* along with its multiple pharmacological properties, it has been considered as an important indicator for quality control in pharmaceuticals and aloe based products (Ding *et al*., 2014). The additional therapeutic properties of *Aloe vera* are the presence of the brown gel which is a natural sunscreen protectant (Bhatnagar, 2016).

2.5 NUTRIENT ANALYSIS OF *Aloe vera* GEL AND ALOE GEL CONTAINING JUICES DURING STORAGE

Parameters that are routinely used in the evaluation and identification of commercial *Aloe vera* gels are pH, malic acid, and conductivity (Ni and Tizard, 2004). The International Aloe Science Council (IASC) has presented guidelines for levels of these parameters in *Aloe vera* gels (Waller *et al.*, 2004). Wei *et al*. (2004) reported that the stability of health beverage prepared from fresh aloe leaves concluded that the stability was negatively affected by high sucrose, citric acid concentrations and low concentrations of vitamin C and sodium chloride. According to Nindo *et al*. (2010) concentration and interaction of polysaccharides, dissolved solids and processing method should be analyzed for determining the quality of the *Aloe vera* product. Dayu (2012) reported that aloe gel is a translucent slippery liquid which is light yellow to beige in color, tasteless, odorless to mildly vegetative odour which contain 0.5 per cent total solids with a minimum of 5 per cent polysaccharide content by dry weight with below acceptable level of microbial population (aerobic microorganisms and fungi > 2.0

log CFU/ml and Enterobacteriaceae > 1.0 log CFU/g). According to Chandegara and Varshney (2013) for judging the quality and purity of extracted aloe gel the quality parameters such as fiber content, viscosity, refractive index, optical density and total soluble solids plays an important role.

2.5.1 Sensory parameters

Gorloff (1983) reported that in gel preparation processes, organoleptic properties are important when the gel is intended for internal use. High quality gel appears opaque, slightly off - white in colour and is viscous (Vogler and Ernst, 1999). Color is also an indicator of changes that occur in food during storage or processing (Esteve *et al*., 2005). Decrease in sensorial profile of RTS beverage during storage was also reported (Jakhar and Pathak, 2012). Chandegara and Varshney (2013) reported that the appearance of aloe gel as clear yellow or green liquid. The colour and appearance scores showed a declining trend in treated Liquidized aloe gel juice and flavour scores were significantly higher in treated products than control throughout the study period (Jairath *et al*., 2015). Suraiti *et al*. (2019) reported time of storage, temperature and interaction has real effect on the colour of the aloe gel.

2.5.2 pH and Acidity

Bor and Jasper (1988) reported that the pH which supports the growth of most microbes ranged from 6.6 - 7.5. Lodi and Rossin, (1995) reported wide variation in pH (4.8 - 6.4) of fresh juice during storage. According to them, the conversion of malic acid into lactic acid during storage lowers the pH of fruit juices. In the case of aloe gel, malic acid is an excellent indicator of gel freshness. This acid is produced naturally in aloe leaves during Crassulacean Acid Metabolism (CAM). Under poor handling or storage conditions, in the presence of bacteria, malic acid can be broken down to form lactic acid, which might have resulted in the increase in pH during storage. According to Richard and Piere (2006) increasing pH is used as an indicator of bacterial spoilage in food (*i.e.* the food with high pH is more susceptible to microbial spoilage). Ruggeri *et al*. (2008) reported reduction in pH of fermented milks due to the persistant growth and metabolic activity of lactic acid bacteria. Yadav *et al*., (2010) reported that variations in pH during storage are due to the change in chemical properties which are affected by storage conditions. Study also reported that significant decrease in pH of the samples during storage might be due to increase in their titratable acidity and the acidity and pH are inversely proportional to each other.

Adubofuor *et al*. (2010) reported that the pH of the studied samples ranged from 3.83 - 3.90 which was within the expected range of 3 - 5 for fruits and vegetable juices. Lactic acid occurrence is an indication of gel decay (Brien *et al*., 2011). The pH of the beverage was found to decrease with increase in storage and could be correlated inversely with the acidity of product reported by Pawar *et al*. (2011). Decrease in pH with storage time, affects the organoleptic qualities of fruits blends (Awsi and Dorcus, 2012). Singh *et al*. (2012) reported that during manufacturing of lassi, containing aloe juice titrable acidity increased with increase in the level of *Aloe vera* juice from 0 to 15 per cent. pH of *Aloe vera* gel ranged from 4.55 to 4.81 as plant maturity increased from 1 to 4 years (Ahlawat *et al*., 2013). Chandegara and Varshney (2013) reported that the pH value of *Aloe vera* gel ranged from 3.5 - 4.7. Kumar *et al*. (2013) and Sasikumar (2015) reported the pH value of aloe gel as 4.4. Elbandy *et al*. (2014) reported pH of aloe gel as 4.33. Talib *et al*. (2016) reported a pH value of 4.34 in aloe gel. According to Rahman *et al*. (2017), the pH of the aloe gel remains stable due to addition of citric acid and pH of aloe gel is acidic between 4 - 5.

Increasing trend in acidity due to the formation of various organic acids in the fruits such as sulphurous acid was reported by Baramanary *et al*. (1995). Sandhu *et al*. (2001) observed increasing trends in acidity with increasing storage period. According to Rodrigo *et al.* (2003) increase in acidity indicated the start of spoilage or fermentation of the sample. Kumar *et al*. (2013) and Sasikumar (2015) conducted study about acidity of aloe juice and reported acidity of aloe juice as 1.2 percent. Singh *et al*. (2014) observed that the acidity of *bael* RTS decreased during the storage period of 6 months. Elbandy *et al*. (2014) reported an acidity of 0.10 percent and Talib *et al*. (2016) reported an acidity of 1.15 percent in aloe gel.

2.5.3 Viscosity

Viscosity is defined as the internal friction of a fluid or its tendency to resist flow (Salehi, 2020).

Viscosity increased from 31.43 to 52.70 cP suggesting an increase in concentration of aloe polysaccharides which impart pseudoplastic behavior to *Aloe vera* gel (Gowda *et al*., 1979). Gowda *et al*. (1979) also reported that when viscosity is higher, product will be better and biologically active. Chiou (2003) reported that decrease in viscosity of aloe gel was observed with increase in heating time. Ni *et al*. (2004) reported that once the gel was extracted from aloe leaves its viscous characteristics degrade rapidly. Viscosity is a parameter that reflects the fruit juice quality, it affects the taste and the ability to hold its solid portion in suspension than the product shelf life (Bravo *et al*., 2012). Dhall (2013) says that glucomannan in aloe gel is able to form a cross linking bond that affects its viscosity. Higher temperature reduced viscosity which led to decrease in biological activity of *Aloe vera* gel (Chandegara *et al*., 2015). Izadi *et al*. (2015) reported that due to increase in temperature, viscosity was reduced, when temperature reached 55 ^{0}C, the viscosity reduced to 9.2 cP and no change in viscosity was observed after this temperature in *Aloe vera* gel.

2.5.4 Refractive index

Refractive index of *Aloe vera* gel increased from1.3341 to 1.3348, with a significant increase up to three years of plant maturity (Ahlawat *et al*., 2013). Refractive index of aloe gel was reported to range from 1.3340 - 1.3355 by Aloe CORP and 1.33789 to 1.34390 by M/S Delta International. The gel with lowest refractive index is the best treatment for extraction process (Chandegara and Varshney, 2013). High refractive index indicates more impurities in the gel (Chandegara *et al*., 2015). Chandegara *et al*. (2015) observed the minimum and maximum refractive index as 1.33603 and 1.33610 in aloe gel with 1.31 ^{0}Brix and also reported that refractive index of aloe gel is near to that of distilled water, more impurities leads to higher refractive index.

2.5.5 Optical density

The optical density of 1.020 to 1.437 (abs) for *Aloe vera* leaves were reported (Wang and Strong, 1993). Optical density and anthocyanin concentration were highly correlated (Haffner *et al*., 2002). More optical density indicates the impurities in the extracted gel (Chandegara and Varshney, 2013). Optical density is the measure of transparency of liquid and also a measure of quality for *Aloe vera* gel (Chandegara *et al*., 2015). The study by Palonen and Weber (2019) reported that the optical density of raspberry fruit juice was affected by genotype, storage temperature and storage duration.

2.5.6 Aloin content

Paez *et al*. (2000) have reported presence of aloin in yellow leaf latex of *Aloe vera*. According to Ding *et al*. (2014) the aloin constitutes up to 30 per cent of leaf exudates and also reported less aloin content in dry samples due to rapid degradation by temperature. The higher aloin content was reported in fresh leaf samples with an average of 199.76 ± 0.74 mg. For dry latex, the average aloin content was 176.26 ± 0.16 mg and the aloin content in dry gel samples were 5.11 ± 0.12 mg. The results showed 17 and 20 per cent for fresh and dry exudate (Machado *et al*., 2016).

2.5.7 Specific gravity

Chandegara and Varshney (2013) reported that the specific gravity of aloe gel ranged from 1.0030 - 1.0070 according to Aloecorp and 1.0221 to 1.0339 by M/S Delta International.

Specific gravity of *Aloe vera* gel ranged from 1.0042 to 1.0064 with a significant increase up to three years of plant maturity which may be attributed to an increase in the dissolved solid content of gel (Ahlawat *et al*., 2013).

2.5.8 Total solids

Total solid is the amount of dry material remaining after the water is evaporated (Richard and Pierrie, 2006). According to Chandegara and Varshney (2012) total solids yield (without preservatives and additives) present in aloe leaves by hand filleting method was 0.45 - 0.65 percent and after whole leaf processing was 1.30 - 3.50 percent. Gel total solids increased from 0.68 and 1.1 percent with increasing age of *Aloe vera* up to four years implying accumulation of bio-molecules in the gel (Ahlawat *et al*., 2013). High total solid content implies that samples were able to retain fibres in the sample (Francis *et al*., 2017).

2.5.9 TSS and Carbohydrates

Wills *et al*. (1980) reported TSS content in apple juices decreased due to conversion of sugar into starch. Increase in TSS content with the increase in storage period was reported in juice of mandarin, sweet orange and lemon (Mehta and Bajaj, 1983). According to Lee and Nagy (1988) the fructose, glucose and sucrose got degraded in grapefruit juice during storage at 37 ^{0}C for 16 weeks. During storage the heat processed juices showed increase in sugar levels which might be due to the inactivation of enzymes, which may be responsible for decreasing acidity and conversion of polysaccharides into simple sugars (Ghorai and Khurdiya, 1998). Haffner *et al*. (2002) reported that the soluble solids content in raspberry genotypes varied between 7.0 and 11.7 per cent, which did not change significantly during storage.

Whimey and Rolfes (2005) reported carbohydrate content of 56.27 per cent in the proximate analysis of aloe leaves and is the highest parameter, so aloe leaves are a good source of carbohydrate readily accessible fuel for physical performance and regulate nerve tissue. The polysaccharides found in aloe gel are not stable, especially under stress conditions such as heat, and in the presence of acid and enzymatic activities; when it is the main factor contributing to health benefit (Hamman, 2008). Garg *et al*. (2008) observed the increase in reducing and total sugar content during storage in blended aonla juices.

For preservation of good juice quality, retention or minimum increase in TSS content of juice during storage is desirable (Bhardwaj and Pandey, 2011). Six months after storage, the total soluble solids of bottle gourd basil leaf juice increased from 11.3 percent to 11.5 percent showing a negligible change (Majumdar *et al*., 2011). Boghani *et al*. (2012) developed a blended *Aloe vera* aonla based ready to-serve and the TSS ranged from 12 per cent to 14.4 per cent, after storage for six months. Increase in TSS during storage of juices is because of conversion of polysaccharides and other constituents of juice into sugars (Jakhar and Pathak, 2012). In apple cultivars the increase in TSS was attributed to the breakdown of starch into sugars or the hydrolysis of cell wall polysaccharides (Jan and Rab, 2012). Kausar *et al*. (2012) reported in cucumber – melon functional drink TSS mean values increased from 15.49 - 16.09 percent during storage, while reducing sugars increased from 1.9 per cent to 2.48 per cent and non-reducing sugars decreased from 9.36 per cent to 8.70 per cent.

Barandozi and Enferadi (2012) reported the presence of glucuronic acid, galactose, glucose, mannose and xylose with various proportions in aloe juice. Boghani *et al*. (2012) and Kumar *et al.* (2013) reported TSS of aloe juice as 2.1 ^{0}Brix. Reduction in total soluble solids was noticed due to the presence of microorganisms which causes deterioration of fruit blends because of sugar fermentation (Ana *et al*., 2013). Ibironke *et al*. (2013) reported decrease in TSS content of tomato pulp during storage due to the breaking of solids. TSS of *Aloe vera* gel was 3 per cent according to Elbandy *et al*. (2014). Kilima *et al*. (2014) reported that increase in reducing sugars was probably due to the hydrolysis of polysaccharides like starch, cellulose, pectin, etc., and conversion into simple sugars (glucose, fructose) during storage. TSS of blanched and unblanched aloe gel was 2.2 ^{0}Brix and 2.0 ^{0}Brix respectively as reported by Shubra *et al*. (2014). Glucose was the most abundant monosaccharide present in the neutral polysaccharide from liquidized aloe gel juice as revealed by chromatography (Vidic *et al*., 2014). According to Talib *et al*. (2016) TSS of 1 ^{0}Brix was found in aloe gel. Talib *et al*. (2016) reported total, reducing and non-reducing sugars of aloe juice as 0.70, 0.22 and 0.48 percentage respectively.

2.5.10 Aminoacids

Aloe vera gel provides 20 of the 22 required amino acids for human and 7 of the 8 essential amino acids (Surjushe *et al*., 2008). Determination of the essential amino acids of *Aloe vera* gel was undertaken by Sugiastuti *et al*. (2012). The *Aloe* *vera* gel was hydrolyzed using 6N hydrochloric acid, to convert the protein into free amino acids. Then free amino acids were analyzed by high performance liquid chromatography (HPLC) using phenylisothiocyanate (PTIC) derivatization process. The results indicated threonine (7.4 ppm), valine (12.5 ppm), methionine (11.6 ppm), isoleucine (14 ppm), leucine (17.7 ppm), phenylalanine (13.8 ppm) and lysine (36.5 ppm). The non-essential amino acid, proline present in *Aloe vera* is a constituent of collagen, whose role is to ensure the perfect holding capacity and elasticity of epithelial tissues (Kar and Bera, 2018).

2.5.11 Vitamins

Ascorbic acid transformed to diketoglutaric acid due to reaction with air and metal ions may contribute to the losses of ascorbic acid (Harris, 1975 and Addo, 1981). Ascorbic acid is water soluble and it is readily lost *via* leaching from cut or bruised surfaces of raw material which results in chemical degradation of the product (Tannenbaum *et al*., 1985). Vitamin C begins to degrade immediately after harvest and degrades steadily during prolonged storage (Murcia *et al*., 2000).Vitamin C content of all roselle - fruit blends decreased during storage with the advancement of storage period, which was probably due to the sensitivity of vitamin C to oxygen, light and heat (Ziena, 2000). Incorporation of air into the juice during extraction, finishing and bottle filling leads to loss of ascorbic acid (Farnworth *et al*. 2001). The amount of ascorbic acid in fresh spiced fruit drinks slightly decreased with increasing amount of spices; lower than from the value for guava juice and almost similar to passion fruit juice and higher than the value for lemon juice (Suntornsuk, *et al*., 2002). Oxygen is the most destructive ingredient in juice causing degradation of vitamin C (Padayatty *et al*., 2003).

The retention of vitamin C is used as an estimate for the overall nutrient retention of food products because it is a least stable nutrient and is highly sensitive to oxidation and leach into water soluble media during storage (Franke *et al*., 2004). Majumdar *et al*. (2009) reported, 74 percent loss in vitamin C, after 6 months of storage in cucumber-litchi-lemon blended juice. Jain and Khurdiya, (2009) observed the loss in vitamin C during storage of aonla juice.

According to Inga *et al*. (2007) the vitamin C is an important parameter for assessing the nutritional quality of fruits blends, as it degrades during storage. The trend of decrease in vitamin C in storage studies was reported in developed beverage and pickle (Mishra *et al*., 2010; Puranik *et al.*, 2011). According to Pena *et al*. (2011) when vitamin C retention decreased to 50 percent of its initial amount, shelf life of the product ends. A loss in vitamin C was observed in guava blended with *Aloe vera* and roselle during 120 days of storage at ambient temperature (Kumar *et al*., 2012). Vitamin C content of *Aloe* gel was 47 - 61 mg per 100 ml (Chandegara and Varshney, 2013). Ascorbic acid content of aloe gel was 7 mg/100g as reported by Kumar *et al*. (2013) and Sasikumar (2015). According to Talib *et al*. (2016) ascorbic acid content of aloe juice was 1.54 mg/100ml.

2.5.12 Enzymes

The viscous pseudoplastic nature of *Aloe vera* gel was lost shortly after extraction due to enzymatic degradation (Gowda *et al*., 1979). Enzyme addition increased the release of various phenolics and other nutritionally important components, in the juice apart from promoting juice extraction (Markham *et al*., 2000). In the agro industrial sector, the use of enzymes led to a process optimisation, reducing process related energy costs, improving nutritional safety and quality of food, the development of new products and new applications for a number of farming products (Minnusi *et al*., 2002).

Amylases have a wide range of application in various industries such as in the food, bread making, paper industries, textiles, sweeteners, glucose and fructose syrups, fruit juices, detergents, fuel ethanol from starches, alcoholic beverages, digestive aid, and spot remover in dry cleaning and also α-amylases are being used in clinical, medicinal, and analytical chemistry (Pandey *et al*., 2000). Chandegara and Varshney (2013) reported amylase content of 1100 – 1600 units per 100ml and lipase content of 600 – 800 units per 100ml in aloe gel.

2.5.13 Protein

Protein serve as an enzymatic catalyst and third highest parameter in aloe leaves (Whimey and Rolfes, 2005). Aloe protein of 14 kDa from the *Aloe vera* leaf gel was isolated by an ion exchange chromatography using DEAE-cellulose and CM - cellulose column. This is a new protein possessing antifungal and anti inflammatory properties and thus sets a platform to be used as a medicinal plant product (Das *et al*., 2010). Protein content of 0.11g per 100g of aloe gel was reported by Chandegara and Varshney (2013). A protease inhibitor protein with the molecular mass of 11,804.931 Da was isolated from *Aloe vera* leaf gel which was designated as AVPI-12 and the isoelectric point of the protein was about 7.43. The inhibition of the fibrinogenolytic and fibrinolytic activities of plasmin by AVPI-12 suggested that the inhibitor has potential for use in antifibrinolytic treatment (Siritapetawee *et al*., 2013).

2.5.14 Calcium and Iron

Calcium content reported in *Aloe vera* drinks in Thai food composition table, as 800 milligram and iron content as 15 milligram (Chandegara and Varshney, 2012).

The process of making *Aloe vera* leaf powder was standardized and its nutritional and physico - chemical characteristics were assessed. The recovery rate of *Aloe vera* leaf powder was 3.2 percent. The powder contained substantial amounts of iron (64.8 mg/100 g) (Gautam and Awasthi, 2007).

2.5.15 Ash

Antia *et al*. (2006) reported that ash content is a reflection of mineral preserved in the sample, it represents total mineral content which is essential for proper functioning of tissues and act as second messengers in biological cascade mechanisms and important part of proximate analysis for nutrient evaluation. Ash content of 0.25 per cent was reported by Chandegara and Varshney (2013). Ash content in blanched aloe gel was 0.26 percent and unblanched aloe gel was 0.25

percent as reported by Shubhra *et al* (2014). An ash content of 0.23 percent was reported by Talib *et al*. (2016) in aloe gel.

2.5.16 Calories

According to Gautam and Awasthi (2007) *Aloe vera* leaf powder (AVLP) standardized recorded 231 kcal energy value. Chandegara and Varshney (2013) reported calorie content of 3.3/100g in aloe gel. According to USDA *Aloe vera* juice contains 131 calories out of which 3 calories come from fat**.** The calculated energy value of *Aloe vera* – aonla blended functional squash using stevioside recorded 53.65 kcal/100g (Sharma *et al*., 2018).

2.5.17 Fat

Aloe vera provides four plant steroids *viz*., cholesterol, campesterol, β-sisosterol and lupeol all of which have anti-inflammatory action and lupeol also possesses antiseptic and analgesic properties (Surjushe, *et al*., 2008). According to Chandegara and Varshney (2013) fat content in aloe gel was reported as 0.09 g per 100 g. The total fat content of (1 cup serving) *Aloe vera* Juice is 0.3 g as per USDA standards.

2.5.18 Crude fiber

The fibre content of aloe gel ranged from 0.074 to 0.088 percent of fresh weight of pulp (Wang and Strong, 1993). The crude fibre plays an important role in the composition of *Aloe vera* gel and due to presence of pectic substances, cellulose and hemicelluloses it stimulate intestinal transition (Femenia *et al*. 1999; Femenia *et al*. 2003). The effect of high hydrostatic pressure (HHP) on colour, dietary fibre, vitamin C content, polysaccharides content, physico - chemical and structural properties of *Aloe vera* gel at three pressure levels (300, 400 and 500 MPa for 3 min) after 35 days of storage at 4 ± 1 °C revealed that ash, crude fibre and carbohydrates content increased with increasing pressure (Galvez *et al*., 2014).

Chandegara and Varshney (2013) reported that fiber content is directly related to purity of gel and is the criteria of efficiency which reported crude fiber content as 0.10 percent. Shubra *et al*. (2014) reported a crude fibre content of 0.26 percent in aloe gel and the study also revealed that blanching effect had negligible effect on crude fibre content.

2.5.19 Microbial population

According to WHO microbiological standards, microbial populations in untreated samples of *Aloe* gel were higher than the acceptable limits, the acceptable limits are aerobic microorganisms and fungi > 2.0 log cfu/ml and enterobacteria > 1.0 log cfu/g (WHO, 1998). Ethanolic extracts of *Aloe vera* showed advanced antibacterial property than aqueous extracts (Choi and Chung, 2003). Ensure microbial safety to identify the undesirable quality losses and to maximize the quality improvement (Holdsworth, 2004). *Aloe vera* is reported to have antibacterial and antioxidant properties (Rashid *et al*., 2018).

The effect of high hydrostatic pressure (HHP) treatment (300, 400 and 500 MPa for 1 and 3 min at 20 ^{0}C) on the microbiological shelf life and microbiota composition of *Aloe vera* gel during 90 days of storage at 4 °C revealed aerobic mesophilic and psychrotrophic bacteria, as well as moulds and yeasts, which were enumerated after HHP treatment and through cold storage. Results showed that HHP treatment at or over 400 MPa for 3 min were effective to keep the microbial counts to undetectable levels during the whole storage period, and consequently the microbiological shelf life of *Aloe vera* gel was extended for more than 90 days at 4 ^{0}C. The microbiota in the untreated *A. vera* gel was dominated by Gram negative bacteria (mostly *Rahnella aquatilis*) and yeasts (mostly *Rhodotorula mucilaginosa*). In contrast, Gram positive bacteria tentatively identified as *Arthrobacter* spp. and *Micrococcus* or *Kocuria* spp. were the predominant microorganisms in samples pressurized at 300 MPa for 1 and 3 min, while *Bacillus megaterium* predominating in samples treated at 400 MPa for 1 min. At 400 MPa for 3 min and above, the microbial growth was completely suppressed for 90 days; however, viable spore formers were detected by enrichment (Reyes *et al*., 2012).

2.5.20 Minimum Inhibitory Concentration

The MIC of leaf extract of *Chenopodium ambroisoides* for complete inhibition of test fungus *Fusarium oxysporum* was determined by Usual Poisoned Food Technique (Grover and Moore, 1962). According to Moreira *et al*. (2005) the procedure followed to determine Minimum Inhibitory Concentration of essential oil of spices was agar diffusion assay. Mazzola *et al*. (2009) reported the determination of minimum inhibitory concentration (MIC) for every chemical agent, by using the method of successive dilution. To study and compare the behavior of selected microorganisms, the samples were subjected to minimal inhibitory concentration (MIC). The minimum inhibitory concentration (MIC) is the concentration at which an agent experiences the complete inhibition of organism growth (Vipra *et al*., 2013).

Aloe vera sap and leaves extracts were investigated using agar well diffusion technique with different concentrations to evaluate the antimicrobial activity. Both types of extract revealed antimicrobial inhibitory effect. The sap extract were more effective than the leaves extract against all test microorganisms. The highest mean diameter of zone (MDIZ) showed with sap water extract (100 μg/ml) was against *Pseudomonas aeruginosa* and *Bacillus subtilis* (47 mm). The minimum inhibitory concentration (MIC) was ≤ 6.25 μg/ml for almost all test microorganisms and different types of extracts except acetone extract from leaves which exhibited MIC ≥ 50 μg/ml (Abakar *et al*., 2017). The extracts from *Aloe vera, Aloe volkensii* and *Aloe secundiflora* inhibited growth of *Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumonia, Escherichia coli, Erwinia carotovora, Candida albicans* and *Fusarium oxysporum*. The zones of inhibition varied significantly (F = 3.424051, P = 0.005007) between the extracts of *Aloe vera, Aloe volkensii* and *Aloe secundiflora*. The minimum inhibitory concentration, minimum bactericidal concentration and minimum fungicidal concentration of the extracts varied significantly (F = 4.696456, P = 0.000534) (Waithaka *et al*., 2018).

In a comparative study of antimicrobial activity of *Aloe vera* gel and antibiotics against isolates from fast food (Shireen *et al*., 2015) obtained lower zones of inhibition which may have emanated from the type of secondary metabolites synthesized by the *Aloe* sp. (Gharibi *et al*., 2015)

2.6 STABILIZATION PROPERTIES OF HERBAL EXTRACTS AND ESSENTIAL OILS

The most active constituents (essential oils) of many spices having wide spectra of antimicrobial activity are aromatic phenolic compounds, such as thymol and carvacrol in oregano and thyme, eugenol in clove and cinnamon, and cinnamic aldehyde in cinnamon (Karapinar and Aktug, 1987; Beuchat and Golden, 1989). Researchers have reported that phenolic compounds from different plant sources could inhibit various food borne pathogens (Nychas, 1995; Smid and Gorris, 1999; Prashanth *et al*., 2001; Kim *et al*., 2005).

Tannin rich plants have antibacterial potential due to their basic character that allows them to react with proteins to form stable water soluble compounds, thereby killing the bacteria by directly damaging its cell membrane (Elmarie and Johan, 2001). Flavonoids are a major group of phenolic compounds reported for their antiviral (Chiang *et al*., 2003), spasmolytic properties (Amor *et al*., 2005) and antimicrobial (Bastos *et al*., 2009).

Liliaceae, Zingiberaceae, Theaceae and Punicaceae family have obvious synergistic effects on stabilizing aloe and potential as natural antiseptics and oxidation resistant materials (Guo *et al.*, 2004). Many plant flavonoids (e.g., epigallocatechin, catechin, myricetin, quercetin) are reported to have antimicrobial activity (Cushnie and Lamb, 2005). Polyphenols and flavanoids are usually recognized as the medicinal plant compounds which are responsible for the antioxidant activity (Gajera *et al*., 2005).

The use of synthetic antioxidants (e.g., butylated hydroxyanisole and butylated hydroxytoluene) as well as other synthetic food additives has been avoided mainly due to their potential toxic effects (Garau *et al*., 2007). Alkaloids isolated from plants were found to have antimicrobial properties (Ahmed *et al*., 2010). Thus, there is an urgent need to find natural antioxidants that may be used to prevent food spoilage. Medicinal plants could be subjected to a wide range of microbial contamination during pre and post harvest stages and can harbor high microbial loads (Bhowmik *et al*., 2016).

Western society appears to be experiencing a trend of 'green' consumerism (Silva, 1996; Smid and Gorris, 1999), which favours use of fewer synthetic food additives and products with a smaller impact on the environment. A few preservatives containing essential oils are already commercially available. 'DMC Base Natural' is a food preservative produced by DOMCA S.A., Alhendı´n, Granada, Spain and comprises 50 per cent essential oils from rosemary, sage and citrus and 50 per cent glycerol (Yepes *et al.,* 1997).

'Protecta One' and 'Protecta Two' are blended herb extracts recognized as safe (GRAS) food additives in the US produced by Bavaria Corp. Apopka, FL, USA and are classed as generally. Carbohydrates in foods do not appear to protect bacteria from the action of essential oils as much as fat and protein (Shelef *et al*., 1984). A low water and/or salt level facilitates the similar action of essential oils (Shelef *et al*., 1984; Tassou *et al*., 1995; Wendakoon and Sakaguchi, 1995; Skandamis and Nychas, 2000). Products with low moisture content can be stored at ambient temperatures for long periods of time (Vega *et al*., 2007).

However, food products are sensitive to drying temperature, which can induce degradation (Attanasio *et al*., 2004; Sablani, 2006; Luangmalawat *et al*., 2008). Antioxidative properties of essential oils and various extracts from many plants have great interest in the food industry, so possible growing trend of using as natural additives has emerged to replace synthetic antioxidants (Pittella *et al*., 2009).

In addition, the conditions and techniques used for drying can induce structural changes in the food, causing variation in its original microstructure such as physical and chemical changes which can render the product unacceptable to consumers (Attanasio *et al*., 2004). Biochemical functions of natural antioxidant extracts from vegetables, fruits, and medicinal plants are having an increase in interest among consumers because of the property to prevent oxidative damage and promoting health (Pittella *et al*., 2009).

According to Saumendu *et al.* (2010), the antibiotic and antimicrobial activities of different extracts of *Gymnema sylvestre* were determined against a number of pathogens, namely, *S. aureus*, *E. coli*, *and B. subtilis.* David and Sudarsanam (2013) reported that the antimicrobial activity of leaf extracts of *Gymnema sylvestre*, can be attributed to the presence of phytochemicals like flavonoids, terpenoids, amino acids, glycosides, tannins, amino acids and carbohydrates. The aqueous and methanol leaf extract showed significant antibacterial and antifungal activities against the selected microorganisms when compared to the standard drugs respectively.

Similar study on antimicrobial effect of ethanolic extract of *Gymnema sylvestre* against *Bacillus pumilus*, *B. subtilis*, *P. aeruginosa*, and *S. aureus* showed promising antimicrobial effect (Satdive *et al*., 2003). Tiwari *et al*. (2014) reported that the methanolic and ethanolic leaf extract of *Gymnema sylvestre* possessed antibiotic and antimicrobial activities. The methanol extract of leaves of *Gymnema sylvestre* possessed significant antioxidant activities (Behera, 2019).

Zainol *et al*. (2003) reported that naturally occurring alkaloids and synthetic derivatives in *Centella asiatica* have bactericidal effects. Pittella *et al.* (2009) indicated the presence of bioactive molecules in *Centella asiatica* and can be used as a prototype for development of new drugs and/or as a source of antioxidant and antitumor pharmaceutical raw material. It has been reported recently that *Centella asiatica* has antimicrobial ability, and the root extracts of *Centella asiatica* were more effective both as antibacterial, antifungal and even as antiyeast (Yadav and Agarwala, 2011). Nasution *et al*. (2018) indicated in study that both the leaves and root of *Centella asiatica* have bactericidal effects on pathogenic microorganisms and ethanol was referred as the best solvent to extract antimicrobial substances from this plant compared with chloroform and water.

The antifungal activity of the *Achyranthes aspera* was reported by Elumalai *et al*. (2009). The plant root of *Achyranthes aspera* was reported to possess antioxidant property and *Achyranthes aspera* stem can be used as a good source of safe and natural antioxidant compounds (Priya *et al*., 2010).

Chemical constituents like flavonoids, triterpenoids, polyphenolic compounds and steroids were responsible for antioxidant and antibacterial activities and these chemical constituents were in the methanolic extract of aerial parts of *Achyranthes aspera* (Tahiliana and Kar, 2000). The compounds responsible for antioxidant activity and antibacterial activity were revealed by the preliminary phytochemical screening of the extracts which comprised of phenolic compounds, flavonoids, alkaloids, steroids, tannins etc. and might serve as a substitute for synthetic drugs (Beaulah *et al*., 2011).

Jain *et al*. (2015) studied the antibacterial and antifungal activity of *Tridax procumbens* using petroleum ether, methanol and aqueous extracts of the plant parts and tested against bacterial strains *i.e., Bacillus subtilis* and *Escherichia coli* and fungal strains *Trichoderma reesei* and *Fusarium oxysporium*. The results showed that antibacterial and antifungal activities of methanolic extract were effective while the antibacterial activity of aqueous extract was devoid of significant activity.

The antimicrobial activity of ethanol and methanol extracts of root, stem, flower, leaf and whole plant of *T. procumbens* against *E.coli, Klebsiella pneumonia, Proteus vulgaris, B. subtilis and Staphylococcus aureus* were reported by Kumar and Naidu (2011). Bharathi *et al*. (2012) demonstrated that methanolic extract of *Tridax procumbens* stem was effective against *E.coli*.

Strong antifungal activity of *Tridax procumbens* against *Aspergillus niger* was reported by Nair *et al.* (2005) and Dangi *et al.* (2013). Jain *et al*. (2015) revealed that the antibacterial activity of *Tridax procumbens* leaves may be due to presence of various active principles.

Chang and Lin (2012) reported antioxidant activities of water, methanol and 95 per cent ethanol extracts of air dried fruit of *Terminalia chebula*. Saha and Verma (2016) reported *Terminalia chebula* fruits as potential source of natural antioxidants due to presence of rich polyphenolic and the total antioxidant capacity of the polyphenolic extract of *Terminalia chebula* fruits ranged from 0.140 to 0.387 at graded concentrations of 50–500 μg/mL.

Terminalia chebula aqueous extracts exhibited antifungal activity against a number of dermatophytes and yeasts (Ray and Majumdar, 1976; Dutta *et al*., 1998). *Terminalia chebula* exhibited antibacterial activity against more number of bacterial species (Malekzadeh *et al*., 2001). According to Michalak (2006) phenolic compounds present in *Terminalia chebula* are effective hydrogen donors, which makes them good antioxidants.

Ellagitannin (punicalagin) in pomegranate (*P. granatum*) is the active substance responsible for its antimicrobial activity (Machado *et al*., 2002). Jurenka (2008) based on the research on chemical constituents of pomegranate revealed the presence of ellagitannins (punicalagin), anthocyanins (delphinidin, cyanidin and pelargonidin 3- glucosides and 3,5-diglucosides), ellagic acid, punicic acid, flavonoids, flavonols and many other constituents which were further correlated to the antioxidant potential of the plant . The antibacterial activity of the plant was contributed by the presence of alkaloids in plant extract (Zuo *et al*., 2008).

According to Cushnie and Lamb (2005) the antibacterial activity of plant extract may be exerted due to the presence of plant secondary metabolites such as flavonoids. Akter *et al*. (2013) reported that the rind extract of the plant showed the presence of plant secondary metabolites like alkaloids, tannins, flavonoids and glycosides. Akter *et al*. (2014) in their study suggested that, rind extract of *Punica granatum* exhibited significant antimicrobial activity against a number of pathogenic bacterial strains due to the presence of the phytochemical constituents and minimum inhibitory concentration of the extracts support the antimicrobial property, rind part of *Punica granatum* plant possesses strong antimicrobial activity.

Cao *et al*. (1996) reported green tea as an excellent source of polyphenols, which are natural antioxidants that can be used as alternatives to synthetic antioxidants, since they are typically less harmful. The antioxidants present in green tea inhibit oxidation of organic molecules, and therefore are very important, not only for living systems, but also for food preservation (Masuda *et al*., 2003). Kumaran *et al*. (2009) says that the beneficial effect of green tea extract on the health is widely recognized due to its antioxidant properties. Jigisha *et al*. (2012) and Steinmann *et al*. (2013) reported that the antimicrobial effects of green tea against a variety of gram positive and gram negative bacteria (e.g. *Escherichia coli*, *Salmonella* spp., *Staphylococcus aureus*, *Enterococcus* spp.) some fungi (e.g., *Candida albicans*) and a variety of viruses (e.g., HIV, herpes simplex, influenza) . Antimicrobial studies using different green tea extracts revealed that there is a huge potential for the use of green tea in antimicrobial therapy (Reygaert, 2014).

Juliani *et al*. (2004) reported that cinnamaldehyde containing oils (non-phenolic) showed lower antimicrobial activities than eugenol containing oils. The antimicrobial activity of the essential oil of *Cinnamomum zeylanicum* has been related to its cinnamaldehyde content (Prabuseenivasan *et al*., 2006). Some of the publications deal with the mode of action of the essential oil components in combination with other natural preservatives or antibiotics (Yoon *et al*., 2011). Essential oils containing aldehydes or phenols, such as cinnamaldehyde, citral, carvacrol, eugenol or thymol as major components has the highest antibacterial activity, followed by essential oils containing terpene alcohols (Bassole and Juliani, 2012).

According to Kolesnikova *et al*. (1976) essential oils are natural plant products containing complex mixture of components and thus having multiple antimicrobial properties. In basil, the strongest antimicrobial activity was attributed to eugenol (19 per cent) and linalool (54 per cent) content and a synergistic effect of these two were observed. In addition to this, a fresh herb of basil has a smoother flavor than a dried one and many alcoholic beverages with basil have excellent sensory properties. These beverages are rich in amounts of vitamin C, thiamin and riboflavin and also have a good shelf life too due to their antimicrobial substances. The antimicrobial activity in essential oils appears due to oxygenated terpenoids, particularly phenolic terpenes, phenyl propanoids and alcohols (Bassole and Juliani, 2012).

According to Wendakoon and Sakaguchi (1995) the antibacterial activity of essential oil of clove may be attributed to eugenol, the antimicrobial compound of clove. The microbial inhibition of eugenol might be related to either membrane disruption or by inactivation of enzymes and genetic material. Sulieman *et al*. (2007) reported that clove oil has a potential antimicrobial activity against *E.coli, Staphylococcus aureus* and *Salmonella lyphimirium*. According to Gulcin *et al*. (2012), the antioxidant activity of clove oil compared with synthetic antioxidants measured as the scavenging of the DPPH radical decreased in the following order: clove oil > BHT > alfa-tocopherol > butylated hydroxyanisole > Trolox. The clove essential oil is widely used for its antimicrobial, antifungal and antioxidant action, as well as anesthetic and analgesic effects. These characteristics are strongly attributed to its major compound, eugenol, present in the oil in concentrations that can reach up to 90 per cent (Hongfang *et al*., 2017; Radunz *et al*., 2019).

Clove essential oil contains 89 per cent eugenol, 5 to 15 per cent eugenol acetate and β-cariofileno (Jirovetz *et al*., 2006). Another important compound found in the essential oil of clove in concentrations up to 2.1 per cent is α-humulen. Other volatile compounds present in lower concentrations in clove essential oil are β-pinene, limonene, farnesol, benzaldehyde, 2 - heptanone and ethyl hexanoate. Among spices, clove showed the higher content of polyphenols and antioxidant compounds (Rojas *et al*., 2014).

At a concentration ≤ 2%, lemon grass essential oil inhibited the growth of several kinds of microorganisms including periodontal pathogens, especially the reference strains *Actinomyces naeslundii* and *Porphyromonas gingivalis,* which were resistant to tetracycline hydrochloride. Increased tissue healing response seen in the experimental group can be explained by the antioxidant activity of lemongrass essential oil gel components (Warad *et al*, 2013).

Helander *et al*. (1998) reported that the antibacterial activity of essential oil of cinnamon may be attributed to cinnamaldehyde the antimicrobial compound of cinnamon. Cinnamon extract inhibited *E.coli* 0157:H7 and *Salmonella lyphimirium* due to cinnmaladehyde which exhibited its antibacterial activity due to its lipophilicity of terpenoids and phenyl propanoids which can penetrate the membrane and reach the inner part of the cell and impair bacterial enzyme system). Hoque *et al.* (2007) reported that essential oil of cinnamon at 5 per cent showed highest antibacterial activity against *Staphylococcus aureus.*

Cymbopogon citratus contains minor component; limonene, which was found to possess an efficient antibacterial property and that may have contributed to the higher antimicrobial activity (Lin *et al*., 1986).

Cardamom (*Eletteria cardamomum*), belonging to the family of *Zingiberaceae* has been reported to possess several pharmacological properties, including antimicrobial activity (Vaidya and Rathod, 2014).

2.7 ALOE JUICE BLENDED PRODUCTS

Boghani *et al*. (2012) made efforts to develop blended RTS beverage using papaya and aloe juice and found that papaya and aloe juice had high moisture but papaya dominated in reducing sugar, TSS and total sugar content, the blended juice could be stored for period of three months without change in chemical and organoleptic qualities. The study of *Aloe vera* fortified beverage with lemon oil revealed that the combined juice have better sensory and nutritional characters than when used individually and most recommended formulation was 90 percent lemon and 10 percent *Aloe vera* (Shailaja *et al*., 2018). The study on Orange – *Aloe vera* blended RTS revealed that the treatments were found to be suitable at

45 days of storage by the analysis of physicochemical, sensory attributes and microbiological analysis (Kausar *et al*., 2020).

Chauhan *et al*. (2012) reported slight increase in microbial count in functional herbal RTS beverage. Sunita and Ananya (2013) reported increase in TSS content in aloe juice with bael juice blended RTS beverage from 14.10 ^{0}Brix to 14.25 ^{0}Brix. Sasikumar (2015) reported the highest score and acceptability for aloe gel incorporated fruit beverage. Hirdyani (2015) and Hirdyani and Soni (2016) reported microbial load in RTS beverage prepared using traditional medicinal plants.

MATERIALS AND METHODS

3. MATERIALS AND METHODS

The study on "Protocol development for gel stabilization and nutraceuticals in *Aloe vera* (L.) Burm. f. was carried out at the Department of Plantation Crops and Spices, College of Agriculture, Vellayani during March 2016 to December 2019. The details about experimental materials and methodology adopted for the study are presented in this chapter.

The project envisages formulating a low cost stabilization technique for aloe gel using herbal extracts and aromatic oils and development of protocols for the preparation of dried latex and marketable nutraceuticals using aloe gel.

The study was carried out as four experiments. Keeping quality and natural spoilage flora of fresh gel was studied in the first experiment (Experiment I). The second experiment (Experiment II) consisted of standardization of curacao aloe (dried latex products) preparation. The third experiment (Experiment III) was on gel standardization using herbal extracts/essential oils. The fourth experiment (Experiment IV) consisted of preparation of nutraceuticals such as aloe herbal health drink and aloe herbal powder from the stabilized liquidized aloe gel juice standardized from Experiment III.

3.1 STUDY ON KEEPING QUALITY AND NATURAL SPOILAGE FLORA OF FRESH GEL

3.1.1 Raw material collection and gel preparation

Uniform mature aloe leaves were harvested from the aloe field maintained in the Dept. of Plantation Crops and Spices, College of Agriculture, Vellayani. Harvested leaves with their basal portion intact were washed using distilled water. One centimeter of the leaf base was removed using a sharp knife and the latex was allowed to drain by keeping vertically for one hour. After one hour draining of the latex, the base of the leaf was cut (1 cm) and washed again with distilled water. The latex thus collected was set apart for standardization of curacao aloe (dried latex) preparation. Then the tapering point of the leaf top along with the short and sharp spines located along the leaf margins were cut using a sharp knife and the outer rind was removed using a peeler. Then the gel fillets were removed from the bottom rind by scooping it using a spoon. The separated gel fillets were liquidized and filtered to remove the cellular material. The gel in this crude form was used for the study.

The crude gel was kept in closed glass bottles at ambient condition and daily observations on microbial population, colour change, odour, pH, viscosity, refractive index and optical density of the samples were recorded from the day of extraction upto one week of storage.

Treatment

1. D_1 - First day of storage

2. D_2 - Second day of storage

3. D_3 - Third day of storage

4. D_4 - Fourth day of storage

5. D_5 - Fifth day ofstorage

6. D_6 - Sixth day of storage

7. D_7 - Seventh day of storage

Design of the experiment: Completely Randomized Design

Treatments: 7 (daily observations for a period of one week)

Replication: 3

3.1.1.1 Microbial population

The microbial population in the liquidized aloe gel juice was estimated using the serial dilution plate technique (Johnson and Curl, 1972). Plate count is the most frequently used method to measure microbial numbers. As microbial numbers in original sample is not known, original sample is diluted several times in sterile water to ensure that separate colonies are developed and that some colony counts will be in specified range. The process is called serial dilution. Usually a tenfold serial dilution is used in the process where 1 ml of sample is transferred to a 9 ml of the diluent to get 1 : 10 dilution, 1 ml of this was transferred to another tube of 9 ml of diluent to get 1 : 100 dilution and so on till the required dilutions are obtained.

In this experiment, the 1 ml of liquidized aloe gel juice sample was pipetted out and transferred to 9 ml of sterile water in the test tube and shaken for 5 - 10 minutes using rotary shaker. From the suspension different dilutions of 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5}, 10^{-6} and 10^{-7} were prepared. For finding the fungal population of the liquidized aloe gel juice, the dilutions of 10^{-3} and 10^{-4} were taken and 1ml of these dilutions were poured into petri plates containing 25 ml of Rose Bengal Agar medium. The Rose Bengal Agar medium was prepared according to the procedure of Martin (1950) (Appendix I. C). For finding the bacterial and *E.coli* populations in liquidized aloe gel juice, the dilutions of 10^{-6} and 10^{-7} were taken and 1ml of these dilutions were poured into petriplates containing 25ml of Nutrient Agar Medium and 25 ml of Eosine Methylene Blue Agar medium, respectively. The procedure for the preparation of Nutrient Agar Medium by Timonin (1940) (Appendix I. A) and Eosine Methylene Blue Agar medium by Levine (1918) (Appendix I. B) was followed. These plates were kept at room temperature for 24 hours to observe the bacterial and *E.coli* colonies and 72 hours to observe the fungal colonies.

$$\begin{array}{c}\text{Microbial population}\\ \text{(Colony forming unit/ml)}\end{array} = \frac{\text{No. of colonies formed x dilution factor}}{\text{Volume sampled}}$$

3.1.1.2 Colour change

Visual evaluation of the liquidized aloe gel juice was undertaken using the quality standard prepared by International Aloe Science Council (IASC). According to IASC (2017) the colour of liquidized aloe gel juice varies from clean to light yellow or beige.

3.1.1.3 Odour

The odour of the liquidized aloe gel juice was observed following the visual evaluation procedure of quality standard prepared by International Aloe Science

Council (IASC). According to IASC (2017) odourof liquidised aloe gel juice varied from odourless to mildly vegetative.

3.1.1.4 pH

pH of liquidized aloe gel juice was determined (Jackson, 1973), using pH meter (ELICO LI120).

3.1.1.5 Viscosity

Viscosity of liquid is defined as the resistance of a liquid to flow. It was measured using Brookfield Viscometer (DV-II+ Pro EXTRA) (Plate 1). 15 ml of sample was inserted in the instrument by rotating the paddle or spindle at 160 rpm and measured in centipoises (cP) (Viswanath *et al*., 2007).

3.1.1.6 Refractive Index

Refractive index is a fundamental physical property of a substance, used to identify a particular substance, confirm its purity, or measure its concentration. The refractive index of the liquidized aloe gel juice was measured using Refractometer (Model RA 130) after calibration.

3.1.1.7 Optical density

Optical density is a measure of transparency of a liquid and it determines the quality of the aloe gel. Optical density was measured using spectrophotometer. The absorbance reading of aloe gel juice was done by setting the wavelength at 400 nm using distilled water as blank (Chandegara *et al*., 2015).

3.2 STANDARDIZATION OF CURACAO ALOE (DRIED LATEX) PREPARATION

The latex extracted was subjected to different methods of processing till complete drying was obtained. The methods of drying included boiling followed by cooling, sun drying, shade drying and oven drying.

Design of the experiment: Completely Randomized Design

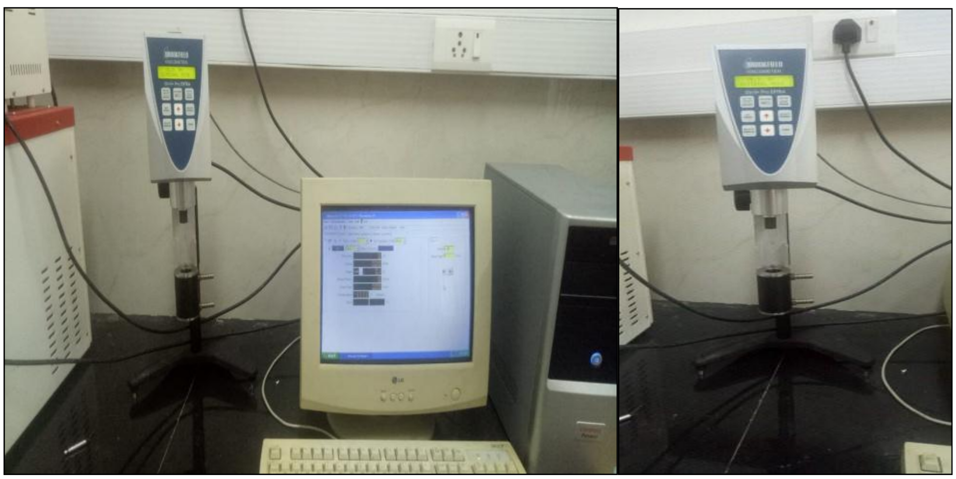


Plate 1. Brookfield Viscometer

Treatments: 4

Treatments

1. L_1 - Boiling followed by cooling

2. L_2 - Sun drying

3. L_3 - Shade drying

4. L_4 - Oven drying (50 ^{0}C – 60 ^{0}C)

The quality assessment of dried latex was assessed by physical appearance and aloin content.

3.2.1 Physical Appearance

The dried aloe latex was evaluated for sensory characteristics *viz*., appearance and colour by 25 member semi trained panel. The panel were asked to score the appearance and colour of the sample using 9 point hedonic scale (Ranganna, 1986) in the order of preference (Appendix II).

3.2.2 Aloin Content

The aloin content of aloe latex dried under different methods of processing was estimated by HPLC method.

No. of treatments: 4

Replication : 5

Design of the experiment: Completely Randomized Design

Standard aloin having 97 per cent purity was purchased from Sigma-Aldrich, USA and HPLC gradient grade acetonitrile was purchased from Merck, India. PBS buffer was prepared. Ultrapure type - I (18.2 MΩ) water prepared in the laboratory using Elga water purification systems was used. One litre of PBS buffer was prepared by adding 8 g of NaCl, 0.2 g of KCl, 1.44g of Na_2HPO_4 and 0.24 g of KH_2PO_4 in 800ml of distilled water, pH adjusted to 7.4 with HCl and made upto 1 litre with distilled water. Polyvinylidene fluoride (PVDF) syringe filters (17 mm, 0.2 µm) were used for injecting the sample (Machado *et al*., 2016).

A stock solution of 400 mg/l of aloin was prepared in PBS buffer and was serially diluted to 1, 0.5, 0.25, 0.1, 0.05 and 0.025 mg/l from 10 mg/l intermediate standard. This serially diluted standard was used for studying the linearity range and spiking for recovery studies. The concentration of the standard solution was calculated using the formula

$$\text{Concentration (mg/l)} = \frac{\text{Weight (mg) x } 10^{6} \text{ x purity (\%)}}{\text{Volume of standard x 100}}$$

Aloin content in dried latex were estimated in the High Performance Liquid Chromatography (Prominence UFLC - LC- 20AT) along with degasser (DGU-20A5), auto-injector (SIL- 20AHT)and 20 AD binary pump with photo diode array detector (PDA SPD-M20 A) supplied by Shimadzu Corporation, Kyoto, Japan. A Luna C18 (250 mm x 4.66 mm i.d, 5 µm particle size) column (Phenomenex) was used. The sample and column temperature were maintained at 10 ^{0}C and 35 ^{0}C respectively. The mobile phase was acetonitrile (A) and water (B) maintaining an isocratic ratio of 60 : 40 throughout the run time. The total run time of the analysis was 15 min with a flow rate of 1 ml min^{-1}. The injection volume was 20 µL.

5 mg of sample was dissolved in 5 ml PBS buffer. From this sample 1 ml was taken in a 1.5 ml HPLC glass vial and used for the analysis. Aloin was identified by comparing its retention time with the standard using characteristic spectra obtained from PDA and the aloin content was expressed in mg/ml.

3.3 GEL STABILIZATION USING HERBAL EXTRACTS/ESSENTIAL OILS

The liquidized aloe gel juice was stabilized using the following seven natural herbal extracts prepared in three forms (aqueous, tincture, decoction) and five essential oils (Plate 2, Plate 3, Plate 4, Plate 5, Plate 6, Plate 7, Plate 8 and Plate 9).

Design of the experiment: Completely Randomized Design

Treatments: 26

(Herbal extracts – 7, Forms – 3, Aromatic oils – 5)

Control: Sodium benzoate (0.08 %)

Total no. of treatments: 7 x 3 +5+1 = 27

Replication: 5

Herbal extracts selected were

1. *Gymnema sylvestre* (leaves)

2. *Centella asiatica* (shoot)

3. *Achyranthes aspera* (shoot)

4. *Tridax procumbens* (shoot)

5. *Terminalia chebula* (fruit)

6. *Punica granatum* (rind)

7. Green tea (dried leaves)

Each of the herbal extracts prepared in three forms (aqueous, tincture and decoction) were used.

Aromatic oils used were

1. Sacred basil oil (*Ocimum basilicum*)

2. Lemon grass oil (*Cymbopogon flexuosus*)

3. Cinnamon bark oil (*Cinnamomum zeylanicum*)

4. Clove oil (*Syzigium aromaticum*)

5. Cardamom oil (*Eletteria cardamomum*)

a) Aqueous extracts: Aqueous extract was prepared by grinding 10 g of fresh herb with 50 ml of distilled water using mortar and pestle followed by filtration.

b) Tincture: The plant material (fresh) and ethyl alcohol are taken in the ratio of 1: 5 (Rasul, 2018). Tincture was prepared by grinding 10g of sample with 50ml of 80 per cent ethanol, followed by filtration.

c) Decoction: The starting ratio of crude drug to water was fixed, as 1:4 the volume was then brought down to one-fourth to its original volume by boiling during the extraction procedure. Then, the concentrated extract was filtered and used. Fresh herb was cleaned and made into small pieces and 10g of pieces of fresh herb was boiled in 40 ml of water for 15minutes to reduce to one fourth of the original volume which was then cooled and strained (Rasul, 2018).

3.3.1 Preliminary Trials

Preliminary trials were conducted using 1 ml, 1.25ml, 1.50ml, 1.75ml and 2ml of herbal extracts and essential oils. Liquidised and filtered aloe gel juice was taken and pH was measured. pH of the juice was adjusted to 3.5 by adding citric acid (0.5 %) and 25 ml each of juice was mixed with varying concentrations namely *viz*., 1 ml, 1.25 ml, 1.50 ml, 1.75 ml and 2 ml of herbal extracts and essential oils. The treatment combinations taken for preliminary trails were described in Appendix V.

The best concentrations of herbal extracts and essential oils were determined based on sensory parameters, pH, refractive index, specific gravity, microbial populations and minimum inhibitory concentration.

3.3.1.1 Sensory parameters

Liquidized aloe gel juice extracted by different treatments were evaluated for sensory characteristics *viz*., appearance, odour and colour by 25 member semi trained panel using 9 point hedonic scale (Ranganna, 1986) in the order of preference (Appendix III).

3.3.1.2 pH

pH value of liquidized aloe gel juice were determined using pH meter (ELICO LI120) as described in 3.1.1.4

3.3.1.3 Refractive index

Refractive index of liquidized aloe gel juice were measured using Refractometer Model RA 130 as described in 3.1.1.6

3.3.1.4 Specific gravity

The specific gravity of liquidized aloe gel juice was determined using the specific gravity bottle (FSSAI, 2015).

3.3.1.5 Microbial populations

Microbial populations in liquidized aloe gel juice were determined using the method described in 3.1.1.1.

3.3.1.6 Minimum inhibitory concentration (MIC)

The minimum inhibitory concentration is defined as the lowest concentration of an antimicrobial agent that inhibits the growth of a microorganism (Tudela *et al*., 2003). MIC was found for the fungus *Aspergillus niger* by Poisoned Food Technique (Grover and Moore 1962) since the microbial load for *E. coli* was absent in the liquidized aloe gel juice samples analysed. The *Aspergilllus niger* was cultured in Rose Bengal Agar medium initially and kept for incubation for seven days. 1 ml of liquidized aloe gel juice of varying concentration (1 ml, 1.25 ml, 1.50 ml, 1.75 ml and 2 ml) were poured into the sterilized petri plates containing Rose Bengal Agar medium and agitated homogenously in a circular mode. Fungal disc of 5mm diameter was cut from the periphery of the seven day old culture of *Aspergillus niger* and was aspetically transferred in the centre of each petri plate containing the sample. The control sets were prepared using sterilized distilled water instead of sample. The assay plates were incubated for six days at $28 \pm 1\ ^{0}C$ and minimum inhibitory concentration was found out by the visual observation of the fungal colonies.

From the preliminary trails the best treatment was selected from stabilized liquidized aloe gel juice added with herbal extracts of aqueous, tincture, decoction and essential oils for final aloe gel stabilization studies.

3.3.2 Aloe gel stabilization

The best concentrations selected from the preliminary trials (3.3.1) were added to 25 ml of liquidized and filtered aloe gel juice and were stored in room temperature for six months for recording the storage life.

A B C D

Plate 2. *Gymnema sylvestre*: (A) shoots (B) aqueous extract (C) tincture extract and (D) decoction extract

A B C D

Plate 3. *Centella asiatica*: (A) shoots (B) aqueous extract (C) tincture extract and (D) decoction extract

A B C D

Plate 4. *Achyranthes aspera*: (A) shoots (B) aqueous extract (C) tincture extract and (D) decoction extract

A B C D

Plate 5. *Tridax procumbens*: (A) shoots (B) aqueous extract (C) tincture extract and (D) decoction extracts

A B C D E

Plate 6. *Terminalia chebula*: (A) dried fruit (B) fruit powder (C) aqueous extract (D) tincture extract and (E) decoction extract

A B C D

Plate 7. *Punica granatum*: (A) dried rind powder (B) aqueous extract (C) tincture extract and (D) decoction extract

A B C D

Plate 8. Green tea: (A) leaf powder (B) aqueous extract (C) tincture extract and (D)decoction extract

A B C D E

Plate 9. Essential oils: (A) cardamom oil (B) cinnamon oil (C) lemon grass oil (D) basil oil and (E) clove oil

Treatment combinations

1. G_1 - Liquidized aloe gel juice + *Gymnema sylvestre* aqueous extract

2. G_2 - Liquidized aloe gel juice + *Gymnema sylvestre* tincture extract

3. G_3 - Liquidized aloe gel juice + *Gymnema sylvestre* decoction extract

4. G_4 - Liquidized aloe gel juice + *Centella asiatica* aqueous extract

5. G_5 - Liquidized aloe gel juice + *Centella asiatica* tincture extract

6. G_6 - Liquidized aloe gel juice + *Centella asiatica* decoction extract

7. G_7 - Liquidized aloe gel juice + *Achyranthes aspera* aqueous extract

8. G_8 - Liquidized aloe gel juice + *Achyranthes aspera* tincture extract

9. G_9 - Liquidized aloe gel juice + *Achyranthes aspera* decoction extract

10. G_{10} - Liquidized aloe gel juice + *Tridax procumbens* aqueous extract

11. G_{11} - Liquidized aloe gel juice + *Tridax procumbens* tincture extract

12. G_{12} - Liquidized aloe gel juice +*Tridax procumbens* decoction extract

13. G_{13} - Liquidized aloe gel juice + *Tridax procumbens* aqueous extract

14. G_{14} - Liquidized aloe gel juice + *Terminalia chebula* tincture extract

15. G_{15} - Liquidized aloe gel juice + *Terminalia chebula* decoction extract

16. G_{16} - Liquidized aloe gel juice + *Punica granatum* aqueous extract

17. G_{17} - Liquidized aloe gel juice + *Punica granatum* tincture extract

18. G_{18} - Liquidized aloe gel juice + *Punica granatum* decoction extract

19. G_{19} - Liquidized aloe gel juice + Green tea aqueous extract

20. G_{20} - Liquidized aloe gel juice + Green tea tincture extract

21. G_{21} - Liquidized aloe gel juice + Green tea decoction extract

22. G_{22} - Liquidized aloe gel juice + Sacred basil oil

23. G_{23} - Liquidized aloe gel juice + Lemon grass oil

24. G_{24} - Liquidized aloe gel juice + Cinnamon bark oil

25. G_{25} - Liquidized aloe gel juice + Clove oil

26. G_{26} - Liquidized aloe gel juice + Cardamom oil

27. G_{27} - Liquidized aloe gel juice + Sodium benzoate (control)

The mixture of liquidized aloe gel juice with three forms of herbal extracts and essential oils were evaluated and best among the treatments were selected based on the following parameters. They are sensory parameters, refractive index, specific gravity, pH value, total solids, mono and polysaccharides, amino acids, vitamins, enzymes, protein, ash, calories, fat, crude fiber and microbial population.

3.3.2.1 Sensory parameters

Sensory parameters of liquidized aloe gel juice were examined using the method described in 3.3.1.1.

3.3.2.2 Refractive index

Refractive index of liquidized aloe gel juice were measured using the method described in 3.1.1.6

3.3.2.3 Specific gravity

Specific gravity of liquidized aloe gel juice were determined using the method described in 3.3.1.4

3.3.2.4 pH value

pH value of liquidized aloe gel juices were determined using the method described in 3.1.1.4.

3.3.2.5 Total solids

Total solids are a measure of all the suspended, colloidal, and dissolved solids in a sample. Sugars are the major soluble solids in juice. Other soluble materials include organic and amino acids, soluble pectins etc. The total solids in liquidized aloe gel juice were estimated using hot air oven at 105 ^{0}C for 10 minutes (AOAC, 2010).

3.3.2.6 Mono and Polysaccharides

3.3.2.6.1 Monosaccharides

Monosaccharide was the reducing sugar present in the sample and was determined by the titrimetric method of Lane and Eynon described by Rangana (1977). Monosaccharide content was expressed in percentage.

3.3.2.6.2 Polysaccharides

Polysaccharide determination in liquidized aloe gel juice was carried out using phenol and concentrated sulphuric acid using standard as 100 μg/ml of glucose (Bhatti *et al*., 2013). Aliquots were taken from this solution to obtain sugar concentrations 60-90 μg/ml. 1 ml of 5 per cent phenol solution was added to 1 ml of sugar solution followed by 5 ml of concentrated H_2SO_4. The absorbance was measured after 10 minutes at 488 nm against blank. To estimate the polysaccharide content in the aloe juice, 1 ml of 5 per cent phenol were added to the 1 ml of sample extract, followed by 5 ml of concentrated H_2SO_4. The absorbance was measured after 10 minutes at 488nm against blank. The polysaccharide of liquidized aloe gel juice was expressed in mg/ml. Polysaccharide content was expressed in percentage.

3.3.2.7 Amino acids

Estimation of aminoacids was done by ninhydrin method (Sadasivam and Manickam, 2008). Ninhydrin, a powerful oxidizing agent, decarboxylated the alpha-amino acids and yielded an intensely coloured bluish purple product which was colorimetrically measured at 570 nm. The amino acids obtained were expressed in ppm.

i. 50 mg leucine was dissolved in 50 ml of water in a volumetric flask. 10ml of this stock standard was taken and diluted to 100ml in another volumetric flask for preparing working standard solution. A series of the solution from

0.1 - 1 ml of this standard solution has given a concentration range of 10 µg- 100 µg. The colour of these samples was read.

ii. 0.8 unit stanous chloride was dissolved in 500 ml of 0.2 M citrate buffer (Ph 5.0). To this solution, 20 g of ninhydrin in 500 ml of methylcellosolve (2 methoxyethanol) was added.

iii. 0.2 M Citrate buffer pH 0.5 was taken.

iv. Equal volumes of water and n - propanol was mixed and used.

1 ml of liquidized aloe gel juice sample were pipetted out and centrifuged. 0.1 ml of supernatant was then pipetted for estimation and diluted it into 1 ml with distilled water. 1 ml of ninhydrin reagent was added to this and tubes were closed with stopper and kept it in boiling water bath for 20 min. The sample was cooled and 5 ml of water - isopropanol mixture was added. This was mixed and the absorbance was measured at 570 nm after 10 min.

3.3.2.8 Vitamins

3.3.2.8.1 Vitamin A

The vitamin A content in liquidized aloe gel juice was estimated by the method of Sadasivam and Manickam (1992). 1 ml of sample was taken and crushed in 10 - 15 ml of acetone, until the colour was removed. Few crystals of anhydrous sodium sulphate were added with the help of mortor and pestle. The supernatant was collected into a beaker and the process was repeated twice. The combined supernatant was transferred into a separatory funnel and 10 - 20ml of petroleum- ether was added and mixed it vigorously. Two layers were separated on standing, discarded the lower layer and collected upper layer in a volumetric flask. Made up the volume to 100 ml with pet - ether and recorded OD at 452nm. In present study the vitamin A content obtained was expressed in mg/100ml.

Vitamin A was calculated by the following formula:

$$\text{Vitamin A} \left(\frac{\mu\text{g}}{100\text{mg}}\right) = \frac{\text{OD x } 13.9 \text{ x } 10^4 \text{ x } 100}{\text{Wt of sample x } 560 \text{ x } 100}$$

3.3.2.8.2 Vitamin C

Ascorbic acid content of the samples was determined by 2, 6-dichlorophenol indophenol (DCPIP) titration method described by Rao and Deshpande (2006). Vitamin C content was expressed in percentage.

3.3.2.9 Enzymes

3.3.2.9.1 Amylase activity

The presence of amylase enzyme was determined by the method of Bird and Hopkins (1954). Starch solution (substrate) was diluted 1 : 1 with a 0.04 M phosphate buffer at pH 5.9. The working iodine reagent was prepared fresh by diluting 1.0 ml of 0.1 N iodine solution to100times. 5ml of substrate solution were added to a test tube and maintained for 10 min at an incubation temperature of 80 ^{0}C in a water bath. Enzyme extract (0.5 ml) was added to the substrate solution and incubated under the test conditions. The sample was added to 5ml stopping reagent (0.1 MHCI). After mixing, 0.5 ml of this mixture was added to 5.0 ml working iodine solution. The intensity of blue color was measured at 500nm in a spectrophotometer. The instrument was set to zero with iodine blank containing neither enzyme nor substrate. The activity of the enzyme were calculated using the formula

Amylase activity (unit/ml) = $D\,[(R_0-R)/R_0] \times 100$

Where,

R_0 is the absorbance of the substrate - iodine complex in the absence of enzyme

R is the absorbance of the digest (sample)

D is the dilution factor of the enzyme.

In present study the amylase activity was expressed in unit per 100ml.

3.3.2.9.2 Lipase activity

Lipase activity was measured following the method described by Sadasivam and Manickam (2008). 20 ml of substrate (2 ml of clear vegetable oil, stirred well with 25 ml of water in the presence of 100 mg bile salts till an emulsion was formed and to hasten the reaction 2 g of gum Arabic were added) was taken in a 500 ml beaker and phosphate buffer (5 ml) of pH7.0 were added into it. The beaker was set on the top of a magnetic stirrer cum hot plate and the contents were stirred slowly. The temperature was maintained at 35^0C and the electrodes of the pH meter were dropped into the reaction mixture and the pH was adjusted to 7.0. 0.5ml of enzyme extract was added and the pH was immediately recorded and the timer was set on to be the pH at zero time. 0.1 N NaOH was added at 10 minutes intervals or as the pH dropped by about 0.2 units to bring the pH to the initial value. The titration was continued for a period of 30 - 60 minutes. The volume of alkali consumed was noted.

$$\text{Activity meq/min/g sample} = \frac{\text{Volume of alkali consumed x strength of alkali}}{\text{Volume of sample x time (in min)}}$$

Amylase activity was expressed in unit per 100ml.

3.3.2.10 Protein

Estimation of proteins by Bradford's colorimetric method (Sadasivam and Manickam, 2008).1.0ml of sample was transferred into centrifuge tube and mixed with1ml 20 per cent TCA, kept for 30 minutes and was centrifuged at 5000 rpm for 30 minutes. The pellet supernatant was discarded. The pellet was washed with acetone two times, centrifuged each time and discarded the supernatant. The pellet was dissolved in 2 ml 0.1N NaOH. 1.0 ml aliquot of solution was taken for estimation. For the estimation aliquots of 0, 0.2, 0.4, 0.6, 0.8 and 1.0 ml were pipetted out of standard solution of protein and transferred to numbered test tubes. This was diluted to 1.0 ml with phosphate buffer by adding 1.0, 0.8, 0.6, 0.4, 0.2, 0 ml in respective tubes. 5 ml dye solution (Bradford reagent) was added in all the tubes, mixed and the absorbance was read at 595 nm, setting zero with first tube (blank). A standard curve was plotted using the absorbance of standard solution of protein. The protein content was calculated from the standard curve and expressed in percentage.

3.3.2.11 Ash

Ash content in liquidized aloe gel juice was determined as described by AOAC (1990) and the content was expressed in percentage.

3.3.2.12 Calories

The energy value in calories was estimated based on the contents of protein, fat and carbohydrate using the factors 4, 4 and 9 (FAO, 2002) respectively as follows

Calorific value (kcal/100g) = 4 x total carbohydrate (%) + 4 x total protein content (%) + 9 x total fat content (%)

3.3.2.13 Fat

The fat content of the sample was determined following the Twisselman method using diethyl ether as solvent (AOAC, 1990) by gravimetric method and expressed in percentage (%).

3.3.2.14 Crude fiber

Crude fibre was determined using the method described by AOAC (1990) and expressed in percentage (%).

3.3.2.15 Microbial population

The microbial population was estimated (fungal, bacterial, *E. coli*) by the method described in 3.1.1.1.

3.4 PREPARATION OF NUTRACEUTICALS

The stabilized liquidized aloe gel juice was used for the preparation of nutraceuticals such as aloe herbal health drink and aloe herbal powder.

3.4.1 Aloe herbal health drink

Aloe herbal health drink was prepared by adding lemon juice, orange juice and honey in proportions of 50 : 50, 25 : 75 and 10 : 90 to stabilized liquidized aloe gel juice followed by pasteurization and flash cooled. The aloe herbal health drink was kept in 30ml glass bottles for taking the observations.

Treatment combinations

1. T_1 **-** Lemon juice+ Liquidized aloe gel juice (50 : 50)
2. T_{2} - Lemon juice + Liquidized aloe gel juice (25 : 75)
3. T_{3} - Lemon juice + Liquidized aloe gel juice (10 : 90)
4. T_{4} - Orange juice + Liquidized aloe gel juice (50 : 50)
5. T_{5} - Orange juice + Liquidized aloe gel juice (25 : 75)
6. T_{6} - Orange juice + Liquidized aloe gel juice (10 : 90)
7. T_{7}- Honey + Liquidized aloe gel juice (50 : 50)
8. T_{8} - Honey + Liquidized aloe gel juice (25 : 75)
9. T_{9} - Honey + Liquidized aloe gel juice (10 : 90)

Treatments: 9

Replication: 6

Design of the experiment: Completely Randomized Design

Organoleptic parameters, chemical/nutrient composition and shelf life of the aloe - herbal health drinks were carried out.

3.4.1.1 Organoleptic parameters

Organoleptic parameters *viz*., odour, color, taste, appearance and overall acceptability of aloe herbal health drink were scored by 25 member semi trained panel using 9-point hedonic scale (Ranganna, 1986) (Appendix IV).

3.4.1.2 Chemical/nutrient composition

Acidity, pH, TSS, vitamin C, calories, iron, calcium, carbohydrate, fat and protein content of aloe herbal health drink were analysed.

3.4.1.2.1 Acidity

Acidity was determined according to the procedure of AOAC (2010). Titrable acidity was expressed in meq/ml.

3.4.1.2.2 pH

pH value of aloe herbal health drink was determined using the method described in 3.1.1.4.

3.4.1.2.3 TSS

The total soluble solids in aloe - herbal health drink were estimated in degree brix by means of the refractometer in 0.1 per cent graduations (AOAC, 2010).

3.4.1.2.4 Vitamin C

Vitamin C content of aloe herbal health drink was determined using the method described in 3.3.2.8.2.

3.4.1.2.5 Energy

Energy value of Aloe herbal health drink was determined using the method described in 3.3.2.12**.**

3.4.1.2.6 Iron

The iron content of aloe herbal health drink was estimated using the diacid digestion and estimation using Atomic Absorption Spectrophotometry (AAS) by Jackson (1973). For the digestion of the sample, 1ml of the sample was taken in a 100ml conical flask and 10 ml of digestion mixture was added (HNO_3 : $HClO_4$ – 9:4) and shaken gently. For measuring the iron content instrument was set with zero using the blank (blank digest). Absorbance/ concentration of iron in the sample were recorded after standardization of the instrument was done with standard iron. Iron content was recorded in percentage (%).

$$\text{Fe (\%)} = \frac{\text{AAS reading (mg/l) x dilution}}{\text{Volume of sample taken}}$$

3.4.1.2.7 Calcium

The calcium content of aloe - herbal health drink was estimated using Ethylene Diamine Tetra Acetic Acid (EDTA) titration method, after wet digestion of the samples using di – acid mixture (Jackson, 1973).

The digestion of the sample was done by taking 5ml of the sample in a100ml standard flask. In the sample 10 ml of diacid digestion mixture (HNO_3: $HClO_4$ – 9:4) was added, gently mixed it and made up the volume to 100ml using distilled water. From this digestion mixture 10 ml of the sample was pipetted out into a conical flask. 5ml of distilled water, 5ml of NaOH, 10 drops of hydroxylamine hydrochloride and tri ethanol amine and 1ml of calcon (indicator) was added into the sample. The resultant pink solution was titrated with EDTA to get a blue end point and calcium content in sample was calculated in percentage.

Calculation:

$$\text{Ca (\%) in the sample} = \frac{\text{Titre value x Normality of EDTA (0.01N)x 0.02x100x100}}{\text{10 x volume of the sample taken}}$$

3.4.1.2.8 Carbohydrate

Estimation of carbohydrate was done by using anthrone reagent (Hedge and Hofrieter, 1962). 2 ml of aloe herbal health drink was taken and subjected to centrifugation at 3000 rpm for 10 mins. 0.5ml of aliquote from the supernatant was taken, made upto 1ml with distilled water and 4.0 ml of anthrone reagent added into it. The tubes were kept in boiling water bath for 10 minutes, cooled rapidly under running water and the absorbance of dark green coloured samples were read at 630 nm.

Pipetted out aliquots of standard solution 0, 0.2, 0.4, 0.6, 0.8 and 1.0 ml (0.1 mg/ml glucose) into test tubes, diluted to 1.0 ml final volume using distilled water by adding 1.0, 0.8, 0.6, 0.4, 0.2 and 0 ml in respective tubes. 4.0ml of anthrone reagent were added and kept the tubes in boiling water bath for 10 minutes. It was then cooled under running water and the absorbance of dark green coloured sample was read at 630 nm The standard graph was drawn by plotting concentration of standard on the X - axis versus absorbance on the Y - axis and amount of carbohydrate was calculated from the graph. The carbohydrate content was expressed in percentage.

3.4.1.2.9 Fat

Fat content in aloe herbal health drink was estimated by the method described in 3.3.2.13 and expressed in percentage.

3.4.1.2.10 Protien

Estimation of protein content in aloe herbal health drink was done by Bradford's colorimetric method as described in 3.3.2.10.

3.4.1.3 Shelf life

Shelf life of aloe herbal health drink was assessed by determining organoleptic parameters *viz*., color, odour, taste, appearance, overall acceptability and microbial population according to the procedure described in 3.4.1.1 and 3.1.1.1 respectively till the product becomes unfit for consumption.

3.4.2 Aloe herbal powder

The stabilized and pasteurized liquidized aloe gel juice was utilized for the preparation of aloe herbal powder by adopting different methods of drying. Each of juice with 100 ml was taken.

Treatments

1. P_1 - Solar drying

2. P_2 - Air drying

3. P_3 - Oven drying (50^0C – 60^0C)

4. P_4 - Freeze drying (- 40^0C – - 50^0C)

Treatments: 4

Control: Fresh gel

Replication: 5

Plate 10: Freeze drier

Design of the experiment: Completely Randomized Design

The stabilized liquidized aloe gel juice were subjected to different methods of drying

1. Solar drying: The samples were dried under direct sunlight for a period of 6 hours daily for one week.

2. Air drying: The samples were dried under laminar air flow using the blower for a period of 6 hours daily for one week.

3. Oven drying: The sample was dried in oven by adjusting the temperature within a range of 50 ^{0}C – 60 ^{0}C for a period of 6 hours daily for one week.

4. Freeze drying: The samples were dried in freeze dryer (Plate 10) with a temperature of - 40 ^{0}C – - 50 ^{0}C for a period of 6 hours daily for one week.

3.5 STATISTICAL ANALYSIS

The data generated from experiments were statistically analysed using one - way analysis of variance (ANOVA) and significance was tested by critical difference. The score for sensory parameters was statistically analysed using Kruskall – Wallis test and the calculated value of Kruskal – Wallis test was compared with the Chi – square value to find the significance.

RESULTS AND DISCUSSION

4. RESULTS AND DISCUSSION

The results obtained from the study on "Protocol development for gel stabilization and nutraceuticals in *Aloe vera* (L.) Burm. f." were analysed, presented and discussed in this chapter under headings

1. Study on keeping quality and natural spoilage flora of fresh gel
2. Standardization of curacao aloe (dried latex) preparation
3. Gel stabilization using herbal extracts/essential oils
4. Preparation of nutraceuticals

4.1 STUDY ON KEEPING QUALITY AND NATURAL SPOILAGE FLORA OF FRESH GEL

Keeping quality of liquidized aloe gel juice stored in closed glass bottles at ambient condition was studied by recording observations daily on changes in microbial population, colour change, odour, pH, viscosity, refractive index and optical density for a period of seven days.

4.1.1. Microbial population

Data on mean fungal, bacterial and *E. coli* population in liquidized aloe gel juice are presented in table 1. The mean fungal population of the liquidized aloe gel juice samples did not show any significant variation between days. How ever significant variation in mean bacterial population was noticed between days. The mean bacterial population on the first day of storage was 6.5 log cfu ml^{-1} which increased to 7.52 log cfu ml^{-1} on the second day of extraction reaching 8.5 log cfu ml^{-1} at the end of seventh day of extraction (Plate 11). *E. coli* was absent in the samples through out the storage period. The microorganisms present on the leaves contaminate *Aloe vera* gel, during extraction and processing and cause spoilage (Coats, 1979). The growth and activity of these microorganisms consequently can adversely affect the overall quality and shelf life of the end product (He *et al.*, 2005). According to Eshun and He (2004) to ensure the microbiological quality of *Aloe vera* gel and to extend its shelf life, it is often necessary to apply thermal processing such as scalding, pasteurization and dehydration which may cause irreversible modifications to the physiological, pharmacological and nutritional properties of its components. Washing of whole *Aloe vera* leaves in a sodium hypochlorite solution (200 ppm) before processing showed limited effectiveness to prevent cross contamination of the final product, which might be because microorganisms were protected by their location in the leaf tissue (internalized, in stomata, cracks, crevices, and cut surfaces) and/or by biofilms made by the microorganisms themselves (Whipps *et al*., 2008). Chandegara and Varshney (2013) supported the present study that the aloe leaves in which base is not intact and sealed would increase the microbial count and reduced the biological activity. The microbial population observed on the first day of storage might be because of the entry of microorganisms from the parent leaf during gel extraction as revealed by Whipps *et al* (2008). The microbial population during seven days of storage is shown in figure1.

4.1.2 Colour change

The colour change of the liquidized aloe gel juice stored for a week is presented in table 2. During the first three days of storage (D_1, D_2, D_3), the liquidized aloe gel juice showed off - white colour which got sedimented in fourth day (D_4) with off - white colour. The colour of the liquidized aloe gel juice became yellow in the subsequent days of storage.

Relative humidity and temperature are two most important environmental parameters that affect product quality (Hernadez and Giacin, 1998). Saeed *et al*. (2004) found that most juices from both fruits and vegetables are unstable and subject to discolouration and spoilage due to contamination by microorganisms. Esteve *et al*. (2005) reported importance of color as an indicator of changes in foods during storage or processing. One important factor affecting the composition of *Aloe vera* gel is the post harvest handling of the leaves, because the decomposition of the gel matrix starts just after its cutting due to natural enzymatic reactions and the activity of bacteria normally present on the leaves can adversely affect the quality of the end product. More over during filtration of aloe juice fibrous material is removed. Filtration influences the stability of *Aloe vera* juice. Poor filtration results in sedimentation of aloe juice on storage (Ahlawat and Khatkar, 2011). The presence of anthraquinones is an important factor leading to non enzymatic browning in aloe products (Tian *et al*., 2002). Suraiti *et al*. (2019) reported that time of storage, temperature and their interaction have effect on the color of aloe gel.

The sedimentation might be because filtration might not have been very effective. More over the physico chemical variation and microbial contamination might have set the changes in colour.

4.1.3 Odour

Th**e** odour of the liquidized aloe gel juice during seven days of storage is presented in table 2. The odour of the liquidized aloe gel juice was mildly vegetative during the first three days of storage .The odour changed to foul smell on the fourth (D_4), fifth (D_5), sixth (D_6) and seventh (D_7) day of storage.

IASC (2017) reported that the odour of liquidized aloe gel juice vary from odourless to mildly vegetative smell. The foul smell from fourth day onwards could be due to changes in physico - chemical properties which might be due to enzymatic changes and spoilage caused by microbial contamination.

Table 1. Mean microbial population of the liquidized aloe gel juice during storage, log cfu ml^{-1}

Treatments	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
D_1 (Day 1)	0.00 x 10^3 (0.00)	3.33 x10^6 (6.50)	0.00 x10^6 (0.00)
D_2 (Day 2)	0.67 x 10^3 (2.00)	34 x10^6 (7.52)	0.00 x10^6 (0.00)
D_3 (Day 3)	0.67 x 10^3 (2.00)	55.67 x10^6 (7.74)	0.00 x10^6 (0.00)
D_4 (Day 4)	1 x 10^3 (2.10)	84.67 x10^6 (7.92)	0.00 x10^6 (0.00)
D_5 (Day 5)	1 x 10^3 (2.10)	105.33 x10^6 (8.04)	0.00 x10^6 (0.00)
D_6 (Day 6)	1 x 10^3 (2.10)	227.33 x10^6 (8.36)	0.00 x10^6 (0.00)
D_7 (Day 7)	1.67 x 10^3 (3.20)	327 x10^6 (8.50)	0.00 x10^6 (0.00)
SEm (±)	0.874	0.041	0.000
CD (0.05%)	NS	0.125	NS

* Figures in parantheses indicate transformed values

* Each value is the mean of three replication

Table 2. Colour and odour of liquidized aloe gel juice during storage

Treatments	Colour	Odour
D_1 (Day 1)	Off – white	Mildly vegetative
D_2 (Day 2)	Off – white	Mildly vegetative
D_3 (Day 3)	Off – white	Mildly vegetative
D_4 (Day 4)	Sedimented and off-white	Foul smell
D_5 (Day 5)	Sedimented and light yellow	Foul smell
D_6 (Day 6)	Sedimented and light yellow	Foul smell
D_7 (Day 7)	Sedimented and light yellow	Foul smell

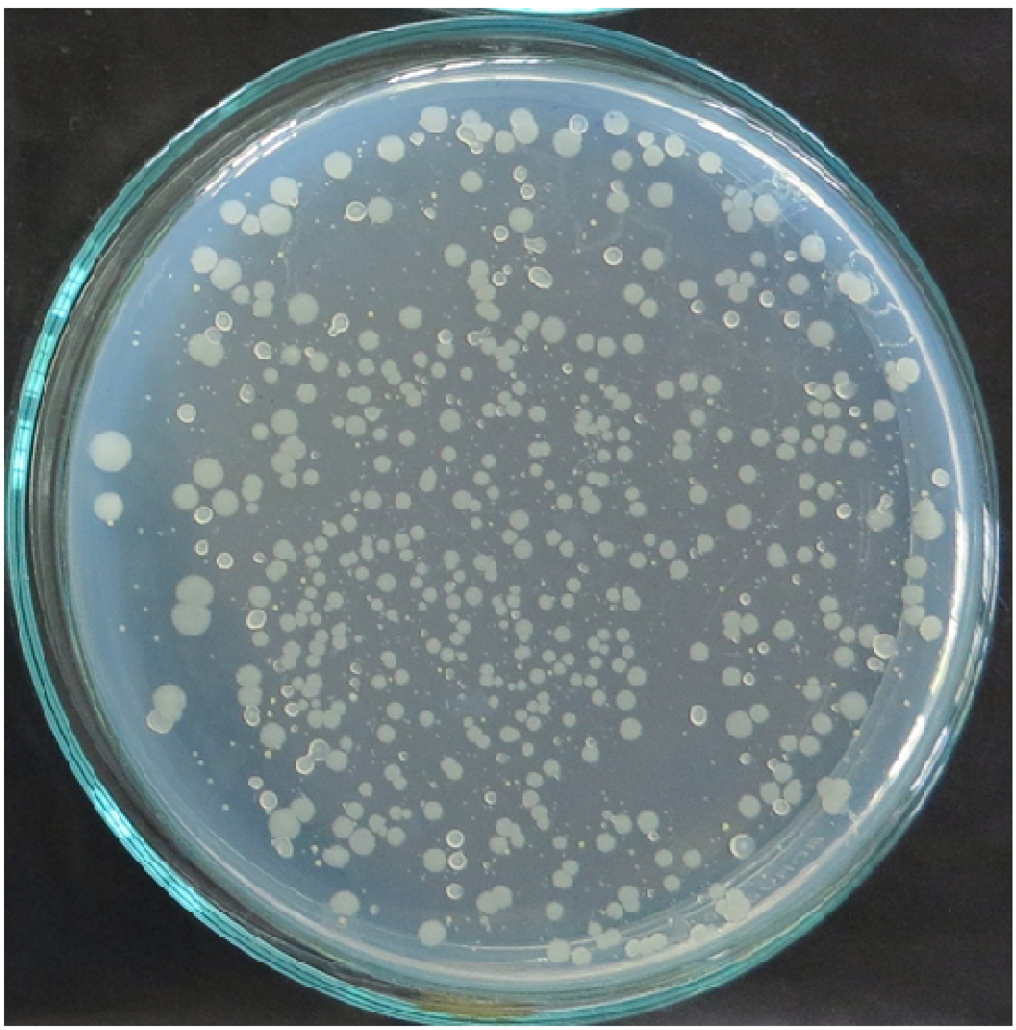


Plate 11. Bacterial population after seven days of storage in liquidized aloe gel juice

4.1.4 pH

The analysis of data on pH of the liquidized aloe gel juice for a period of one week is represented in table 3 and figure 2. pH of the liquidized aloe gel juice varied significantly on storage. The pH of the liquidized aloe gel juice on the first day of extraction was 4.52 that did not differ significantly upto three days of extraction. However the pH increased significantly on the subsequent days reaching 4.85 on the seventh day of storage (D_7).

Gel freshness is indicated by presence of malic acid. This acid is produced naturally in the leaves of aloes and other succulents whose cells contain large, water-filled vacuoles. Under poor handling conditions in the presence of bacteria, malic acid may be broken down to form lactic acid. Gram-negative rod bacteria, which grow in some aloe gels, assimilate malic acid and free glucose and produce other organic acids such as lactate or lactic acid.

Lactic acid is one of the major products of carbohydrate break-down by lactic acid bacteria during malolactic fermentation (MLF). Malolactic enzyme demonstrated to be present in most lactic acid bacteria but not in other bacteria that catalyzes the reaction (Tungala et *al*., 2011). Lodi and Rossin, (1995) reported wide variation in pH (4.8 - 6.4) of cane molasses. In the case of aloe gel, malic acid is produced naturally in aloe leaves during Crassulacean Acid Metabolism (CAM). Under poor handling or storage conditions, in the presence of bacteria, malic acid can be broken down to form lactic acid, which might have resulted in the increase in pH of stored gel. Thus, lactic acid occurrence is an indication of gel decay (Brien *et al*., 2011). Richard and Piere (2006) reported that increase in pH as an indicator of bacterial spoilage in food.

The present findings were supported by the report of Chandegara and Varshney (2013) that the pH value of aloe gel ranged from 3.5 - 4.7. The variation in pH of liquidized aloe gel juice might be due to poor handling/storage conditions. Due to the presence of bacteria, malic acid might have broken down to form lactic acid, which might have resulted in the increased pH during storage.

4.1.5 Viscosity

The viscosity of the liquidized aloe gel juice on storage varied significantly (Table 3). The viscosity of the liquidized aloe gel juice was the highest on the first day of extraction (2.59 cP) and reduced significantly from the second day of extraction. The viscosity of the liquidized aloe gel juice was the minimum (1.36 cP) on the seventh day of extraction (D_7). The viscosity of liquidized aloe gel juice during seven days of storage explained in figure 3.

The study of Gowda *et al*. (1979) supported the findings that when viscosity is higher, products will be better and biologically active.Viscous nature of aloe gel is degraded rapidly immediately after extraction as reported by Ni *et al*. (2004).

Viscosity is a parameter that reflects the fruit juice quality, it affects the taste and the ability to hold its solid portion in suspension than the product shelf life (Bravo *et al*., 2012). The viscosity of fresh aloe solutions at 10 ^{0}C was nearly twice that of the solutions prepared from powders at the same temperature. This can be attributed to the mixing and stirring that occur during the extraction process followed by the drying treatments that caused breakdown of native polysaccharide networks, hence lowering the viscosity (Nindo *et al*., 2010). In present study viscosity decreased and had affected the product quality.

4.1.6 Refractive Index

There was no significant difference in the refractive index of liquidized aloe gel juice on storage (Table 3). Chandegara *et al*. (2015) reported that refractive index of aloe gel is near to that of distilled water. In the present study refractive index was not affected during storage of fresh aloe juice for one week (Figure 4).

4.1.7 Optical density

The optical density of the liquidized aloe gel juice did not differ significantly during storage for seven days of extraction (Table 3). The optical density in the range of 1.020 to 1.437 (abs) for *Aloe vera* leaves were reported by Wang and Strong (1993). Optical density is the physical property of gel and it determines the purity of gel as compared to double distilled water. High optical density indicates the impurities in the extracted gel (Chandegara and Varshney, 2013). Chandegara *et al*. (2015) reported optical density as a measure of transparency of liquid and quality for *Aloe vera* gel.

The study by Palonen and Weber (2019) observed that optical density of raspberry fruit juice was affected by genotype, storage temperature and the storage duration. In the present study optical density was not affected by storage.The change in optical density during seven days of storage is shown in figure 5.

Table 3. pH, viscosity (cP), refractive index and optical density of liquidized aloe gel juice during storage

Treatments	pH	Viscosity (cP)	Refractive Index	Optical density
D_1 (Day 1)	4.52	2.59	1.331	1.63
D_2 (Day 2)	4.62	2.12	1.331	1.63
D_3 (Day 3)	4.69	1.61	1.331	1.65
D_4 (Day 4)	4.73	1.54	1.331	1.73
D_5 (Day 5)	4.76	1.46	1.331	1.78
D_6 (Day 6)	4.81	1.40	1.331	1.80
D_7 (Day 7)	4.85	1.36	1.331	1.89
SEm (±)	0.056	0.087	0.000	0.167
CD (0.05)	0.171	0.263	NS	NS

*Each value is the mean of three replication

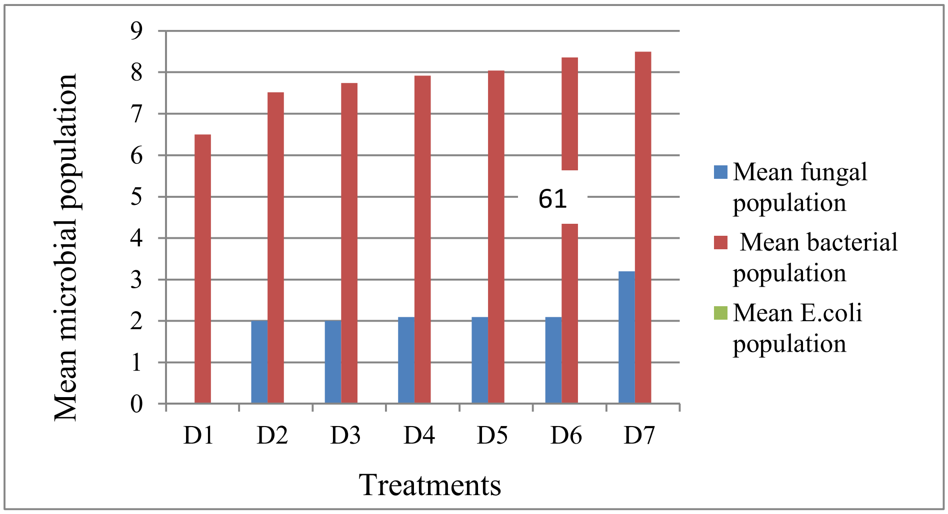


Figure 1. Mean microbial population of liquidized aloe gel juice during storage

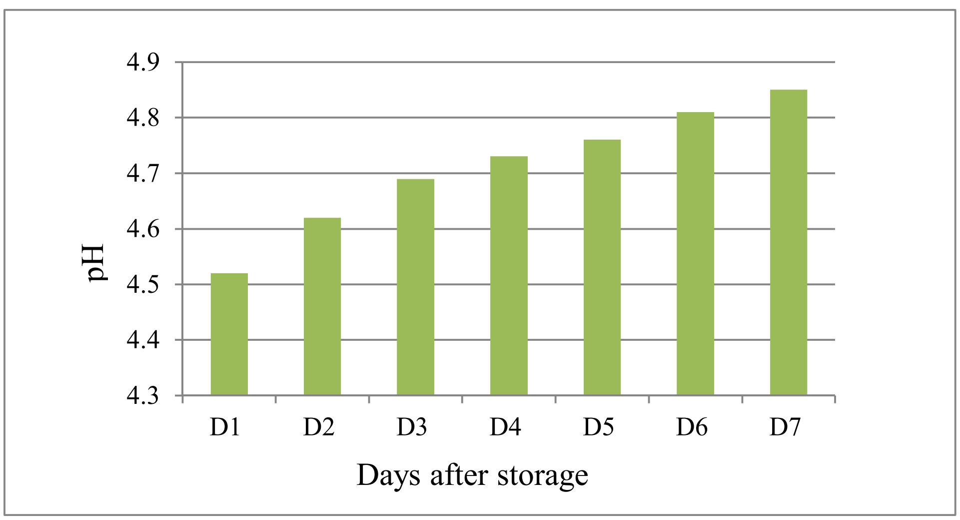


Figure 2. pH of liquidized aloe gel juice during storage

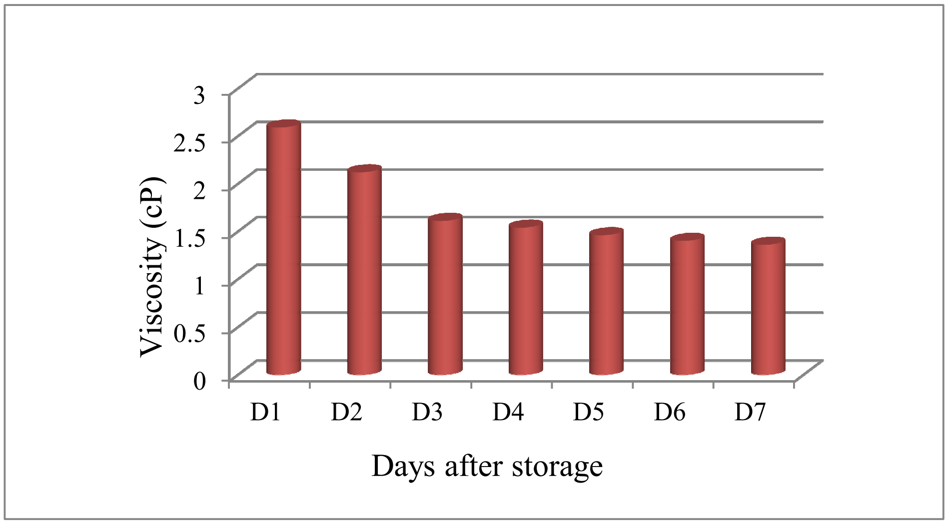


Figure 3. Viscosity of liquidized aloe gel juice during storage

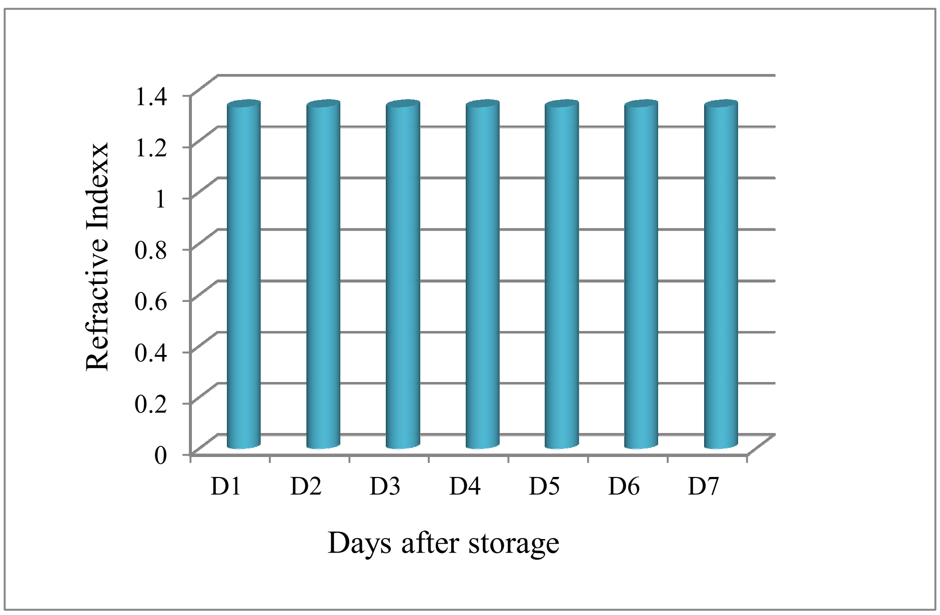


Figure 4. Refractive index of liquidized aloe gel juice during storage

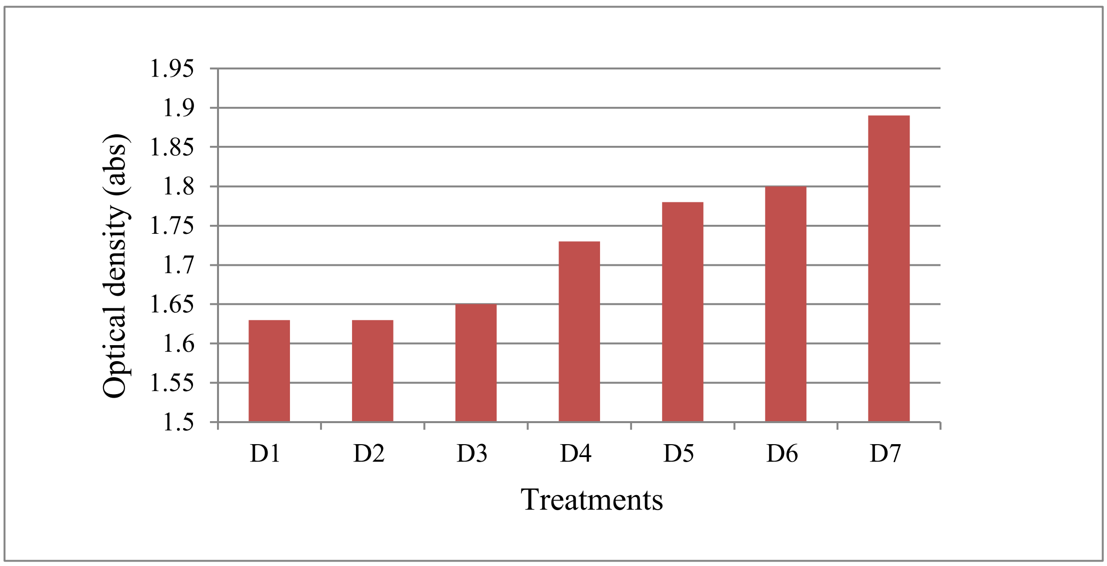


Figure 5. Optical density of liquidized aloe gel juice during storage

4.2. STANDARDIZATION OF CURACAO ALOE (DRIED LATEX) PREPARATION

The latex collected from twenty five good quality aloe leaves constituting one replication were subjected to different methods of drying such as boiling followed by cooling (L_1), sun drying (L_2), shade drying (L_3) and oven drying (L_4) (50 ^{0}C – 60 ^{0}C). There were five replications in each treatment. The physical appearance was scored and aloin content obtained by different drying methods was estimated and analysed statistically.

4.2.1 Physical Appearance

The mean rank value of colour and appearance of curacao aloe (dried latex) scored by semi trained panel consisting of 25 members is presented in table 4**.** Plate 12 represents latex obtained after using different drying methods.

Significant variation in colour and appearance of curacao aloe was noticed among different methods of drying. The results showed that the colour as well as the appearance of dried latex ranked significantly high for shade drying (L_3) which was 79.32 and 80.10 respectively. The least preference was given for the curacao aloe dried by boiling followed by cooling (L_1). The acceptance of shade dried latex was 29.7 per cent more compared to sun dried latex. Latex from *Aloe vera*, is hard and dry produced by squeezing the plant or it is a product of the normal latex flow coming from the leaves of various types of aloes includes 15% derivatives from hydroalcoholic anthracene. This yellow liquid inside the peripheral cells attached to the leaf parenchyma, starts to exit immediately after the leaf is cut and is dried when exposed to sun or fire. This latex contains some variable amounts of alion, Aloe-emodin, chyrsophanic acid, volatile oils and resin (Bahmani, 2016).

4.2.2 Aloin content

Aloin content of curacao aloe dried under different methods of drying is presented in table 5. Aloin content of curacao aloe varied significantly between different methods of drying. The aloin content obtained by shade drying (L_3) was significantly high (271.62 mg/ml) followed by oven drying (218.62 mg/ml), sundrying (159.96 mg/ml) and boiling followed by cooling (91.00 mg/ml). The chromatograms obtained are presented in figure 6.

In consensus with the present study Ding *et al*. (2014) reported that aloin constitutes a very small content of dry samples due to rapid degradation by temperature. Lesser temperature under shade resulted in high aloin content than other methods of drying. Higher aloin content were reported in fresh leaf samples with an average of 199.76 ± 0.74 mg for dry latex, the average aloin content was 176.26 ± 0.16 mg. The aloin content in fresh gel, dry gel as well as fresh and dry latex samples revealed greater aloin content in fresh latex (199.76 ± 0.74 mg) followed by dry latex (176.26 ± 0.16 mg) , fresh gel (7.87 ± 0.05 mg) and dry gel with 5.11 ± 0.12 mg aloin (Machado *et al*., 2016). The dry samples may contain less aloin due to rapid degradation by temperature. Aloin content obtained under different drying methods presented in figure 7. The lower aloin content in different drying methods except shade drying might be due to degradation of aloin under high temperatures under sun drying, oven drying and boiling followed by cooling.

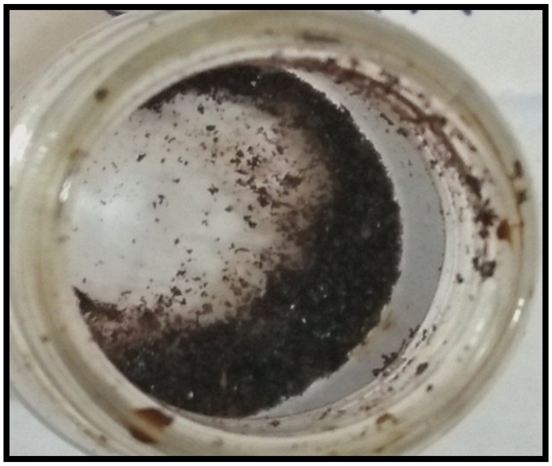


Plate 12A. Boiling followed by cooling

Plate 12B. Sun drying

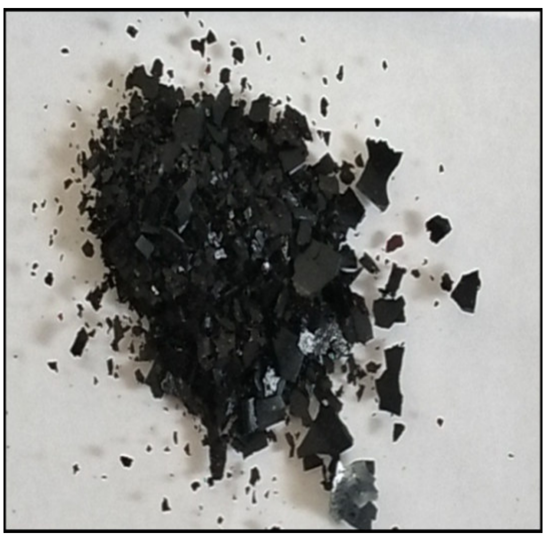


Plate 12C. Shade drying

Plate 12D. Oven drying

Plate 12. Dried latex obtained from different drying process

Table 4. Physical appearance of curacao aloe (dried latex) under different methods of drying

Treatments	Mean score value	
	Colour	Appearance
L_1 (Boiling followed by cooling)	5.88 (18.16)	5.76 (19.38)
L_2 (Sun drying)	2.66 (43.36)	6.08 (41.30)
L_3 (Shade drying)	7.88 (79.32)	8.08 (80.10)
L_4 (Oven drying)	7.2 (61.16)	7.4 (61.22)
KW Value	66.74	65.83
χ^2	9.49	
CD value	3.553	

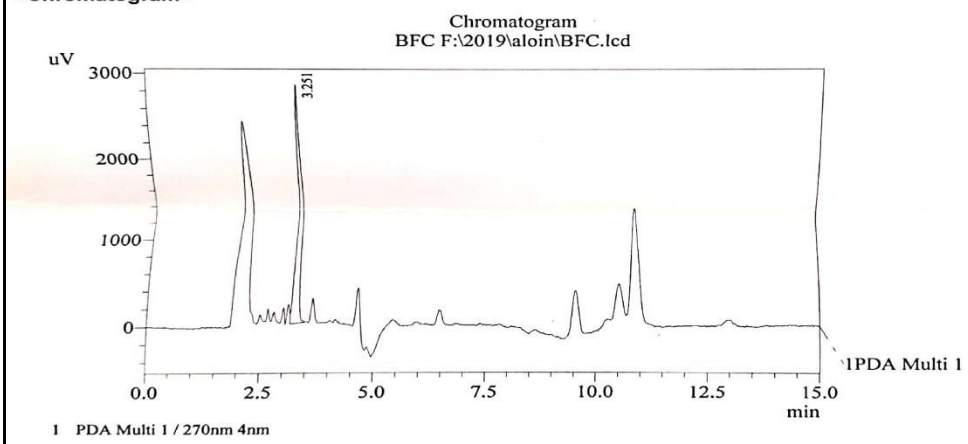
*Figures in parantheses indicate mean rank value

Table 5. Aloin content of curacao aloe (dried latex) under different methods of drying, mg/ml

Treatments	Aloin (mg/ml)
L_1 (Boiling followed by cooling)	91.00
L_2 (Sun drying)	159.96
L_3 (Shade drying)	271.62
L_4 (Oven drying)	218.62
SEm (±)	0.016
CD (0.05)	0.033

*Each value is the mean of five replication

<Chromatogram>



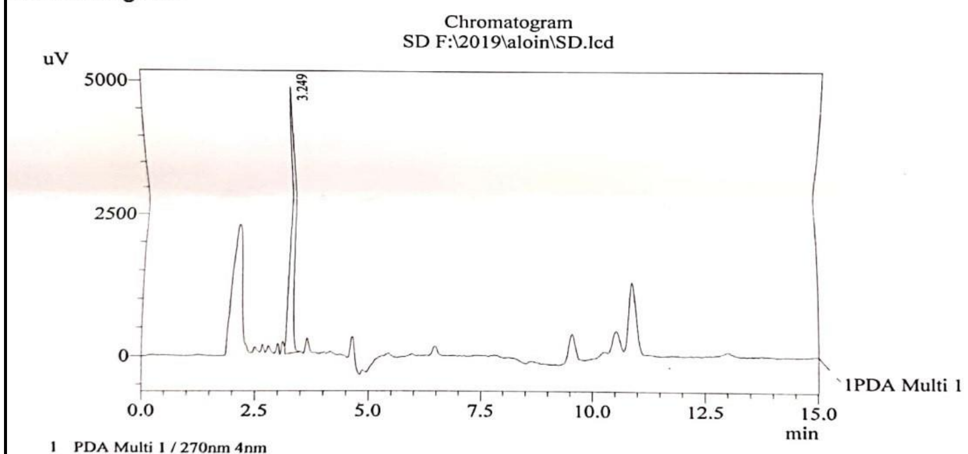
Quantitative Results

PDA

ID#	Name	Ret. Time	Area	Height	Conc.
1	Aloin	3.251	15639	2828	0.091

Figure 6 A. Boiling followed by cooling

<Chromatogram>



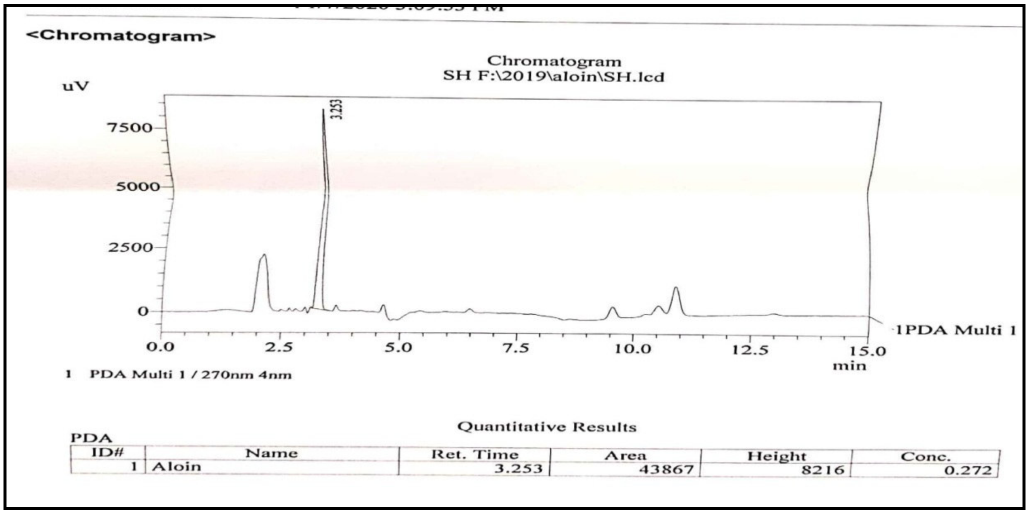
Quantitative Results

PDA

ID#	Name	Ret. Time	Area	Height	Conc.
1	Aloin	3.249	26417	4853	0.160

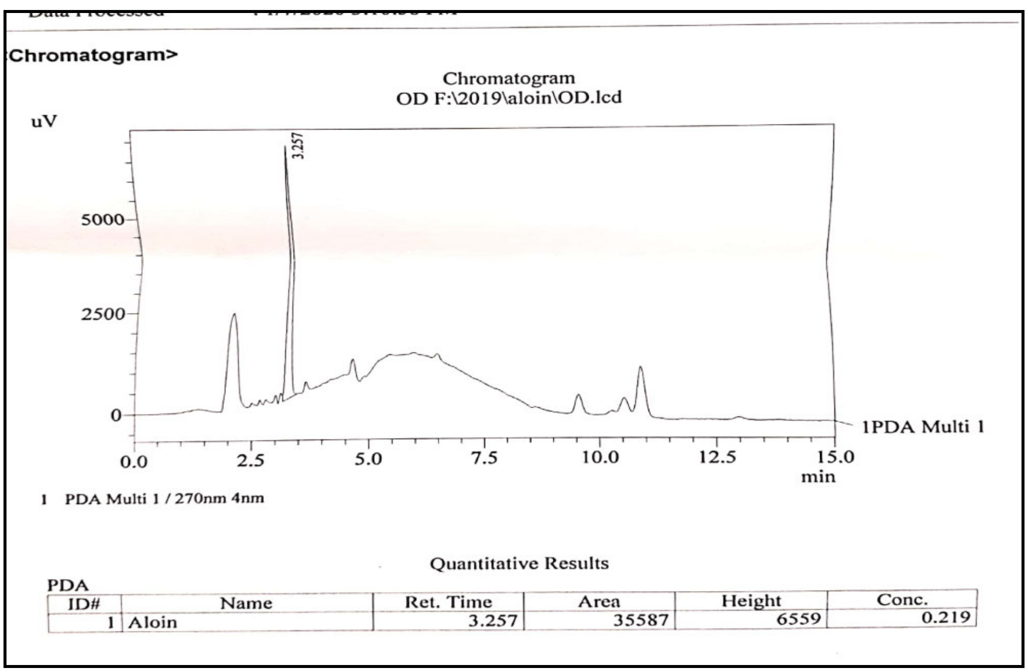
Figure 6 B. Sun drying

Figure 6. Chromatogram of aloin under different methods of drying



ID#	Name	Ret. Time	Area	Height	Conc.
1	Aloin	3.253	43867	8216	0.272

Figure 6C. Shade drying



ID#	Name	Ret. Time	Area	Height	Conc.
1	Aloin	3.257	35587	6559	0.219

Figure 6D. Oven drying

Figure 6. Chromatogram of aloin under different methods of drying

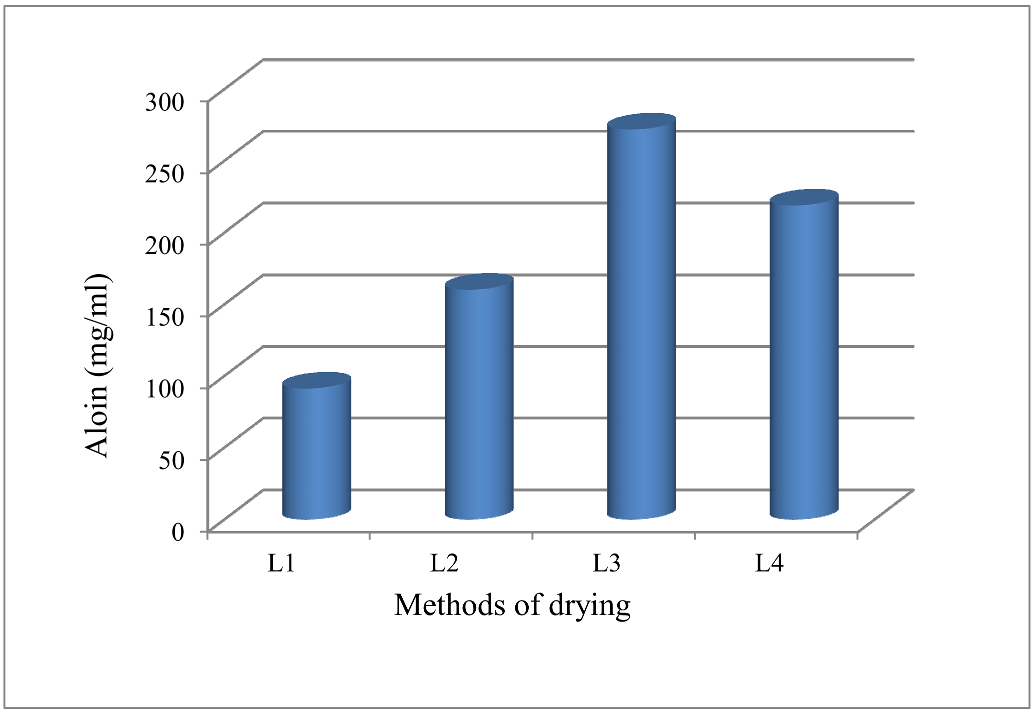


Figure 7. Aloin content of curacao aloe (dried latex) under different methods of drying

4.3. GEL STABILIZATION USING HERBAL EXTRACTS/ESSENTIAL OILS

For the stabilization of liquidized aloe gel juice, different concentrations of three forms (aqueous, tincture, decoction) of seven natural herbal extracts and five essential oils were mixed with liquidized aloe gel juice. Preliminary trials were conducted with varying concentrations of the extract and finally arrived at one concentration for each form of herbal extracts and essential oils.

4.3.1 Preliminary Trials

Preliminary trials were conducted to fix the dosage of herbal extracts and essential oils. The treatments taken for the preliminary trials of each of herbal extracts (aqueous, tincture and decoction) and essential oils were 1ml, 1.25ml, 1.50ml, 1.75ml and 2ml. The best concentration of herbal extracts and essential oils were selected based on sensory parameters, pH, refractive index, specific gravity, microbial populations and minimum inhibitory concentration. The results of sensory parameters, pH, refractive index and specific gravity are attached in the Appendix VI. The final selection of the concentration of herbal extracts and essential oil were based on microbial population and minimum inhibitory concentration.

4.3.1.1 Microbial population of liquidized aloe gel with different concentration of herbal extracts and essential oil

The microbial population of liquidized aloe gel juice added with different concentration of *Gymnema sylvestre* aqueous extract is presented in table 6.

The mean fungal, bacterial and *E.coli* population were absent in all the treatments for the first day of preparation of liquidized aloe gel juice added with 1 ml, 1.25 ml, 1.5 ml, 1.75 ml and 2 ml of *Gymnema sylvestre* aqueous extract.

The mean fungal population on 30^{th} day of storage was not significant. *E.coli* population was absent on 30^{th} day of storage. However on 30^{th} day of storage a significant increase in bacterial population was noticed. The lowest bacterial population was shown by S_1 (6.40 log cfu ml^{-1}) followed by S_2 (6.57 log cfu ml^{-1}). The highest population was observed in S_5 (6.76 log cfu ml^{-1}) and was on par with S_4 (6.74 log cfu ml^{-1}).

The mean microbial population of liquidized aloe gel juice added with tincture extracts of *Gymnema sylvestre* is presented in table 7. The microbial population was absent during the day of preparation. The mean fungal population increased on storage but was insignificant among different concentration of the tincture extracts of *Gymnema sylvestre* on the 30th day of storage. The mean bacterial population on the 30 th day of storage was the highest for S_{10} (6.39 log cfu ml^{-1}) which was on par with S_9 (6.33 log cfu ml^{-1}) and S_8 (6.30 log cfu ml^{-1}). The least mean bacterial population (6.06 log cfu ml^{-1}) was noticed from liquidized aloe gel juice added with 1ml of tincture extract of *Gymnema sylvestre* (S_6). *E. coli* was absent in all the samples taken even after 30 days of storage.

Microbial count of liquidized aloe gel juice added with different concentrations of *Gymnema sylvestre* decoction extracts are presented in table 8. There was no microbial population on the day of preparation. How ever on storage the mean fungal and bacterial count showed significantly different values on 30th day of storage. The mean fungal population was found to be the lowest for S_{15} (3.06 log cfu ml^{-1}). The mean bacterial population was also the least for S_{15} (6.55 log cfu ml^{-1}). There was an increase in mean fungal and bacterial population in liquidized aloe gel juice added with 1 ml of *Gymnema sylvestre* decoction extract (S_{11}). *E. coli* was absent in all the treatments.

The microbial population of liquidised aloe gel juice with different concentration of *Centella asiatica* aqueous extracts are shown in table 9. There was no microbial population on the day of preparation. On the 30th day of preparation the mean fungal and bacterial population increased and was significantly different among different concentrations of aqueous extracts of *Centella asiatica*.The mean fungal population was the highest for S_{20} (3.71 log cfu ml^{-1}) followed by S_{19} (3.66 log cfu ml^{-1}). The mean fungal and bacterial population was the least for the liquidized aloe gel juice added with 1 ml of aqueous extract of *Centella asiatica* (S_{16}). The mean bacterial population on 30th

day of preparation was significantly higher for S_{20} and was on par with S_{19}. *E coli* population was not observed at the day of preparation or on storage.

Microbial population of liquidized aloe gel juice added with *Centella asiatica* tincture extracts is expressed in table 10. Microbial population was absent on the liquidized aloe gel juice added with different concentration of *Centella asiatica* tincture extracts on the day of preparation. *E coli* was also not observed on the day of preparation as well on storage. The mean fungal and bacterial population in the liquidized aloe gel juice showed significant difference among different concentration of *Centella asiatica* tincture extracts on 30^{th} day of preparation. The mean fungal population noticed was the highest for S_{21} (3.54 log cfu ml^{-1}) and was on par with S_{22} (3.52 log cfu ml^{-1}) and S_{23} (3.37 log cfu ml^{-1}). The least mean fungal population was recorded for S_{25} (0.60 log cfu ml^{-1}). Significantly higher population of bacteria was observed in treatment S_{21} (6.64 log cfu ml^{-1}) which was on par with S_{22} (6.59 log cfu ml^{-1}). Liquidized aloe gel juice added with 2ml of *Centella asiatica* tincture extracts S_{25} (6.15 log cfu ml^{-1}) had the least mean bacterial population.

Different concentration of *Centella asiatica* decoction added to liquidized aloe gel juice produced significant variations in the mean fungal and bacterial population on 30^{th} day of preparation (Table 11).

Mean fungal, bacterial and *E coli* population was absent on the day of preparation. Fungal and bacterial population increased significantly with increase in concentration of *Centella asiatica* decoction extracts. Least fungal and bacterial was observed in S_{26}. The mean fungal population noticed was 3.49 log cfu ml^{-1} and bacterial population is 6.52 log cfu ml^{-1} for S_{26}. *E coli* was absent after 30 days of preparation.

The microbial population of liquidized aloe gel juice with different concentration of *Achyranthes aspera* aqueous extracts is shown in table 12. There was no microbial count on the day of preparation. On the 30^{th} day of preparation the mean fungal and bacterial population was significantly different among the different concentrations of aqueous extracts of *Achyranthes aspera*. The mean fungal population was the highest for S_{31} (3.40 log cfu ml^{-1}) which was on par with S_{32} (3.37 log cfu ml^{-1}) and S_{33} (3.27 log cfu ml^{-1}). The mean fungal (3.00 log cfu ml^{-1}) and bacterial population (6.12 log cfu ml^{-1}) was the least for the liquidized aloe gel juice added with 2 ml of aqueous extract of *Achyranthes aspera* (S_{35}). The mean bacterial population on 30th day of preparation was significantly higher for lower concentration of aqueous extract of *Achyranthes aspera*, S_{31} (6.44 log cfu ml^{-1}) and which was on par with S_{32} (6.37 log cfu ml^{-1}). *E.coli* population was not observed at the day of preparation or on storage.

The mean fungal, bacterial and *E.coli* population were absent in all the treatments for the first day of preparation of liquidized aloe gel juice added with various concentration of *Achyranthes aspera* tincture extract. The mean fungal and bacterial population in the liquidized aloe gel juice showed significant difference among different concentration of *Achyranthes aspera* tincture extracts on the 30th day of storage (Table 13). The mean fungal population noticed was significantly higher for S_{36} (3.30 log cfu ml^{-1}) and S_{37} (3.30 log cfu ml^{-1}). The mean fungal population was the least for S_{39} (3.00 log cfu ml^{-1}) and S_{40} (3.00 log cfu ml^{-1}). Significantly higher population of bacteria was observed in treatments S_{36} and S_{37} as 6.24 log cfu ml^{-1}. Liquidized aloe gel juice added with 2ml of *Achyranthes aspera* tincture extracts had the least bacterial population for S_{40} (6.00 log cfu ml^{-1}) which was on par with S_{39} (6.06 log cfu ml^{-1}) and S_{38} (6.12 log cfu ml^{-1}). *E coli* was absent in the treatments at the time of preparation and after 30 days of storage.

Achyranthes aspera decoction at various concentration added to liquidized aloe gel juice produced significant variation in mean fungal and bacterial population on 30th day of preparation (Table 14). Mean fungal, bacterial and *E coli* population was absent on the day of preparation. The mean fungal population noticed was the highest for S_{45} (3.44 log cfu ml^{-1}) which was on par with S_{42} (3.41 log cfu ml^{-1}), S_{44} (3.41 log cfu ml^{-1}) and S_{41} (3.37 log cfu ml^{-1}). Mean bacterial population for S_{45} treatment was 6.58 log cfu ml^{-1} which was on par with S_{42} (6.53 log cfu ml^{-1}) and S_{44} (6.53 log cfu ml^{-1}). The least bacterial population was observed in S_{43} which was 6.12 log cfu ml^{-1}. The mean fungal population was also the least for S_{43} (3.12 log cfu ml^{-1}). *E coli* was absent in the 30^{th} day of storage.

Microbial population of *Tridax procumbens* aqueous extract added to liquidized aloe gel juice at concentrations of 1, 1.25, 1.5, 1.75 and 2 ml and kept for 30 days is presented in table 15. The mean fungal and bacterial population on 30^{th} day of storage was significantly different among all concentrations. The highest fungal population was noticed in S_{46} (3.93 log cfu ml^{-1}) and the least was noticed in S_{50} (3.66 log cfu ml^{-1}). *E. coli* population was absent on the day of preparation as well as on 30^{th} day of storage. A significant increase in bacterial population was noticed on 30^{th} day of storage. The lowest bacterial population was observed in S_{49} (6.06 log cfu ml^{-1}) and S_{50} (6.06 log cfu ml^{-1}).The highest bacterial population was observed in S_{46} (6.35 x 10^6 log cfu ml^{-1}) which was on par with S_{47} (6.34 log cfu ml^{-1}) and S_{48} (6.31 log cfu ml^{-1}).

The mean microbial population of liquidized aloe gel juice added with tincture extracts of *Tridax procumbens* is presented in table 16. The microbial population was absent during the day of preparation. The mean fungal population was non significant among different concentration of the tincture extracts of *Tridax procumbens* on the 30^{th} day of storage. The mean bacterial population on the 30^{th} day of storage was the highest for S_{51} (6.06 log cfu ml^{-1}) and was on par with S_{52} (6.00 log cfu ml^{-1}) and S_{53} (3.60 log cfu ml^{-1}).No bacterial population was noticed from liquidized aloe gel juice added with 2ml of tincture extract of *Tridax procumbens* (S_{55}) even after 30 days of storage which was on par with S_{54} (1.20 log cfu ml^{-1}). *E coli* was absent in all the samples on the day of preparation or even after 30 days of storage.

Microbial count of liquidized aloe gel juice added with different concentration of *Tridax procumbens* decoction extracts are presented in table 17. There was no microbial population on the day of preparation. However on storage the mean fungal and bacterial count was significantly different. The mean fungal population was found to be the lowest for S_{56} and S_{57} as 3.18 log cfu ml^{-1}which was on par with S_{58} (3.30 log cfu ml^{-1}). The mean bacterial population was also the least for S_{56} (6.12 log cfu ml^{-1}) which was on par with S_{57} (6.28 log cfu ml^{-1}).There was an increase in mean fungal and bacterial population in liquidized aloe gel juice added with increased concentration of *Tridax procumbens* decoction extract (S_{60}). *E. coli* was absent in all the treatments on storage.

The microbial population of liquidized aloe gel juice with different concentration of *Terminalia chebula* aqueous extracts is shown in table 18. There was no microbial count on the day of preparation. On the 30th day of preparation the mean fungal and bacterial population increased and was significantly different among different concentrations of aqueous extracts of *Terminalia chebula*.

The mean fungal population was the highest for S_{65} (3.82 log cfu ml^{-1}) which was on par with S_{64} (3.75 log cfu ml^{-1}) .The mean fungal (3.57 log cfu ml^{-1}) and bacterial population (6.82 log cfu ml^{-1}) was the least for the liquidized aloe gel juice added with 1 ml of aqueous extract of *Terminalia chebula* (S_{61}). The mean bacterial population on 30th day of preparation was significantly higher for S_{65} (7.02 log cfu ml^{-1}) and was on par with S_{64} (6.99 log cfu ml^{-1}) and S_{63} (6.98 log cfu ml^{-1}). *E. coli* population was not observed at the day of preparation and also during storage.

The mean fungal, bacterial and *E.coli* population were absent in all the treatments for the first day of preparation of liquidized aloe gel juice added with 1, 1.25, 1.5, 1.75 and 2 ml of *Terminalia chebula* tincture extracts (Table 19). The mean fungal and bacterial population in the liquidized aloe gel juice showed significant difference among different concentration of *Terminalia chebula* tincture extracts. The mean fungal population noticed was the highest for S_{70} (3.00 log cfu ml^{-1}). No fungal population was recorded in liquidized aloe gel juice added with 1 ml of tincture extract of *Terminalia chebula,* S_{66} (0.00 log cfu ml^{-1}) which was on par S_{67} (0.60 log cfu ml^{-1}), S_{68} (0.60 log cfu ml^{-1}) and S_{69} (1.20 log cfu ml^{-1}). Significantly higher population of bacteria was observed in treatment S_{70} (6.12 log cfu ml^{-1}) which was on par with S_{69} (6.06 log cfu ml^{-1}), S_{68} (6.00 log cfu ml^{-1}) and S_{67} (4.80 log cfu ml^{-1}). Liquidized aloe gel juice added with 2 ml of

Terminalia chebula tincture extracts had the least bacterial population for S_{66} (2.40 log cfu ml^{-1}).

Different concentration of *Terminalia chebula* decoction added to liquidized aloe gel juice produced significant variation in mean fungal and bacterial population on 30^{th} day of storage (Table 20). Mean fungal, bacterial and *E coli* population was absent on the day of preparation. The mean fungal population noticed was the highest for S_{75} (3.73 log cfu ml^{-1}) which was on par with S_{74} (3.72 log cfu ml^{-1}). Mean bacterial population showed highest value for S_{75} (6.90 log cfu ml^{-1}) which was on par with S_{74} (6.87 log cfu ml^{-1}) and S_{73} (6.84 log cfu ml^{-1}). The least fungal and bacterial population was observed in S_{71} which was 3.53 log cfu ml^{-1} and 6.49 log cfu ml^{-1} respectively.

The mean fungal, bacterial and *E. coli* population were absent in all the treatments on the first day of preparation of liquidized aloe gel juice added with different concentration of *Punica granatum* aqueous extract prepared from rind (Table 21). The mean fungal and bacterial population on 30^{th} day of storage was significantly different among all concentrations. The highest fungal population was noticed in S_{80} (3.62 log cfu ml^{-1}) which was on par with S_{79} (3.55 log cfu ml^{-1}) and S_{78} (3.53 log cfu ml^{-1}) and the least was noticed in S_{76} (3.37 log cfu ml^{-1}) which was on par with S_{77} (3.43 log cfu ml^{-1}). However on 30^{th} day of storage a significant increase in bacterial population was noticed. The lowest bacterial population was shown by S_{76} (6.28 log cfu ml^{-1}). The highest population was observed in S_{80} (6.66 log cfu ml^{-1}) which was on par with S_{79} (6.62 log cfu ml^{-1}) and S_{78} (6.53 log cfu ml^{-1}). *E. coli* population was absent on 30^{th} day of storage.

The mean microbial population of liquidized aloe gel juice added with different concentration of tincture extracts of *Punica granatum* prepared from rind is presented in table 22. The microbial population was absent during the day of preparation. The mean fungal population on the 30^{th} day of storage was the highest for S_{81} (3.12 log cfu ml^{-1}) which was on par with S_{82} (3.06 log cfu ml^{-1}) and S_{83} (2.40 log cfu ml^{-1}). The least mean fungal population of (0.60 log cfu ml^{-1}) was noticed from liquidized aloe gel juice added with 2 ml of tincture extract of

Punica granatum (S_{85}) which was on par with S_{84} (1.20 log cfu ml^{-1}). The mean bacterial population was non significant among different concentration of the tincture extracts of *Punica granatum* on the 30^{th} day of storage. *E coli* was absent in all the samples taken even after 30 days of storage.

Microbial count of liquidized aloe gel juice added with different concentration of *Punica granatum* decoction extracts prepared from rind are presented in table 23. There was no microbial population on the day of preparation. However on storage the mean fungal count was significantly different on 30^{th} day of storage. The mean fungal population was found to be the lowest for S_{86} (3.37 log cfu ml^{-1}) which was on par with S_{87} (3.43 log cfu ml^{-1}) and S_{88} (3.49 log cfu ml^{-1}). There was an increase in mean fungal population in the liquidized aloe gel juice added with 2 ml of *Punica granatum* decoction extract prepared from rind (S_{90}). The mean bacterial population was non significant among different concentrations of the decoction extracts of *Punica granatum* prepared from rind on the 30^{th} day of storage. *E coli* was absent in all the treatments.

The microbial population of liquidized aloe gel juice with different concentration of green tea aqueous extracts is shown in table 24. There was no microbial count on the day of preparation. On the 30^{th} day of storage, the mean fungal and bacterial population increased and was significantly different among different concentrations of aqueous extracts of green tea. The mean fungal population was the highest for S_{91} (3.41 log cfu ml^{-1}) which was on par with S_{92} (3.31log cfu ml^{-1}). Least mean fungal (3.06 log cfu ml^{-1}) and bacterial population (6.55 log cfu ml^{-1}) was observed in the liquidized aloe gel juice added with 2 ml of aqueous extract of green tea (S_{95}). The mean bacterial population on 30^{th} day of preparation was significantly higher for S_{91} (6.87 log cfu ml^{-1}) which was on par with S_{92} (6.83 log cfu ml^{-1}). *E coli* population was not observed at the day of preparation or on storage.

The mean fungal, bacterial and *E coli* population were absent in all the treatments for the first day of preparation of liquidized aloe gel juice added with various concentration of green tea tincture extracts.

The mean fungal and bacterial population in the liquidized aloe gel juice showed significant difference among different concentration of green tea tincture extracts (Table 25). The mean fungal population noticed was the highest for S_{100} (3.60 log cfu ml^{-1}). The least fungal population was recorded in S_{96} (3.24 log cfu ml^{-1}) followed by S_{97} (3.37log cfu ml^{-1}). Significantly higher population of bacteria was observed in treatment S_{100} (6.53 log cfu ml^{-1}) which was on par with S_{99} (6.50 log cfu ml^{-1}). Liquidized aloe gel juice added with 1 ml of green tea tincture extracts had the least bacterial population for S_{96} (6.34 log cfu ml^{-1}).

Green tea decoction extracts of varying concentration added to liquidized aloe gel juice produced significant variation in mean fungal and bacterial population on 30th day of preparation (Table 26). Mean fungal, bacterial and *E coli* population was absent on the day of preparation. The mean fungal population noticed was the highest for S_{101} (3.79 log cfu ml^{-1}) which was on par with S_{102} (3.76 log cfu ml^{-1}) and S_{103} (3.73 log cfu ml^{-1}). Mean bacterial population was the highest for S_{101} (6.86 log cfu ml^{-1}) which was on par with S_{102} (6.81 log cfu ml^{-1}). The least fungal and bacterial population was observed in S_{105}. The mean fungal population noticed was 3.64 log cfu ml^{-1} and bacterial population is 6.66 log cfu $ml^{-1}$for $S_{105.}$

Microbial population of the liquidized aloe gel juice added with different concentration (1 ml, 1.25 ml, 1.50 ml, 1.75 ml, and 2 ml) of essential oil of basil oil, lemon grass oil cinnamon grass oil, clove oil and cardamom oil did not show any microbial count after 30 days of storage. Hence the minimum concentration of each of the essential oil was taken for the further study. The absence of the microbial population with minimum concentration of essential oil is presented in table 27.

Ethanolic extracts of *Aloe vera* showed advanced antibacterial property than aqueous extracts (Choi and Chung, 2003). The study supported preliminary results that most of the tincture extracts shows less microbial population. The gel stabilization process of *Aloe vera* included admixing heated gel with an antioxidant and adjusting the pH to a range of 3-3.5 followed by cooling (Rezaei *et al*., 2003). According to Holdsworth (2004) to maximize the quality, ensure microbial safety to prevent the undesirable quality losses. The biological activity of aloe gel remained intact when the gel was heated at 65 ^{0}C for periods less than 15 min, extended periods and higher temperatures resulted in reduced biological activity. So best method suggested for pasteurization was HTST (High Temperature Shot Time), followed by flash cooling to 5 ^{0}C or below (Aloecorp, 2012). Gel stabilization and sterilization are the different processing techniques suggested to reduce the degradation (Ramachandra and Rao, 2008). These studies support the present study that due to pasteurization the microbial population was less during preliminary studies. Medicinal plants could be subjected to a wide range of microbial contamination during pre and post harvest stages and can harbor high microbial loads (Bhowmik *et al*., 2016). This supports the presence of microbial count after 30th day of storage in samples added with herbal extracts.

The ethanolic extract of *Gymnema sylvestre* against *Bacillus pumilus*, *B. subtilis*, *P. aeruginosa* and *S. aureus* showed promising antimicrobial effect (Satdive *et al*., 2003). According to *Saumendu et al.* (2010), the antibiotic and antimicrobial activity of different extracts of *Gymema sylvestre* was determined against a number of pathogens, namely, *S. aureus*, *E. coli*, *and B. subtilis.*

The most active constituents (essential oils) of many spices having wide spectra of antimicrobial activity due to aromatic phenolic compounds, such as thymol and carvacrol in oregano and thyme, eugenol in clove and cinnamon, and cinnamaldehyde in cinnamon (Karapinar and Aktug, 1987; Beuchat and Golden, 1989). These studies supported the result of absence of microbial population after 30th day in samples added with essential oils.

Table 6. Mean microbial population of liquidized aloe gel juice with different concentrations of *Gymnema sylvestre* aqueous extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_1 (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.6 x 10^3 (1.80)	2.6 x 10^6 (6.40)	0.00 x 10^6 (0.00)
S_2 (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.8 x 10^3 (2.40)	3.8 x 10^6 (6.57)	0.00 x 10^6 (0.00)
S_3 (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1 x 10^3 (3.00)	4.6 x 10^6 (6.66)	0.00 x 10^6 (0.00)
S_4 (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.2 x 10^3 (3.06)	5.6 x 10^6 (6.74)	0.00 x 10^6 (0.00)
S_5 (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.6 x 10^3 (3.18)	5.8 x 10^6 (6.76)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.42	0.03	0.00
CD (0.05)	NS	NS	NS	NS	0.09	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 7. Mean microbial population of liquidized aloe gel juice with different concentrations of *Gymnema sylvestre* tincture extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	* Mean *E. coli* population
S_6 (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.4 x 10^3 (1.20)	1.2 x 10^6 (6.06)	0.00 x 10^6 (0.00)
S_7 (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.8 x 10^3 (2.40)	1.4 x 10^6 (6.12)	0.00 x 10^6 (0.00)
S_8 (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.8 x 10^3 (2.40)	2 x 10^6 (6.30)	0.00 x 10^6 (0.00)
S_9 (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.2 x 10^3 (3.06)	2.2 x 10^6 (6.33)	0.00 x 10^6 (0.00)
S_{10} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.6 x 10^3 (3.18)	2.6 x 10^6 (6.39)	0.00 x 10^6 (0.00)
SEm±	0.00	0.00	0.00	0.50	0.05	0.00
CD (0.05)	NS	NS	NS	NS	0.15	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 8. Mean microbial population of liquidized aloe gel juice with different concentrations of *Gymnema sylvestre* decoction extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{11} (1ml)	0.00 x 10^3 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	4.2 x 10^3 (3.61)	5.4 x 10^6 (6.73)	0.00 x 10^6 (0.00)
S_{12} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	3.2 x 10^3 (3.50)	5 x 10^6 (6.69)	0.00 x 10^6 (0.00)
S_{13} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	3.4 x 10^3 (3.52)	4.8 x 10^6 (6.67)	0.00 x 10^6 (0.00)
S_{14} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	3.2 x 10^3 (3.50)	4.8 x 10^6 (6.67)	0.00 x 10^6 (0.00)
S_{15} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.2 x 10^3 (3.06)	3.6 x 10^6 (6.55)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.03	0.03	0.00
CD (0.05)	NS	NS	NS	0.11	0.10	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 9. Mean microbial population of liquidized aloe gel juice with different concentrations of *Centella asiatica* aqueous extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{16} (1ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	2.6×10^3 (3.40)	4.4×10^6 (6.64)	0.00×10^6 (0.00)
S_{17} (1.25ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	2.6×10^3 (3.39)	6.4×10^6 (6.80)	0.00×10^6 (0.00)
S_{18} (1.50ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	3.2×10^3 (3.48)	7.8×10^6 (6.89)	0.00×10^6 (0.00)
S_{19} (1.75ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	4.6×10^3 (3.66)	8.6×10^6 (6.93)	0.00×10^6 (0.00)
S_{20} (2ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	5.2×10^3 (3.71)	8.8×10^6 (6.94)	0.00×10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.05	0.01	0.00
CD (0.05)	NS	NS	NS	0.14	0.05	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 10. Mean microbial population of liquidized aloe gel juice with different concentrations of *Centella asiatica* tincture extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli population*	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{21} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.6 x 10^3 (3.54)	4.4 x 10^6 (6.64)	0.00 x 10^6 (0.00)
S_{22} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.4 x 10^3 (3.52)	4 x 10^6 (6.59)	0.00 x 10^6 (0.00)
S_{23} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.4 x 10^3 (3.37)	3.4 x 10^6 (6.52)	0.00 x 10^6 (0.00)
S_{24} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.6 x 10^3 (3.15)	2.4 x 10^6 (6.37)	0.00 x 10^6 (0.00)
S_{25} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.2 x 10^3 (0.60)	1.6 x 10^6 (6.15)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.27	0.05	0.00
CD (0.05)	NS	NS	NS	0.80	0.15	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 11. Mean microbial population of liquidized aloe gel juice with different concentrations of *Centella asiatica* decoction extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{26} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.2 x 10^3 (3.49)	3.4 x 10^6 (6.52)	0.00 x 10^6 (0.00)
S_{27} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.8 x 10^3 (3.57)	4.4 x 10^6 (6.64)	0.00 x 10^6 (0.00)
S_{28} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.6 x 10^3 (3.55)	5.6 x 10^6 (6.74)	0.00 x 10^6 (0.00)
S_{29} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	4.6 x 10^3 (3.66)	7.4 x 10^6 (6.88)	0.00 x 10^6 (0.00)
S_{30} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	4.8 x 10^3 (3.68)	7.6 x 10^6 (6.86)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.09	0.02	0.00
CD (0.05)	NS	NS	NS	0.09	0.07	NS

*Figures in parantheses indicate transformed values

* Each value is the mean of five replication

Table 12. Mean microbial population of liquidized aloe gel juice with different concentrations of *Achyranthes aspera* aqueous extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{31} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.6 x 10^3 (3.40)	2.8 x 10^6 (6.44)	0.00 x 10^6 (0.00)
S_{32} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.4 x 10^3 (3.37)	2.4 x 10^6 (6.37)	0.00 x 10^6 (0.00)
S_{33} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2 x 10^3 (3.27)	1.8 x 10^6 (6.24)	0.00 x 10^6 (0.00)
S_{34} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.8 x 10^3 (3.24)	1.8 x 10^6 (6.24)	0.00 x 10^6 (0.00)
S_{35} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1 x 10^3 (3.00)	1.4 x 10^6 (6.12)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.05	0.05	0.00
CD (0.05)	NS	NS	NS	0.15	0.16	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 13. Mean microbial population of liquidized aloe gel juice with different concentrations of *Achyranthes aspera* tincture extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{36} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2 x 10^3 (3.30)	1.8 x 10^6 (6.24)	0.00 x 10^6 (0.00)
S_{37} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2 x 10^3 (3.30)	1.8 x 10^6 (6.24)	0.00 x 10^6 (0.00)
S_{38} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.6 x 10^3 (3.18)	1.4 x 10^6 (6.12)	0.00 x 10^6 (0.00)
S_{39} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1 x 10^3 (3.00)	1.2 x 10^6 (6.06)	0.00 x 10^6 (0.00)
S_{40} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1 x 10^3 (3.00)	0.8 x 10^6 (6.00)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.03	0.05	0.00
CD (0.05)	NS	NS	NS	0.09	0.16	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 14. Mean microbial population of liquidized aloe gel juice with different concentrations of *Achyranthes aspera* decoction extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{41} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.4 x 10^3 (3.37)	2.8 x 10^6 (6.42)	0.00 x 10^6 (0.00)
S_{42} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.6 x 10^3 (3.41)	3.4 x 10^6 (6.53)	0.00 x 10^6 (0.00)
S_{43} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.4 x 10^3 (3.12)	1.4 x 10^6 (6.12)	0.00 x 10^6 (0.00)
S_{44} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.6 x 10^3 (3.41)	3.4 x 10^6 (6.53)	0.00 x 10^6 (0.00)
S_{45} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.8 x 10^3 (3.44)	3.8 x 10^6 (6.58)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.05	0.05	0.00
CD (0.05)	NS	NS	NS	0.15	0.15	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 15. Mean microbial population of liquidized aloe gel juice with different concentrations of *Tridax procumbens* aqueous extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{46} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	8.6 x 10^3 (3.93)	2.4 x 10^6 (6.35)	0.00 x 10^6 (0.00)
S_{47} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	7.4 x 10^3 (3.87)	2.2 x 10^6 (6.34)	0.00 x 10^6 (0.00)
S_{48} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	5.6 x 10^3 (3.75)	2.2 x 10^6 (6.31)	0.00 x 10^6 (0.00)
S_{49} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	5.4 x 10^3 (3.73)	1.2 x 10^6 (6.06)	0.00 x 10^6 (0.00)
S_{50} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	4.6 x 10^3 (3.66)	1.2 x 10^6 (6.06)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.02	0.07	0.00
CD (0.05)	NS	NS	NS	0.05	0.21	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 16. Mean microbial population of liquidized aloe gel juice with different concentrations of *Tridax procumbens* tincture extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{51} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.6 x 10^3 (1.80)	1.2 x 10^6 (6.06)	0.00 x 10^6 (0.00)
S_{52} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.4 x 10^3 (1.20)	1 x 10^6 (6.00)	0.00 x 10^6 (0.00)
S_{53} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.2 x 10^3 (0.60)	0.6 x 10^6 (3.60)	0.00 x 10^6 (0.00)
S_{54} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.2 x 10^3 (0.60)	0.2 x 10^6 (1.20)	0.00 x 10^6 (0.00)
S_{55} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.0 x 10^3 (0.00)	0.0 x 10^6 (0.00)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.60	0.85	0.00
CD (0.05)	NS	NS	NS	NS	2.50	NS

*Figures in parantheses indicate transformed values * Each value is the mean of five replication

Table 17. Mean microbial population of liquidized aloe gel juice with different concentrations of *Tridax procumbens* decoction extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population
S_{56} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.6 x 10^3 (3.18)	1.4 x 10^6 (6.12)	0.00 x 10^6 (0.00)
S_{57} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.6 x 10^3 (3.18)	2 x 10^6 (6.28)	0.00 x 10^6 (0.00)
S_{58} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2 x 10^3 (3.30)	2.4 x 10^6 (6.37)	0.00 x 10^6 (0.00)
S_{59} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.6 x 10^3 (3.41)	3.4 x 10^6 (6.50)	0.00 x 10^6 (0.00)
S_{60} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3 x 10^3 (3.47)	3.6 x 10^6 (6.55)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.06	0.05	0.00
CD (0.05)	NS	NS	NS	0.16	0.16	NS

*****Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 18. Mean microbial population of liquidized aloe gel juice with different concentrations of *Terminalia chebula* aqueous extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population
S_{61} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.8 x 10^3 (3.57)	6.6 x10^6 (6.82)	0.00 x10^6 (0.00)
S_{62} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	4.4 x 10^3 (3.64)	7.6 x10^6 (6.88)	0.00 x10^6 (0.00)
S_{63} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	4.8 x 10^3 (3.68)	9.6 x10^6 (6.98)	0.00 x10^6 (0.00)
S_{64} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	5.8 x 10^3 (3.75)	9.8 x10^6 (6.99)	0.00 x10^6 (0.00)
S_{65} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	6.6 x 10^3 (3.82)	10.4 x10^6 (7.02)	0.00 x10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.03	0.02	0.00
CD (0.05)	NS	NS	NS	0.08	0.05	NS

*Figures in parantheses indicate transformed values

*Each value is the mean of five replication

Table 19. Mean microbial population of liquidized aloe gel juice with different concentrations of *Terminalia chebula* tincture extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population
S_{66} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0 x 10^3 (0.00)	0.4 x 10^6 (2.40)	0.00 x 10^6 (0.00)
S_{67} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.2 x 10^3 (0.60)	0.8 x 10^6 (4.80)	0.00 x 10^6 (0.00)
S_{68} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.2 x 10^3 (0.60)	1 x 10^6 (6.00)	0.00 x 10^6 (0.00)
S_{69} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.4 x 10^3 (1.20)	1.2 x 10^6 (6.06)	0.00 x 10^6 (0.00)
S_{70} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1 x 10^3 (3.00)	1.4 x 10^6 (6.12)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.50	0.85	0.00
CD (0.05)	NS	NS	NS	1.48	2.51	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 20. Mean microbial population of liquidized aloe gel juice with different concentrations of *Terminalia chebula* decoction extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean Fungal population	* Mean Bacterial population	Mean *E. coli* population	*Mean Fungal population	* Mean bacterial population	Mean *E. coli* population
S_{71} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.4 x 10^3 (3.53)	3.2 x 10^6 (6.49)	0.00 x 10^6 (0.00)
S_{72} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	4.2 x 10^3 (3.62)	4.8 x 10^6 (6.68)	0.00 x 10^6 (0.00)
S_{73} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	4.4 x 10^3 (3.64)	7 x 10^6 (6.84)	0.00 x 10^6 (0.00)
S_{74} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	5.2 x 10^3 (3.72)	7.4 x 10^6 (6.87)	0.00 x 10^6 (0.00)
S_{75} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	5.4 x 10^3 (3.73)	8 x 10^6 (6.90)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.03	0.03	0.00
CD (0.05)	NS	NS	NS	0.08	0.10	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 21. Mean microbial population of liquidized aloe gel juice with different concentrations of *Punica granatum* (rind) aqueous extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{76} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.4 x 10^3 (3.37)	2 x 10^6 (6.28)	0.00 x 10^6 (0.00)
S_{77} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.8 x 10^3 (3.43)	2.8 x 10^6 (6.43)	0.00 x 10^6 (0.00)
S_{78} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.4 x 10^3 (3.53)	3.4 x 10^6 (6.53)	0.00 x 10^6 (0.00)
S_{79} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.6 x 10^3 (3.55)	4.2 x 10^6 (6.62)	0.00 x 10^6 (0.00)
S_{80} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	4.2 x 10^3 (3.62)	4.6 x 10^6 (6.66)	0.00 x 10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.04	0.05	0.00
CD (0.05)	NS	NS	NS	0.11	0.14	NS

*****Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 22. Mean microbial population of liquidized aloe gel juice with different concentrations of *Punica granatum* (rind) tincture extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population
S_{81} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.4 x 10^3 (3.12)	2.4 x10^6 (6.37)	0.00 x10^6 (0.00)
S_{82} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.2 x 10^3 (3.06)	2 x10^6 (6.30)	0.00 x10^6 (0.00)
S_{83} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.8 x 10^3 (2.40)	1.4 x10^6 (6.12)	0.00 x10^6 (0.00)
S_{84} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.4 x 10^3 (1.20)	1 x10^6 (6.00)	0.00 x10^6 (0.00)
S_{85} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	0.2 x 10^3 (0.60)	0.8 x10^6 (4.80)	0.00 x10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.50	0.54	0.00
CD (0.05)	NS	NS	NS	1.49	NS	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 23. Mean microbial population of liquidized aloe gel juice with different concentrations of *Punica granatum* (rind) decoction extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	*Mean *E. coli* population
S_{86} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.4 x 10^3 (3.37)	1.6 x10^6 (6.18)	0.00 x10^6 (0.00)
S_{87} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	2.8 x 10^3 (3.43)	2 x10^6 (6.28)	0.00 x10^6 (0.00)
S_{88} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.2 x 10^3 (3.49)	2.4 x10^6 (6.37)	0.00 x10^6 (0.00)
S_{89} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.6 x 10^3 (3.55)	2.2 x10^6 (6.34)	0.00 x10^6 (0.00)
S_{90} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	3.8 x 10^3 (3.58)	2.6 x10^6 (6.41)	0.00 x10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.04	0.06	0.00
CD (0.05)	NS	NS	NS	0.13	NS	NS

*Figures in parantheses indicate transformed values * Each value is the mean of five replication

Table 24. Mean microbial population of liquidized aloe gel juice with different concentrations of green tea aqueous extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population
S_{91} (1ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	2.6×10^3 (3.41)	7.4×10^6 (6.87)	0.00×10^6 (0.00)
S_{92} (1.25ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	2.2×10^3 (3.31)	6.8×10^6 (6.83)	0.00×10^6 (0.00)
S_{93} (1.50ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	1.6×10^3 (3.18)	6.2×10^6 (6.79)	0.00×10^6 (0.00)
S_{94} (1.75ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	1.4×10^3 (3.12)	5.2×10^6 (6.71)	0.00×10^6 (0.00)
S_{95} (2ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	1.2×10^3 (3.06)	3.6×10^6 (6.55)	0.00×10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.07	0.02	0.00
CD (0.05)	NS	NS	NS	0.20	0.07	NS

*****Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 25. Mean microbial population of liquidized aloe gel juice with different concentrations of green tea tincture extracts, log cfu ml^{-1}

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population
S_{96} (1ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	1.8×10^3 (3.24)	2.2×10^6 (6.34)	0.00×10^6 (0.00)
S_{97} (1.25ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	2.4×10^3 (3.37)	2.4×10^6 (6.37)	0.00×10^6 (0.00)
S_{98} (1.50ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	3.2×10^3 (3.50)	2.4×10^6 (6.37)	0.00×10^6 (0.00)
S_{99} (1.75ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	3.6×10^3 (3.55)	3.2×10^6 (6.50)	0.00×10^6 (0.00)
S_{100} (2ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	4×10^3 (3.60)	3.4×10^6 (6.53)	0.00×10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.04	0.04	0.00
CD (0.05)	NS	NS	NS	0.11	0.11	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 26. Mean microbial population of liquidized aloe gel juice with different concentrations of green tea decoction extracts, log cfu ml $^{-1}$

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population
S_{101} (1ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	6.2 x 10^3 (3.79)	7.2 x10^6 (6.86)	0.00 x10^6 (0.00)
S_{102} (1.25ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	5.8 x 10^3 (3.76)	6.4 x10^6 (6.81)	0.00 x10^6 (0.00)
S_{103} (1.50ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	5.4 x 10^3 (3.73)	5.6 x10^6 (6.75)	0.00 x10^6 (0.00)
S_{104} (1.75ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	4.6 x 10^3 (3.66)	5.8 x10^6 (6.76)	0.00 x10^6 (0.00)
S_{105} (2ml)	0.00 x 10^3 (0.00)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	4.4 x 10^3 (3.64)	4.6 x10^6 (6.66)	0.00 x10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.02	0.02	0.00
CD (0.05)	NS	NS	NS	0.06	0.06	NS

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 27. Mean microbial population of liquidized aloe gel juice with concentrations of essential oils, log cfu ml^{-1}

Treatments	1st day			30th day		
	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population	*Mean fungal population	* Mean bacterial population	Mean *E. coli* population
S_{106} (1ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)
S_{111} (1ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)
S_{116} (1ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)
S_{121} (1ml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)
S_{126} (lml)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^3 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)
SEm (±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	NS	NS	NS	NS

*Figures in parantheses indicate transformed values * Each value is the mean of five replication

4.3.1.2 Minimum inhibitory concentration

The minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Gymnema sylvestre* aqueous, tincture and decoction extracts are presented in table 28, 29, 30 respectively. *Aspergillus* sp was found to have 100 per cent inhibition on the aqueous, tincture and decoction extracts of *Gymnema sylvestre* from the sample taken on first day of preparation. During 30^{th} day of observation all the treatments showed zero per cent inhibition.

The minimum inhibitory concentration of liquidized aloe gel juice with aqueous, tincture and decoction extracts of *Centella asiatica* on *Aspergillus* sp are presented in Table 31, 32, 33 respectively. All the treatments of aqueous extracts showed zero per cent inhibition on 30^{th} day of observation. Application of 1 ml, 1.25 ml, 1.50 ml, 1.75 ml of tincture extracts showed zero per cent inhibition while 2 ml of tincture extract had shown 100 per cent inhibition during the 30^{th} day of observation (S_{25}).

The minimum inhibitory concentration of liquidized aloe gel juice added with various concentrations of *Achyranthes aspera* aqueous, tincture and decoction extracts on the day of preparation showed 100 per cent inhibition of *Aspergillus* sp. (Table 34, 35 and 36). The treatments containing aqueous extracts especially S_{31} and S_{32} with 1 ml and 1.25 ml showed zero per centage inhibition on 30^{th} day of storage. The treatment S_{33} and S_{34} showed 11.11 per cent inhibition, while S_{35} with 2 ml has shown 22.22 per cent inhibition. The percentage inhibition of *Aspergillus* sp by liquidized aloe gel juice with different concentration of tincture extracts of *Achyranthes aspera* was zero on 30^{th} day of storage. The liquidized aloe gel juice with different concentration of decoction extracts of *Achyranthes aspera* after 30 days of storage has shown zero per cent inhibition of *Aspergillus* sp for 1 ml, 1.25 ml, 1.75 ml and 2 ml. The minimum inhibitory concentration of liquidized aloe gel juice with 1.5ml of *Achyranthes aspera* decoction extracts stored at 30 days showed 11.11 per cent inhibition of *Aspergillus* sp.

The minimum inhibitory concentration of liquidized aloe gel juice added with different concentration of *Tridax procumbens* aqueous, tincture and decoction extracts from 1^{st} day of preparation and 30^{th} day of storage on *Aspergillus sp.* are presented in table 37, 38 and 39 respectively. The treatments had shown 100 per cent inhibition for 1, 1.25, 1.5, 1.75 and 2 ml of aqueous, tincture and decoction extracts on the day of preparation and zero percentage inhibition on 30^{th} day of storage for all treatments of aqueous, tincture and decoction extracts of *Tridax procumbens* except the treatment S_{50} (11.11per cent) which was 2ml of aqueous extract of *Tridax procumbens* and S_{55} (22.22 per cent) which was 2 ml of tincture extract of *Tridax procumbens* added to liquidized aloe gel juice.

The minimum inhibitory concentration of liquidized aloe gel juice added with 1 ml, 1.25 ml, 1.50 ml, 1.75 ml and 2 ml of aqueous, tincture and decoction extracts of *Terminalia chebula* on *Aspergillus* sp are presented in table 40, 41 and 42 respectively. All the treatments had shown 100 per cent inhibition of *Aspergillus* sp on the samples taken from 1^{st} day of preparation and zero percentage inhibition of *Aspergillus* sp from 30^{th} day of storage.

Studies on the minimum inhibitory concentration of liquidized aloe gel juice added with aqueous, tincture and decoction extracts from the rind of *Punica granatum* on *Aspergillus* sp on the 1^{st} and 30^{th} day of storage are presented in Table 43, 44 and 45 respectively. Samples drawn from the 1^{st} day of preparation for all the treatments showed 100 per cent inhibition on *Aspergillus* sp, but zero percentage inhibition was noticed on all treatments on 30^{th} day of storage.

The minimum inhibitory concentration of liquidized aloe gel juice added with aqueous, tincture and decoction extracts of green tea on *Aspergillus* sp are presented in Table 46, 47, 48 respectively. All the treatments showed zero percentage inhibition on 30^{th} day of observation except the treatment S_{95} which exhibited 66.67 per cent inhibition on *Aspergillus* sp.

The minimum inhibitory concentration of liquidized aloe gel juice added with different concentration of essential oils *viz*., basil oil, lemon grass oil, cinnamon grass oil, clove oil and cardamom oil are presented in table 49. All the essential oils of concentration 1ml, 1.25 ml, 1.5 ml, 1.75 ml and 2 ml at 30th day of storage showed 100 per cent inhibition on *Aspergillus* sp. Hence the minimum concentration of each essential oil was taken for further studies.

The minimum inhibitory concentration, minimum bactericidal concentration and minimum fungicidal concentration of the extracts from *Aloe vera, Aloe volkensii* and *Aloe secundiflora* inhibited growth of *Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumonia, Escherichia coli, Erwinia carotovora, Candida albicans* and *Fusarium oxysporum* and it varied significantly (Waithaka *et al*., 2018).

The minimum inhibitory concentration (MIC) showed ≤ 6.25 μg/ml for almost all test microorganisms and different types of extracts except acetone extract from *Aloe vera* leaves which exhibited MIC ≥ 50 μg/ml (Abakar *et al*., 2017). The minimum inihibitory concentration of samples added with essential oil showed hundred percent inhibition to *Aspergillus* sp which might be due to the presence of phenolic compounds which had antimicrobial activity. However, with herbal extracts varying response was noticed with respect to aqueous, tincture and decoction of herbal extracts. The one with maximum inhibitory properties from each herbal extract was selected for further continuing the experiment.

Table 28. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Gymnema sylvestre* aqueous extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_1 (1ml)	100% inhibition	0% inhibition
S_2 (1.25ml)	100% inhibition	0% inhibition
S_3 (1.50ml)	100% inhibition	0% inhibition
S_4 (1.75ml)	100% inhibition	0% inhibition
S_5 (2ml)	100% inhibition	0% inhibition

Table 29. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Gymnema sylvestre* tincture extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_1 (1ml)	100% inhibition	0% inhibition
S_2 (1.25ml)	100% inhibition	0% inhibition
S_3 (1.50ml)	100% inhibition	0% inhibition
S_4 (1.75ml)	100% inhibition	0% inhibition
S_5 (2ml)	100% inhibition	0% inhibition

Table 30. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Gymnema sylvestre* decoction extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_1 (1ml)	100% inhibition	0% inhibition
S_2 (1.25ml)	100% inhibition	0% inhibition
S_3 (1.50ml)	100% inhibition	0% inhibition
S_4 (1.75ml)	100% inhibition	0% inhibition
S_5 (2ml)	100% inhibition	0% inhibition

Table 31. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Centella asiatica* aqueous extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{16} (1ml)	100% inhibition	0% inhibition
S_{17} (1.25ml)	100% inhibition	0% inhibition
S_{18} (1.50ml)	100% inhibition	0% inhibition
S_{19} (1.75ml)	100% inhibition	0% inhibition
S_{20} (2ml)	100% inhibition	0% inhibition

Table 32. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Centella asiatica* tincture extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{21} (1ml)	100% inhibition	0% inhibition
S_{22} (1.25ml)	100% inhibition	0% inhibition
S_{23} (1.50ml)	100% inhibition	0% inhibition
S_{24} (1.75ml)	100% inhibition	0% inhibition
S_{25} (2ml)	100% inhibition	100% inhibition

Table 33. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Centella asiatica* decoction extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{26} (1ml)	100% inhibition	0% inhibition
S_{27} (1.25ml)	100% inhibition	0% inhibition
cS_{28} (1.50ml)	100% inhibition	0% inhibition
S_{29} (1.75ml)	100% inhibition	0% inhibition
S_{30} (2ml)	100% inhibition	0% inhibition

Table 34. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Achyranthes aspera* aqueous extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{31} (1ml)	100% inhibition	0% inhibition
S_{32} (1.25ml)	100% inhibition	0% inhibition
S_{33} (1.50ml)	100% inhibition	11.11% inhibition
S_{34} (1.75ml)	100% inhibition	11.11% inhibition
S_{35} (2ml)	100% inhibition	22.22 % inhibition

Table 35. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Achyranthes aspera* tincture extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{36} (1ml)	100% inhibition	0% inhibition
S_{37} (1.25ml)	100% inhibition	0% inhibition
S_{38} (1.50ml)	100% inhibition	0% inhibition
S_{39} (1.75ml)	100% inhibition	0% inhibition
S_{40} (2ml)	100% inhibition	0% inhibition

Table 36. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Achyranthes aspera* decoction extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{41} (1ml)	100% inhibition	0% inhibition
S_{42} (1.25ml)	100% inhibition	0% inhibition
S_{43} (1.50ml)	100% inhibition	11.11% inhibition
S_{44} (1.75ml)	100% inhibition	0% inhibition
S_{45} (2ml)	100% inhibition	0% inhibition

Table 37. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Tridax procumbens* aqueous extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{46} (1ml)	100 % inhibition	0% inhibition
S_{47} (1.25ml)	100 % inhibition	0% inhibition
S_{48} (1.50ml)	100 % inhibition	0% inhibition
S_{49} (1.75ml)	100 % inhibition	0% inhibition
S_{50} (2ml)	100 % inhibition	11.11% inhibition

Table 38. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Tridax procumbens* tincture extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{51} (1ml)	100 % inhibition	0 % inhibition
S_{52} (1.25ml)	100 % inhibition	0 % inhibition
S_{53} (1.50ml)	100 % inhibition	0 % inhibition
S_{54} (1.75ml)	100 % inhibition	0 % inhibition
S_{55} (2ml)	100 % inhibition	22.22 % inhibition

Table 39. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Tridax procumbens* decoction extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{56} (1ml)	100 % inhibition	0 % inhibition
S_{57} (1.25ml)	100 % inhibition	0 % inhibition
S_{58} (1.50ml)	100 % inhibition	0 % inhibition
S_{59} (1.75ml)	100 % inhibition	0 % inhibition
S_{60} (2ml)	100 % inhibition	0 % inhibition

Table 40. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Terminalia chebula* aqueous extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{61} (1ml)	100% inhibition	0 % inhibition
S_{62} (1.25ml)	100% inhibition	0 % inhibition
S_{63} (1.50ml)	100% inhibition	0 % inhibition
S_{64} (1.75ml)	100% inhibition	0 % inhibition
S_{65} (2ml)	100% inhibition	0 % inhibition

Table 41. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Terminalia chebula* tincture extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{66} (1ml)	100% inhibition	0 % inhibition
S_{67} (1.25ml)	100% inhibition	0 % inhibition
S_{68} (1.50ml)	100% inhibition	0 % inhibition
S_{69} (1.75ml)	100% inhibition	0 % inhibition
S_{70} (2ml)	100% inhibition	0 % inhibition

Table 42. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Terminalia chebula* decoction extracts

Treatments	*Aspergillus* sp	
	1^{st} day	30^{th} day
S_{71} (1ml)	100% inhibition	0 % inhibition
S_{72} (1.25ml)	100% inhibition	0 % inhibition
S_{73} (1.50ml)	100% inhibition	0 % inhibition
S_{74} (1.75ml)	100% inhibition	0 % inhibition
S_{75} (2ml)	100% inhibition	0 % inhibition

Table 43. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Punica granatum* (rind) aqueous extracts

Treatments	*Aspergillus* sp	
	1st day	30th day
S_{76} (1ml)	100 % inhibition	0 % inhibition
S_{77} (1.25ml)	100 % inhibition	0 % inhibition
S_{78} (1.50ml)	100 % inhibition	0 % inhibition
S_{79} (1.75ml)	100 % inhibition	0 % inhibition
S_{80} (2ml)	100 % inhibition	0 % inhibition

Table 44. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Punica granatum* (rind) tincture extracts

Treatments	*Aspergillus* sp	
	1st day	30th day
S_{81} (1ml)	100 % inhibition	0 % inhibition
S_{82} (1.25ml)	100 % inhibition	0 % inhibition
S_{83} (1.50ml)	100 % inhibition	0 % inhibition
S_{84} (1.75ml)	100 % inhibition	0 % inhibition
S_{85} (2ml)	100 % inhibition	0 % inhibition

Table 45. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of *Punica granatum* (rind) decoction extracts

Treatments	*Aspergillus* sp	
	1st day	30th day
S_{86} (1ml)	100 % inhibition	0 % inhibition
S_{87} (1.25ml)	100 % inhibition	0 % inhibition
S_{88} (1.50ml)	100 % inhibition	0 % inhibition
S_{89} (1.75ml)	100 % inhibition	0 % inhibition
S_{90} (2ml)	100 % inhibition	0 % inhibition

Table 46. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of green tea aqueous extracts

Treatments	*Aspergillus* sp	
	1st day	30th day
S_{91} (1ml)	100 % inhibition	0% inhibition
S_{92} (1.25ml)	100 % inhibition	0% inhibition
S_{93} (1.50ml)	100 % inhibition	0 % inhibition
S_{94} (1.75ml)	100 % inhibition	0 % inhibition
S_{95} (2ml)	100 % inhibition	66.67% inhibition

Table 47. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of green tea tincture extracts

Treatments	*Aspergillus* sp	
	1st day	30th day
S_{96} (1ml)	100 % inhibition	0 % inhibition
S_{97} (1.25ml)	100 % inhibition	0 % inhibition
S_{98} (1.50ml)	100 % inhibition	0 % inhibition
S_{99} (1.75ml)	100 % inhibition	0 % inhibition
S_{100} (2ml)	100 % inhibition	0 % inhibition

Table 48. Minimum inhibitory concentration of liquidized aloe gel juice with different concentrations of green tea decoction extracts

Treatments	*Aspergillus* sp	
	1st day	30th day
S_{101} (1ml)	100 % inhibition	0 % inhibition
S_{102} (1.25ml)	100 % inhibition	0 % inhibition
S_{103} (1.50ml)	100 % inhibition	0 % inhibition
S_{104} (1.75ml)	100 % inhibition	0 % inhibition
S_{105} (2ml)	100 % inhibition	0 % inhibition

Table 49. Minimum inhibitory concentration of liquidized aloe gel juice with concentrations of essential oils

Treatments	*Aspergillus* sp	
	1st day	30th day
S_{106} (1ml)	100 % inhibition	100 % inhibition
S_{111}(1ml)	100 % inhibition	100 % inhibition
S_{116}(1ml)	100 % inhibition	100 % inhibition
S_{121}(1ml)	100 % inhibition	100 % inhibition
S_{126}(lml)	100 % inhibition	100 % inhibition

4.3.2 Final aloe gel stabilization treatments

The best concentrations from aqueous, tincture and decoction of herbal extracts and essential oils (1 ml) from basil, lemon grass, cinnamon bark, clove and cardamom and sodium benzoate (1 ml) as control were selected from the preliminary trials and subjected to final stabilization treatments and stored in room temperature for six months for recording observations on the storage life.

4.3.2.1 Sensory parameters

The sensory parameters included appearance, odour and colour. The appearance of the liquidized aloe gel juice with different forms of herbal extracts and essential oils stored for six months are depicted in table 50.

The mean rank value for the appearance of liquidized aloe gel juice showed significant difference among herbal extracts and essential oils. The highest mean rank value for the first day (644.22) at the time of storage was noticed for liquidized aloe gel juice added with clove oil (1ml), G_{25} which was followed by juice mixed with basil oil, G_{22} (618.43). The mean rank value for appearance of liquidized aloe gel juice stabilized with 1ml of cardamom oil was 615.58 followed by 1 ml of cinnamon bark oil (609.50). Among the herbal extracts, *Terminalia chebula* tincture 1 ml, G_{14} (498.18) was on par with *Centella asiatica* decoction ml), G_3 (262.12) which was followed by (*Achyranthes aspera* aqueous, 2 ml) G_7 (285.94).The least mean rank value was noticed in *Gymnema sylvestre* decoction, 2 ml (262.12). The mean rank value of liquidized aloe gel juice treated with sodium benzoate (control) was 499.82.

The appearance of the liquidized aloe gel juice added with herbal extracts and essential oil during the first month after storage was significantly different. The mean rank value was the highest for liquidized aloe gel juice added with 1 ml clove oil, G_{25} (642.62) followed by 1 ml cardamom oil, G_{26} (614.60). The treatment G_{26} was on par with 1ml basil oil (G_{22}), 1 ml cinnamon bark oil, G_{24} (607.14) and 1 ml lemon grass oil, G_{23} (605.04). The least value for the first month after storage was noticed in G_{19}

(238.00) which were on par with G_3 (241.76). Among the herbal extracts, *Terminalia chebula* tincture 1 ml, G_{14} (493.70) recorded the highest mean rank value. The mean rank value noticed in the control was 487.40.

A significant variation was noticed in the mean rank value of the appearance for the liquidized aloe gel juice treated with herbal extracts and essential oils during second month after storage. The mean rank value for the second month after storage was the highest for liquidized aloe gel juice added with clove oil (1ml), G_{25} (642.00) which was followed by 1 ml basil oil, G_{22} (613.18) and 1 ml cardamom oil, G_{26} (613.12). *Terminalia chebula* tincture 1 ml, G_{14} (487.40) and *Terminalia chebula* decoction 1 ml, G_{15} (470.68) recorded superior mean rank value among herbal extracts and were on par. The least mean rank value among the treatments was noticed in *Terminalia chebula* aqueous 1 ml, G_{13} (236.66) which was on par with green tea aqueous 2 ml, G_{19} (237.00) and *Achyrathes aspera* aqueous 2 ml G_7 (254.86). The mean rank value for the control was 458.24.

Appearance at third month after storage also showed significant difference among the treatments. The highest value was observed for liquidized aloe gel juice added with 1 ml clove oil, G_{25} (635.80) which was followed by 1 ml cardamom oil, G_{26} (611.84). The mean rank value for appearance for the third month after storage was least in green tea aqueous 2 ml, G_{19} (153.08) which was followed by *Gymnema sylvestre* decoction 2 ml, G_3 (204.20).

The mean rank value for appearance of the liquidized aloe gel juice significantly varied among herbal extracts and essential oils during fourth month after storage. The mean rank value for the fourth month after storage was the highest for the treatment with 1 ml clove oil, G_{25} (611.02) which was on par with 1 ml cardamom oil, G_{26} (608.98), 1 ml basil oil, G_{22} (601.34), 1 ml cinnamon bark oil, G_{24} (599.94) and 1 ml lemon grass oil, G_{23} (596.80). The least mean rank value was noticed in green tea aqueous 2 ml, G_{19} (96.64) which were followed by *Achyranthes aspera* aqueous 2 ml, G_7 (150.96). The mean rank value for the control was 446.16.

The appearance of liquidized aloe gel juice added with herbal extracts and essential oils varied significantly during fifth month after storage. The mean rank value for appearance was the highest in liquidized aloe gel juice added with 1ml clove oil, G_{25} (610.60) which was on par with 1 ml cardamom oil, G_{26} (595.66) and 1 ml basil oil, G_{22} (593.90). The least value for the fifth month after storage was noticed in green tea aqueous 2 ml, G_{19} (83.32) which was followed by *Achyranthes aspera* aqueous 2 ml, G_7 (123.00). The mean rank value for the liquidized aloe gel juice added with 1ml of sodium benzoate, G_{27} was 375.46.

During six months after storage the mean rank value of appearance for all the treatments were reduced. A significant difference in appearance was noticed among all treatments at six months after storage. The highest mean rank value among the treatments were noticed in liquidized aloe gel juice added with 1 ml clove oil, G_{25} (545.24) which was followed by 1 ml cardamom oil, G_{26} (454.24). The least value among the treatments in green tea aqueous 2 ml, G_{19} (67.58) which was followed by *Gymnema sylvestre* decoction 2 ml, G_3 (111.40). The mean rank value for the control was 237.56.

The mean rank value of control (liquidized aloe gel juice with 1 ml sodium benzoate) ranged from 499.82 to 237.56 from 0 MAS to 6 MAS.

The colour of the liquidized aloe gel juice with herbal extracts and essential oils stored for six months are presented in table 51.

The mean rank value for the colour of the liquidized aloe gel juice showed significant difference among herbal extracts and essential oils at the time of storage (0 MAS). The mean rank value for the first day was highest for liquidized aloe gel juice added with 1 ml clove oil, G_{25} (648.52) which were followed by 1ml cardamom oil, G_{26} (616.86). The least mean rank value was noticed in *Terminalia chebula* tincture 1 ml, G_4 (235.30) which were followed by green tea aqueous 2 ml, G_{19} (247.16) and *Punica granatum* aqueous 1 ml, G_{16} (254.04).

From the first month after storage the colour of liquidized aloe gel juice added with herbal extracts showed a decreasing trend in mean rank value. Significant difference in the mean rank value was noticed among treatments. The mean rank value for the treatments added with essential oil was higher. The mean rank value for the liquidized aloe gel juice added with 1 ml clove oil,G_{25} was significantly higher (637.76) which was followed by 1 ml cardamom oil, G_{26} (609.36). The mean rank value for the treatment with *Achyranthes aspera* decoction 1.5 ml, G_9 was 474.78 which was the highest among herbal extracts. The least value for the first month after storage was noticed in *Centella asiatica* aqueous 1 ml, G_4 as 221.36 which was on par with *Punica granatum* aqueous 1 ml, G_{16} (240.44) and green tea aqueous 2 ml, G_{19} (230.90). The liquidized aloe gel juice added with 1 ml sodium benzoate, G_{27} had a mean rank value of 429.94.

A significant variation among the treatments were seen in the mean rank values of colour for the liquidized aloe gel juice treated with herbal extracts and essential oils during second month after storage. The mean rank value for the second month after storage was the highest for liquidized aloe gel juice added with1 ml clove oil, G_{25} (632.86) which was followed by 1 ml basil oil, G_{22} (593.62). The colour was the best in *Achyranthes aspera* decoction of 1.5 ml, G_9 among the herbal extracts used (452.66). The least mean rank value among the treatments was seen in *Centella asiatica* aqueous 1 ml, G_4 (185.80) which were on par with green tea aqueous 2 ml, G_{19} (205.00). The mean rank value for colour in control (G_{27}) was 373.90.

Significant variation in mean rank values of colour was observed among the herbal extracts and essential oils added to liquidized aloe gel juice during the third month of storage. The mean rank value for colour was the highest for the treatment of liquidized aloe gel juice added with1 ml clove oil, G_{25} (631.48) which was followed by1 ml cardamom oil, G_{22} (592.56). Among the herbal extracts higher rank value (440.58) was noticed in *Achyranthes aspera* tincture, 2 ml (G_8) which was on par with *Centella asiatica* tincture 2 ml (G_5) with a mean rank value of 431.39. The least value among the treatments for the third month after storage was noticed in *Centella asiatica* aqueous 1 ml, G_4 (109.40) which were followed by green tea aqueous 2 ml, G_{19} (169.64). The mean rank value for colour in control was 306.02.

The mean rank value for the colour of liquidized aloe gel juice added with herbal extracts and essential oils showed significant variance during fourth month after storage. The mean rank value in fourth month of storage for liquidized aloe gel juice added with essential oil ranged from 541.72 in 1 ml lemon grass oil to 1 ml clove oil, G_{25} (621.16) which was the highest followed by 1 ml cardamom oil G_{26} (570.24) and 1 ml basil oil (543.10). The least mean rank value was noticed in *Centella asiatica* aqueous 1 ml, G_4 (100.38) which were on par with green tea aqueous 2 ml, G_{19} (102.64).

After fifth month of storage significant difference among the treatments was observed. The highest value was observed in liquidized aloe gel juice added with 1 ml clove oil, G_{25} (594.04) which was followed by 1 ml cardamom oil G_{26} (539.74). The mean rank value for colour was the highest for *Centella asiatica* tincture 2 ml, G_5 (393.40) among the herbal extracts added and the least value was observed for green tea aqueous 2 ml, G_{19} (42.92) which was followed by G_4 (89.44). The mean rank value for the colour in control was 237.78.

The sixth month after storage showed a substantial reduction in the mean rank value of all the treatments compared to zero MAS. The best treatment as revealed by the highest mean rank value among the treatments was noticed in liquidized aloe gel juice added with1ml clove oil, G_{25} (592.90) which was followed by 1 ml cardamom oil, G_{26} (520.84). Among herbal extracts the mean rank value was higher for *Achyranthes aspera* tincture 2 ml, G_8 (345.24). The least value among the treatments was observed in green tea aqueous 2 ml, G_{19} (34.48) which were on par with *Centella asiatica* aqueous 1 ml, G_4 (53.24). The mean rank value for the control was 216.76.

The mean rank value of liquidized aloe gel juice added with control (sodium benzoate) ranged from 444.26 to 216.76 from 0 MAS to 6 MAS.

The odour of the liquidized aloe gel juice with herbal extracts and essential oils stored for the six months is presented in table 52.

The mean rank value for the odour of the liquidized aloe gel juice added with herbal extracts and essential oils at the day of preparation (0 MAS) showed significant difference among the treatments. The mean rank value for the first day was significantly highest for liquidized aloe gel juice added with 1ml of clove oil, G_{25} (650.62). The liquidized aloe gel juice added with other essential oils 1 ml basil oil, G_{22} (621.94), 1 ml cardamom oil, G_{26} (621.42) 1 ml lemon grass oil, G_{23} (619.58) and 1ml cinnamon bark oil, G_{24} (609.78) were on par. The highest value among the herbal extracts was noticed in *Tridax procumbens* tincture 2 ml, G_{11} (411.94) which was followed by liquidized aloe gel juice with green tea tincture extract 1 ml , G_{20} (387.34) and 2 ml of *Centella asiatica* tincture, G_5 (386.38). However, least mean rank value was noticed in *Terminalia chebula* decoction 1 ml, G_{15} (276.48) which was on par with G_{16} (*Punica granatum* aqueous 1ml), green tea aqueous 2 ml, G_{19} (295.92), *Punica granatum* decoction 1 ml, G_{18} (289.92), *Punica granatum* aqueous 1ml ,G_{16} (279.82), *Terminalia chebula* aqueous 1 ml, G_{13} (281.68), *Tridax procumbens* decoction 1 ml, G_{12} (295.92), *Gymnema sylvestre* decoction 2 ml, G_3 (289.90) and *Gymnema sylvestre* aqueous 1 ml, G_1 (293.96). The mean odour rank value of control (G_{27}) was 512.92.

From the first month after storage onwards significant difference among the mean rank value of odour was getting reduced. The highest mean rank value was noticed in G_{25} (636.68) which was on par with G_{26} (620.80). The least value for the first month after storage was noticed in G_{16} (265.78) which was on par with G_1 (285.64) G_3 (281.68) G_9 (281.68) G_{10} (285.08) G_{12} (279.70) G_{13} (267.66), G_{15} (274.24) and G_{18} (279.82). Among herbal extracts highest value was noticed in G_{11}

(405.28) with *Tridax procumbens* tincture 2 ml followed by G_5 (373.90). The mean odour rank value for the control was 502.32.

A significant variation among the treatments were noticed in the mean rank value of the odour for the liquidized aloe gel juice treated with herbal extracts and essential oils during second month after storage. The mean rank value for the second month after storage was highest for G_{25} (636.34) liquidized aloe gel juice added with 1 ml clove oil which were on par with G_{26} ,1 ml cardamom oil (620.74). The mean odour rank value in the treatment G_{22}, 1 ml basil oil was 611.80 while for 1 ml lemon grass oil and1 ml cinnamon bark oil was 608.16 and 603.50 respectively.The least mean rank value among the treatments was seen for G_3 (240.16) which was on par with G_{13} (253.16), G_{15} (253.64) and G_{16} (252.56). Among the herbal extracts the highest value was noticed in G_1, *Tridax procumbens* tincture 2ml (394.90) added with liquidized aloe gel juice followed by $G_{5,}$ *Centella asiatica* tincture with 2ml (365.20). The mean rank value for odour was 401.16 for control which was lower than essential oils but higher than herbal extracts.

The mean rank value of odour for third month after storage also showed significant variation among the treatments. The highest rank value for odour was observed in $G_{25,}$ 1ml clove oil (630.64) added with liquidized aloe gel juice which was on par with G_{26}, 1ml cardamom oil (620.30). The least mean rank value was noticed for G_3 (221.48) which were on par with G_{13} (241.72). G_5, *Centella asiatica* tincture 2ml (349.76) showed highest mean rank for odour among the treatments with herbal extracts followed by G_2, *Gymnema sylvestre* tincture 1 ml (320.30).The mean rank value of odour for control was 346.56 which was lower than essential oils but higher than herbal extracts.

Liquidized aloe gel juice added with essential oils and herbal extracts showed significant variation among the treatments during fourth month after storage. The highest mean rank value after fourth month of storage was noticed in liquidized aloe gel juice added with1ml clove oil, G_{25} (622.42) which was on par with 1ml cardamom oil, G_{26} (617.68) and 1ml cinnamon bark oil, G_{24} (602.88). The least value was noticed during third month after storage in G_3 (207.62) which was on par with G_9 (219.84), G_{12} (219.84) and G_{16} (229.20). Highest value among the herbal extracts was noticed in G_2 *Gymnema sylvestre* tincture, 1ml (312.02) which was on par with G_5 (305.56). The mean rank value of odour for control was 263.40.

The mean rank of odour after the fifth month of storage also showed significant variation among the treatments. The mean rank value of odour was the highest in clove oil 1 ml, G_{25} (620.30) which was on par with G_{26} cardamom oil, 1 ml (605.90) and G_{24} cinnamon bark oil, 1 ml (600.06). The least value after fifth month of storage was noticed in G_7 (177.62) which were on par with G_9 (186.22) and G_{16} (229.20). Highest value among the herbal extracts was noticed in G_5 (285.30) which was on par with G_2 (281.44), G_1 (270.54), G_8 (279.82) and G_{19} (270.82).

During the sixth month after storage the mean rank value of odour for all the treatments was significantly different. The mean rank value of odour observed was higher amongst essential oil treatments and this was significantly higher in liquidized aloe gel juice added with, 1 ml clove oil G_{25} (593.70) which was followed by 1 ml cardamom oil, G_{26} (528.06), 1ml basil oil, G_{22} (430.46), 1ml lemon grass oil, G_{23} (404.16) and 1 ml cinnamon bark oil, G_{24} (404.16). The least value among the treatments was observed in G_{16} (145.64) which were on par with G_{15} (165.68). Highest value among herbal extracts was noticed in G_5 (281.68) which were on par with G_8 (277.80). As for control the mean rank value for odour was 314.26.

Jairath *et al*. (2015) observed a declining trend in colour and appearance in treated liquidized aloe gel juice and flavour scores were significantly higher in treated products than control throughout the study period. In the present study odour or flavor of the treated samples showed a decreasing trend. Suraiti *et al*. (2019) reported that the time of storage, temperature and interaction had real effect to the colour of the aloe gel, the study supported the present result that the colour, taste and odour of the samples decreased by the increase of storage time.

Femenia *et al*. (1999) determined higher glucose content compared to fructose and found 26.7 g glucose/100 g dry matter in *Aloe vera* gel compared to 0.64 g fructose/100 g dry matter. Bozzi *et al*. (2007) studied the quality and authenticity of commercial *Aloe vera* gel powders and reported 11.85 and 5.3 g/100g dry matter of free glucose and free fructose respectively in fresh *Aloe vera* gel. Miranda *et al*. (2009) reported that the *Aloe vera* gel showed water activity (a_w) greater than 0.98, which means that the gel is very susceptible to attack of moulds. According to Scala *et al*. (2013) two of the main sugars present in aloe gel stored after high hydrostatic pressure processing were fructose and glucose (9.3 and 25.2 g/100 g dry matter, respectively). According to Mulay and Ogale (2018) during the preparation of *Aloe vera* leaf juice care has to be taken to preserve the content of acetylated polysaccharides which readily degrade during prolonged storage, bacterial fermentation and elevated temperature. Quality of commercial *Aloe vera* juice is strongly dependent on processing and storage conditions. Enzymatic - thermal degradation and bacterial fermentation may affect the quality and decrease the value of final product. IASC ensures presence of acetylated polysaccharides at or above a minimum level (≥ 5% dry weight). The degradation of polysaccharides of aloe gel may result in the change in appearance, colour and odour on storage. The use of herbal extracts has to an extent resulted in varying intensities of polysaccharide degradation as well as microbial contamination which changed the appearance, colour and odour of the liquidized aloe gel juice. Analysis on the effect of essential oil on the liquidized aloe gel juice revealed better appearance, colour and odour for samples added with 1 ml clove oil followed by those mixed with 1 ml cardamom oil at six month of storage. The antioxidant activity of clove oil due to phenolic compounds might have prevented the microorganisms to make changes in appearance, colour and odour during storage thus giving a comparatively stable product.

Table 50. Effect of herbal extracts and essential oils on appearance of liquidized aloe gel juice

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	7.12 (345.10)	4.16 (333.44)	2.44 (317.50)	2.04 (272.98)	1.76 (245.26)	1.72 (195.54)	1.64 (159.62)
G_2 (*Gymnema sylvestre* tincture 1ml)	6.88 (409.98)	4.8 (387.40)	2.96 (362.24)	2.52 (322.08)	2.52 (281.32)	2.32 (248.10)	2.16 (137.36)
G_3 (*Gymnema sylvestre* decoction 2ml)	6.48 (262.12)	3.12 (241.76)	2.36 (236.66)	2.28 (204.20)	1.92 (200.36)	1.72 (167.08)	1.64 (111.40)
G_4 (*Centella asiatica* aqueous 1ml)	7.56 (420.48)	3.8 (296.94)	2.68 (274.06)	2.24 (262.22)	1.88 (209.30)	1.76 (204.80)	1.76 (191.70)
G_5 (*Centella asiatica* tincture 2ml)	7.16 (414.56)	5.08 (390.14)	3.4 (382.70)	3.24 (315.06)	2.04 (299.28)	1.92 (262.22)	1.56 (228.40)
G_6 (*Centella asiatica* decoction 1ml)	8.24 (494.40)	4.08 (454.48)	2.68 (280.16)	2.32 (245.18)	1.96 (235.66)	1.80 (212.68)	1.6 (209.02)
G_7 (*Achyranthes aspera* aqueous 2 ml)	6.84 (285.94)	3.28 (280.16)	2 (254.86)	2 (238.14)	1.92 (150.96)	1.80 (123.00)	1.68 (121.50)
G_8 (*Achyranthes aspera* tincture 2ml)	7.16 (399.20)	4.44 (341.76)	2.92 (323.32)	2.80 (286.98)	2.44 (274.06)	2 (248.54)	1.76 (229.56)
G_9 (*Achyranthes aspera* decoction 1.50 ml)	7 (308.36)	4.56 (306.44)	2.84 (279.30)	2.72 (253.70)	2 (246.60)	1.64 (232.52)	1.56 (226.06)
G_{10} (*Tridax procumbens* aqueous 2ml)	7.76 (365.24)	3.88 (344.00)	2.76 (270.56)	2.44 (238.14)	1.92 (231.26)	1.76 (218.80)	1.48 (209.20)
G_{11} (*Tridax procumbens* tincture 2ml)	7.36 (411.52)	4.28 (386.50)	2.72 (318.30)	2.44 (310.30)	2.36 (275.20)	2.32 (212.76)	2.00 (212.42)
G_{12} (*Tridax procumbens* decoction 1ml)	7.2 (353.10)	4.32 (271.72)	2.88 (267.24)	2.40 (263.38)	1.76 (238.78)	1.72 (226.34)	1.64 (180.72)
G_{13} (*Terminalia chebula* aqueous 1ml)	7.16 (345.36)	4.32 (271.84)	3.36 (236.66)	2.64 (234.58)	1.92 (218.80)	1.6 (194.78)	1.52 (173.08)
G_{14} (*Terminalia chebula* tincture 1ml)	7.96 (498.18)	5.76 (493.70)	4.72 (487.40)	3.32 (387.46)	2.12 (322.54)	2.04 (278.52)	1.8 (277.74)
G_{15} (*Terminalia chebula* decoction 1ml)	6.80 (388.50)	5 (375.70)	4.6 (470.68)	3.48 (413.94)	3.32 (321.12)	1.68 (253.78)	1.68 (254.86)

Table 50. Effect of herbal extracts and essential oils on appearance of liquidized aloe gel juice (continued)

G_{16} (*Punica granatum* aqueous 1ml)	7.08 (347.22)	4.28 (264.14)	3.60 (263.62)	3.56 (263.18)	1.88 (238.00)	1.52 (229.62)	1.6 (217.80)
G_{17} (*Punica granatum* tincture 2ml)	6.6 (349.88)	4.8 (327.42)	3.56 (303.10)	2.68 (273.36)	2.36 (268.14)	1.92 (219.86)	1.76 (199.62)
G_{18} (*Punica granatum* decoction 1ml)	7.36 (330.26)	4.40 (324.10)	3.44 (281.96)	2.68 (280.50)	2.16 (228.40)	1.56 (160.20)	1.28 (196.28)
G_{19} (green tea aqueous 2ml)	5.44 (287.78)	2.84 (238.00)	2.36 (237.00)	1.72 (153.08)	1.68 (96.64)	1.68 (83.320)	1.6 (67.58)
G_{20} (green tea tincture 1ml)	7.92 (476.24)	5.48 (452.30)	4.6 (346.66)	3.72 (329.64)	2.36 (260.16)	2.16 (229.96)	1.72 (184.46)
G_{21} (green tea decoction 2ml)	7.48 (374.04)	3.76 (335.58)	3.2 (272.96)	2.28 (254.86)	2.08 (198.60)	1.68 (177.96)	1.44 (160.72)
G_{22} (basil oil 1ml)	7.28 (618.43)	6.8 (613.94)	6.70 (613.18)	6.16 (606.32)	6.16 (601.34)	5.68 (593.90)	5.56 (294.88)
G_{23} (lemon grass oil 1ml)	7.72 (605.50)	6.44 (605.04)	6.20 (602.86)	5.96 (597.10)	5.80 (596.80)	5.60 (539.78)	5.44 (453.44)
G_{24} (cinnamon bark oil 1ml)	7 (609.70)	6.44 (607.14)	6.36 (607.12)	5.88 (605.68)	5.76 (599.94)	5.76 (566.62)	5.68 (308.36)
G_{25} (clove oil 1ml)	6.96 (644.22)	6.92 (642.62)	6.76 (642.00)	6.60 (635.80)	6.56 (611.02)	6.52 (610.60)	6.52 (545.24)
G_{26} (cardamom oil 1ml)	6.96 (615.58)	6.56 (614.60)	6.44 (613.12)	6.16 (611.84)	5.8 (608.98)	5.72 (595.66)	5.72 (454.24)
G_{27} (control - sodium benzoate 1ml)	6.68 (499.82)	5.8 (487.40)	4.72 (458.24)	3.76 (456.90)	3.12 (446.16)	2.80 (375.46)	2.28 (237.56)
KW value	207.58	456.37	518.92	472.59	405.12	403.77	393.38
χ^2	38.89						
CD value	21.67						

*Figures in parenthesis indicates mean rank value

Table 51. Effect of herbal extracts and essential oils on colour of liquidized aloe gel juice

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	7.28 (480.18)	5.12 (469.2)	5.6 (409.14)	5 (317.50)	3.88 (308.80)	2 (211.82)	1.48 (210.16)
G_2 (*Gymnema sylvestre* tincture1ml)	7.52 (459.90)	5.88 (441.74)	5.08 (395.84)	4.8 (370.40)	3.16 (363.80)	2.32 (329.42)	2.04 (323.10)
G_3 (*Gymnema sylvestre* decoction 2ml)	6.36 (475.52)	4.8 (297.66)	4.32 (280.60)	3.92 (273.28)	3.76 (255.80)	1.76 (244.70)	1.64 (231.48)
G_4 (*Centella asiatica* aqueous 1ml)	6.72 (235.30)	3.72 (221.36)	2.88 (185.80)	1.84 (109.40)	1.6 (100.30)	1.52 (89.44)	1.4 (53.14)
G_5 (*Centella asiatica* tincture 2ml)	6.28 (487.32)	5.8 (436.48)	5.32 (433.76)	4.6 (431.39)	3.56 (425.94)	2.36 (393.40)	2. 08 (311.52)
G_6 (*Centella asiatica* decoction 1ml)	6.76 (409.20)	5.36 (400.84)	5.16 (354.88)	3.76 (328.10)	3.36 (297.26)	1.72 (249.64)	1.4 (195.40)
G_7 (*Achyranthes aspera* aqueous 2 ml)	6.32 (274.68)	4.48 (266.62)	4.28 (256.86)	3.48 (254.42)	1.96 (245.40)	1.84 (202.92)	1.68 (167.24)
G_8 (*Achyranthes aspera* tincture 2ml)	7.28 (488.06)	6.12 (469.46)	5.52 (450.90)	4.68 (440.58)	3 (369.82)	2.36 (350.82)	2.32 (345.24)
G_9(*Achyranthes aspera* decoction 1.50 ml)	6.6 (493.26)	6.04 (474.78)	5.68 (452.66)	4.76 (391.72)	3.28 (308.30)	1.68 (242.76)	1.6 (240.44)
G_{10} (*Tridax procumbens* aqueous 2ml)	6.52 (289.04)	4.36 (285.10)	4.16 (256.52)	3.2 (252.64)	2.68 (249.08)	1.76 (217.74)	1.64 (190.62)
G_{11} (*Tridax procumbens* tincture 2ml)	6.64 (414.40)	4.68 (328.82)	4.28 (315.38)	3.92 (294.50)	2.64 (280.28)	2.6 (271.00)	2.04 (256.28)
G_{12} (*Tridax procumbens* decoction 1ml)	6.84 (348.30)	4.4 (265.60)	3.92 (221.36)	2.92 (212.54)	2.04 (199.90)	1.8 (180.36)	1.52 (171.06)
G_{13} (*Terminalia chebula* aqueous 1ml)	6.76 (333.20)	4.36 (266.40)	3.72 (222.12)	2.8 (193.08)	2.04 (180.36)	1.72 (178.44)	1.56 (155.18)
G_{14} (*Terminalia chebula* tincture 1ml)	6.12 (423.08)	5.76 (412.20)	4.68 (376.98)	3.44 (354.72)	2.88 (329.60)	2.28 (320.00)	2.28 (247.98)
G_{15} (*Terminalia chebula* decoction 1ml)	6.72 (321.40)	4.88 (269.44)	3.68 (269.06)	3.48 (256.34)	2.16 (219.92)	1.72 (204.08)	1.56 (176.00)

Table 51. Effect of herbal extracts and essential oils on colour of liquidized aloe gel juice (continued)

G_{16} (*Punica granatum* aqueous 1ml)	5.76 (254.04)	4.68 (240.44)	3.92 (232.10)	3.2 (216.62)	2.04 (216.36)	1.72 (180.36)	1.6 (125.60)
G_{17} (*Punica granatum* tincture 2ml)	5.72 (345.64)	5.04 (329.28)	3.52 (298.88)	3.28 (225.66)	2.16 (201.68)	2.24 (153.10)	2.08 (144.62)
G_{18} (*Punica granatum* decoction 1ml)	6.92 (298.90)	4.6 (249.98)	3.68 (236.16)	3.28 (229.50)	2.2 (213.04)	1.64 (207.48)	1.52 (176.00)
G_{19} (green tea aqueous 2ml)	6.16 (247.16)	3.36 (230.90)	2.16 (205.00)	1.96 (169.64)	1.68 (102.64)	1.68 (42.92)	1.56 (34.48)
G_{20} (green tea tincture 1ml)	6.88 (359.64)	5.28 (356.70)	4.8 (349.50)	4.08 (351.90)	3.12 (341.48)	2.12 (341.24)	1.52 (210.84)
G_{21} (green tea decoction 2ml)	6.76 (328.10)	4.44 (296.86)	4.04 (254.04)	3.76 (249.98)	2 (231.54)	1.72 (203.18)	1.64 (177.40)
G_{22} (basil oil 1ml)	6.36 (606.88)	6.16 (604.24)	6.08 (593.62)	6.08 (592.56)	5.4 (543.10)	5.16 (525.38)	4.72 (452.60)
G_{23} (lemon grass oil 1ml)	7 (604.24)	6.2 (596.30)	6.12 (586.50)	5.92 (573.72)	5.28 (541.72)	4.96 (498.82)	4.64 (387.30)
G_{24} (cinnamon bark oil 1ml)	7.52 (614.44)	6.32 (601.12)	6.28 (591.70)	6.16 (575.72)	5.04 (570.52)	5.4 (491.12)	4.4 (485.88)
G_{25} (clove oil 1ml)	8.2 (648.52)	6.96 (637.76)	6.92 (632.86)	6.68 (631.48)	6.52 (621.16)	5.88 (594.04)	5.44 (592.90)
G_{26} (cardamom oil 1ml)	7.8 (616.86)	7.04 (609.36)	6.04 (590.72)	5.96 (583.14)	5.32 (570.24)	5.24(539. 74)	5 (520.84)
G_{27} (control - sodium benzoate 1ml)	6.56 (444.26)	5.87 (429.94)	5.28 (373.90)	3.8 (306.02)	2.24 (292.84)	1.92 (237.78)	1.6 (216.76)
KW value	474.39	483.42	383.14	382.04	461.18	256.09	372.71
χ^2	38.89						
CD value	21.67						

*Figures in parenthesis indicates mean rank value

Table 52. Effect of herbal extracts and essential oils on odour of liquidized aloe gel juice

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	5.88 (293.96)	3.64 (285.64)	3 (281.68)	1.76 (274.24)	1.52 (274.28)	1.52 (270.54)	1.44 (216.52)
G_2 (*Gymnema sylvestre* tincture1ml)	6.29 (347.02)	3 (342.90)	2.56 (328.44)	1.84 (320.30)	1.72 (312.02)	1.6 (281.44)	1.48 (259. 90)
G_3 (*Gymnema sylvestre* decoction 2ml)	5.9 (289.90)	2.44 (281.68)	1.92 (240.16)	1.88 (221.48)	1.52 (207.62)	1.52 (204.10)	1.32 (200.48)
G_4 (*Centella asiatica* aqueous 1ml)	6.29 (362.50)	3.12 (340.34)	1.88 (319.66)	1.68 (285.08)	1.68 (274.28)	1.52 (253.6)	1.48 (207.62)
G_5 (*Centella asiatica* tincture 2ml)	6.17 (386.38)	3.44 (373.90)	2.8 (365.20)	2.44 (349.76)	1.6 (305.56)	1.6 (285.3)	1.52 (281.68)
G_6 (*Centella asiatica* decoction 1ml)	6.17 (330.96)	2.96 (324.84)	2.56 (316.84)	1.96 (316.48)	1.56 (279.82)	1.44 (263.40)	1.4 (259.96)
G_7 (*Achyranthes aspera* aqueous 2 ml)	6.12 (378.44)	1.88 (312.02)	1.76 (304.34)	1.72 (281.68)	1.52 (274.24)	1.44 (177.62)	1.44 (159.00)
G_8 (*Achyranthes aspera* tincture 2ml)	6.67 (375.16)	2.88 (307.34)	2.68 (299.58)	1.88 (297.94)	1.6 (285.08)	1.56 (279.82)	1.48 (277.80)
G_9 (*Achyranthes aspera* decoction 1.50 ml)	6.08 (321.34)	2.08 (281.68)	1.76 (274.80)	1.56 (263.40)	1.52 (219.84)	1.52 (186.22)	1.4 (183.16)
G_{10} (*Tridax procumbens* aqueous 2ml)	5.96 (298.34)	2.48 (285.08)	1.88 (265.78)	1.72 (259.96)	1.48 (248.00)	1.44 (242.60)	1.40 (207.62)
G_{11} (*Tridax procumbens* tincture 2ml)	6.62 (411.94)	2.72 (405.28)	1.92 (394.90)	1.56 (300.74)	1.52 (276.22)	1.48 (258.70)	1.48 (210.16)
G_{12} (*Tridax procumbens* decoction 1ml)	5.96 (295.92)	1.96 (279.70)	1.88 (279.06)	1.64 (249.10)	1.52 (219.84)	1.48 (207.52)	1.40 (170.30)
G_{13} (*Terminalia chebula* aqueous 1ml)	5.79 (281.68)	2.24 (267.66)	1.8 (253.16)	1.6 (241.72)	1.52 (239.20)	1.44 (210.88)	1.32 (192.06)
G_{14} (*Terminalia chebula* tincture 1ml)	5.88 (363.20)	2.48 (318.78)	2.8 (279.10)	2 (276.22)	1.52 (269.26)	1.52 (250.56)	1.36 (248.26)
G_{15} (*Terminalia chebula* decoction 1ml)	5.96 (276.48)	2.12 (274.24)	1.92 (253.64)	1.68 (253.16)	1.44 (252.06)	1.44 (249.10)	1.4 (165.68)

Table 52. Effect of herbal extracts and essential oils on odour of liquidized aloe gel juice (continued)

G_{16} (*Punica granatum* aqueous 1ml)	5.5 (279.82)	1.84 (265.78)	1.80 (252.56)	1.72 (249.10)	1.56 (229.20)	1.40 (198.72)	1.36 (145.64)
G_{17} (*Punica granatum* tincture 2ml)	6.13 (347.98)	2.72 (323.70)	2.76 (313.70)	2 (292.54)	1.56 (276.04)	1.48 (258.70)	1.36 (252.56)
G_{18} (*Punica granatum* decoction 1ml)	6 (289.92)	2.32 (279.82)	2.8 (263.72)	1.68 (263.10)	1.48 (253.64)	1.4 (249.10)	1.4 (227.72)
G_{19} (green tea aqueous 2ml)	5.92 (295.92)	2.76 (290.38)	1.88 (285.62)	1.76 (273.00)	1.6 (271.30)	1.52 (270.82)	1.48 (207.62)
G_{20} (green tea tincture 1ml)	6.08 (387.34)	3.56 (306.60)	2.2 (287.08)	1.8 (277.76)	1.56 (274.28)	1.52 (267.62)	1.36 (252.56)
G_{21} (green tea decoction 2ml)	6.04 (306.22)	2.92 (299.54)	2.08 (290.38)	1.6 (263.40)	1.52 (259.96)	1.44 (243.16)	1.4 (219.84)
G_{22} (basil oil 1ml)	7.16 (621.94)	6.44 (612.20)	6.04 (611.80)	6 (598.36)	5.96 (594.52)	5.68 (591.34)	2.62 (430.46)
G_{23} (lemon grass oil 1ml)	6.8 (619.58)	6.68 (609.52)	6.58 (608.16)	6.24 (601.34)	6.12b (585.86)	5.92 (583.36)	4.88 (404.16)
G_{24} (cinnamon bark oil 1ml)	6.84 (609.78)	6.54 (603.90)	6.52 (603.50)	6.52 (603.08)	5.8 (602.88)	5.76 (600.06)	5.32 (404.16)
G_{25} (clove oil 1ml)	8.65 (650.62)	7.58 (636.68)	7.28 (636.34)	6.88 (630.64)	6.6 (622.42)	6.36 (620.30)	6.32 (593.70)
G_{26} (cardamom oil 1ml)	7.72 (621.42)	7.08 (620.80)	6.88 (620.74)	6.88 (620.30)	6.28 (617.68)	5.92 (605.90)	5.88 (528.06)
G_{27} (control - sodium benzoate 1ml)	6.56 (512.92)	4.56 (502.32)	4.16 (401.16)	2.32 (346.56)	1.84 (263.40)	1.64 (317.04)	1.4 (314.26)
KW value	136.14	427.26	363.35	358.10	439.32	355.70	385.06
χ^2	38.89						
CD value	21.67						

*Figures in parenthesis indicates mean rank value

4.3.2.2 Refractive Index

Effect of herbal extracts and essential oils on refractive index of liquidized aloe gel juice are presented in table 53. There was no significant variation on refractive index of liquidized aloe gel juice added with different concentration of aqueous, tincture and decoction of herbal extracts, essential oils (1 ml) of basil oil, lemon grass oil, cinnamon bark oil, clove oil and cardamom oil as well as sodium benzoate 1 ml from first day of storage upto six month after storage.

Present study shows that the refractive index ranged from 1.333 to 1.338 from first day to six month of storage. Refractive index of *Aloe vera* gel increased from 1.3341 to 1.3348, with a significant increase up to three years of plant maturity (Ahlawat *et al*., 2013). Refractive index of aloe gel was reported to range from 1.3340 – 1.3355, by Aloe CORP and 1.33789 to 1.34390 by M/S Delta International (Chandegara and Varshney, 2013). Aloe gel with the lowest refractive index was considered as the best treatment for extraction process (Chandegara and Varshney, 2013). High refractive index indicated more impurities in the gel (Chandegara *et al*., 2015). Chandegara *et al*. (2015) during the study on aloe gel found the minimum and maximum refractive index as 1.33603 and 1.33610 in aloe gel with 1.31 ^{0}Brix and also reported that refractive index of aloe gel is near to that of distilled water, more impurities leads to high refractive index. The value of refractive index obtained in the present study is near to the values in the studies quoted above. However the refractive indices of different samples were non significant.

4.3.2.3 Specific gravity

Effect of herbal extracts and essential oils on specific gravity of liquidized aloe gel juice is presented in table 54. The treatments were found to be non significant from zero month after the month of storage upto third month after storage.

Fourth month after storage the treatments showed significant variation in the case of specific gravity. The specific gravity was highest for *Tridax procumbens* aqueous 2 ml, G_{10} (1.013) which was followed by *Terminalia chebula* aqueous 1ml,

G_{13} (1.011). The least value of 1.003 was shown by G_5, G_8 and G_{22} which were on par with G_{11}, G_{24} and G_{25} (1.004).

After fifth month of storage the treatments showed significant difference among them. The highest value was noticed in *Tridax procumbens* aqueous 2 ml, G_{10} (1.014) which was followed by G_9, G_{15} and G_{19} as 1.012. The specific gravity noticed was the least after fifth month of storage in G_{22}, G_{24} and G_{25} as 1.004 which was on par with G_5, G_8 and G_{11} (1.005).

The specific gravity of liquidized aloe gel juice added with different herbal extracts, essential oil and sodium benzoate and stored for six months showed significant variation among the treatments. The highest value among the treatments was shown by *Tridax procumbens* aqueous 2 ml, G_{10} (1.015) which was on par with G_3, G_9, G_{12} and G_{13} with a value of 1.014. The least specific gravity was noticed in G_{25} (1.004) which was on par with G_{24} (1.005).

Chandegara and Varshney (2013) reported that the specific gravity of aloe gel ranged from 1.0030 – 1.0070. According to Aloecorp specific gravity was 1.0221 while by M/S Delta International the specific gravity reported was 1.0339. According to Ahlawat *et al*., (2013) specific gravity of *Aloe vera* gel ranged from 1.0042 to 1.0064 with a significant increase of up to three years of plant maturity which may be attributed to an increase in the dissolved solid content of gel. In the present study specific gravity ranged from 1.000 to 1.014 after six months of storage. The study supports the present work that from first month of storage onwards the specific gravity showed an increasing trend.

Table 53. Effect of herbal extracts and essential oils on refractive index of liquidized aloe gel juice

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	1.333	1.334	1.334	1.335	1.336	1.337	1.338
G_2 (*Gymnema sylvestre* tincture1ml)	1.333	1.334	1.334	1.334	1.335	1.336	1.336
G_3 (*Gymnema sylvestre* decoction 2ml)	1.333	1.335	1.335	1.336	1.336	1.337	1.338
G_4 (*Centella asiatica* aqueous 1ml)	1.333	1.333	1.334	1.335	1.336	1.337	1.338
G_5 (*Centella asiatica* tincture 2ml)	1.333	1.333	1.334	1.334	1.334	1.335	1.336
G_6 (*Centella asiatica* decoction 1ml)	1.333	1.333	1.334	1.335	1.336	1.337	1.338
G_7 (*Achyranthes aspera* aqueous 2 ml)	1.333	1.333	1.334	1.335	1.336	1.337	1.338
G_8 (*Achyranthes aspera* tincture 2ml)	1.333	1.333	1.334	1.334	1.334	1.335	1.335
G_9 (*Achyranthes aspera* decoction 1.50 ml)	1.333	1.334	1.334	1.335	1.337	1.337	1.338
G_{10} (*Tridax procumbens* aqueous 2ml)	1.333	1.334	1.336	1.337	1.338	1.338	1.338
G_{11} (*Tridax procumbens* tincture 2ml)	1.333	1.333	1.334	1.334	1.335	1.335	1.336
G_{12} (*Tridax procumbens* decoction 1ml)	1.333	1.334	1.335	1.336	1.337	1.337	1.338
G_{13} (*Terminalia chebula* aqueous 1ml)	1.333	1.335	1.335	1.336	1.337	1.337	1.338
G_{14} (*Terminalia chebula* tincture 1ml)	1.333	1.334	1.334	1.335	1.335	1.336	1.336
G_{15} (*Terminalia chebula* decoction 1ml)	1.333	1.334	1.335	1.336	1.336	1.338	1.338

Table 53. Effect of herbal extracts and essential oils on refractive index of liquidized aloe gel juice (continued)

G_{16} (*Punica granatum* aqueous 1ml)	1.333	1.334	1.334	1.335	1.335	1.337	1.337
G_{17} (*Punica granatum* tincture 2ml)	1.333	1.334	1.334	1.335	1.335	1.336	1.337
G_{18} (*Punica granatum* decoction 1ml)	1.333	1.334	1.335	1.336	1.337	1.337	1.338
G_{19} (green tea aqueous 2ml)	1.333	1.334	1.335	1.336	1.337	1.337	1.338
G_{20} (green tea tincture 1ml)	1.333	1.334	1.335	1.335	1.335	1.336	1.336
G_{21} (green tea decoction 2ml)	1.333	1.334	1.335	1.336	1.336	1.337	1.337
G_{22} (basil oil 1ml)	1.333	1.333	1.334	1.334	1.334	1.335	1.335
G_{23} (lemon grass oil 1ml)	1.334	1.334	1.334	1.334	1.336	1.337	1.337
G_{24} (cinnamon bark oil 1ml)	1.333	1.333	1.333	1.334	1.334	1.335	1.335
G_{25} (clove oil 1ml)	1.333	1.334	1.334	1.334	1.334	1.335	1.335
G_{26} (cardamom oil 1ml)	1.333	1.333	1.334	1.334	1.335	1.336	1.336
G_{27} (control - sodium benzoate 1ml)	1.333	1.334	1.335	1.335	1.337	1.337	1.338
SEm (±)	0.000	0.000	0.000	0.000	0.000	0.000	0.000
CD (0.05)	N S	N S	N S	N S	N S	N S	N S

*Each value is the mean of five replication

Table 54. Effect of herbal extracts and essential oils on specific gravity of liquidized aloe gel juice

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	1.000	1.001	1.003	1.006	1.007	1.009	1.013
G_2 (*Gymnema sylvestre* tincture1ml)	1.000	1.001	1.002	1.003	1.006	1.007	1.008
G_3 (*Gymnema sylvestre* decoction 2ml)	1.000	1.005	1.006	1.008	1.009	1.011	1.014
G_4 (*Centella asiatica* aqueous 1ml)	1.000	1.001	1.002	1.005	1.007	1.010	1.013
G_5 (*Centella asiatica* tincture 2ml)	1.001	1.000	1.001	1.002	1.003	1.005	1.008
G_6 (*Centella asiatica* decoction 1ml)	1.000	1.000	1.003	1.005	1.007	1.010	1.013
G_7 (*Achyranthes aspera* aqueous 2 ml)	1.000	1.001	1.003	1.005	1.008	1.011	1.013
G_8 (*Achyranthes aspera* tincture 2ml)	1.001	1.000	1.001	1.002	1.003	1.005	1.006
G_9 (*Achyranthes aspera* decoction 1.50 ml)	1.000	1.002	1.003	1.007	1.010	1.012	1.014
G_{10} (*Tridax procumbens* aqueous 2ml)	1.000	1.003	1.007	1.011	1.013	1.014	1.015
G_{11} (*Tridax procumbens* tincture 2ml)	1.001	1.003	1.002	1.002	1.004	1.005	1.008
G_{12} (*Tridax procumbens* decoction 1ml)	1.000	1.004	1.005	1.008	1.010	1.012	1.014
G_{13} (*Terminalia chebula* aqueous 1ml)	1.000	1.004	1.006	1.009	1.011	1.012	1.014
G_{14} (*Terminalia chebula* tincture 1ml)	1.000	1.002	1.003	1.005	1.006	1.007	1.008
G_{15} (*Terminalia chebula* decoction 1ml)	1.000	1.004	1.005	1.007	1.009	1.012	1.013

Table 54. Effect of herbal extracts and essential oils on specific gravity of liquidized aloe gel juice (continued)

G_{16} (*Punica granatum* aqueous 1ml)	1.000	1.002	1.003	1.005	1.006	1.010	1.012
G_{17} (*Punica granatum* tincture 2ml)	1.001	1.014	1.003	1.004	1.007	1.009	1.010
G_{18} (*Punica granatum* decoction 1ml)	1.000	1.004	1.005	1.007	1.009	1.011	1.013
G_{19} (green tea aqueous 2ml)	1.000	1.005	1.005	1.008	1.009	1.012	1.013
G_{20} (green tea tincture 1ml)	1.000	1.000	1.004	1.005	1.006	1.007	1.008
G_{21} (green tea decoction 2ml)	1.000	1.003	1.005	1.007	1.009	1.011	1.011
G_{22} (basil oil 1ml)	1.000	1.001	1.001	1.002	1.003	1.004	1.006
G_{23} (lemon grass oil 1ml)	1.000	1.000	1.002	1.004	1.008	1.010	1.011
G_{24} (cinnamon bark oil 1ml)	1.000	1.000	1.001	1.003	1.004	1.004	1.005
G_{25} (clove oil 1ml)	1.001	1.001	1.002	1.003	1.004	1.004	1.004
G_{26} (cardamom oil 1ml)	1.000	1.000	1.002	1.004	1.006	1.007	1.008
G_{27} (control - sodium benzoate 1ml)	1.000	1.000	1.004	1.006	1.009	1.011	1.013
SEm (±)	0.000	0.000	0.000	0.000	0.000	0.000	0.000
CD (0.05)	N S	N S	N S	N S	0.001	0.001	0.001

*Each value is the mean of five replication

4.3.2.4 pH value

Effect of herbal extracts and essential oils on pH of liquidized aloe gel juice are presented in table 55. The pH was found to be non-significant among the treatment after zero month of storage.

After first month of storage the treatments were shown significant increase in pH. The highest value among the treatments was noticed in *Gymnema sylvestre* aqueous 1 ml, G_1 (6.75) which were followed by *Gymnema sylvestre* decoction 2 ml, G_3 (6.62). The least value for pH among the treatments was noticed in G_{26} (3.49) which was on par with G_{25} (3.50), G_{23} (3.51) and G_{22} (3.51).

Significant variation was noticed among the treatments during second month of storage also. The highest value was noticed in *Gymnema sylvestre* aqueous 1 ml, G_1 (7.16) which were followed by *Gymnema sylvestre* decoction 2 ml, G_3 (6.36). The least value was noticed in G_{18} (3.43) which were on par with G_5 (3.46) and G_{25} (3.55).

The pH of the treatments was significantly different showed a decreasing trend from third month of storage. The highest value was noticed in *Gymnema sylvestre* decoction 2 ml, G_3 (6.27) which were followed by *Gymnema sylvestre* aqueous 1 ml, G_1 (6.10). Among the treatments the least value was shown by G_4 (2.27) which were followed by G_{16} (2.46). The pH of the liquidized aloe gel juice added with essential oil showed a pH range from 3.31 in cardamom oil (G_{26}) to 3.54 in basil oil (G_{22}).

During the fourth month after storage significant variation in pH was noticed among the treatments. pH was noticed to be highest for *Gymnema sylvestre* decoction 2 ml, G_3 (6.04) which was followed by *Gymnema sylvestre* aqueous 1 ml G_1 (6.01). The least value among the treatments was noticed in G_{16} (2.34) which was followed by G_{18} (2.50). The pH of the liquidized aloe gel juice added with essential oil showed a pH range from 3.25 in cardamom oil (G_{26}) to 3.55 in clove oil (G_{25}).

Fifth month after storage significant variations among the treatments was noticed in the pH recorded. The highest value was shown by *Gymnema sylvestre* aqueous 1 ml, G_1 (5.95) which were followed by *Gymnema sylvestre* decoction 2 ml, G_3 (5.87). Least value was shown among the treatment in G_{16} (2.23) which was followed by G_{18} (2.43). The pH of the liquidized aloe gel juice added with essential oil at fifth month of storage showed a pH range from 2.89 in cardamom oil (G_{26}) to 3.49 in clove oil (G_{25}).

On sixth month after storage, the pH of the liquidized aloe gel juice added with herbal extracts, essential oil and sodium benzoate showed significant variation. Significantly high pH value among the treatments was shown by G_1 (5.30) which were followed by G_3 (5.03). G_{16} showed the least value among the treatments as 2.04 which was on par with G_4 (2.05). The pH of the liquidized aloe gel juice added with essential oil varied from 2.82 in cardamom oil (G_{26}) to 3.49 in clove oil (G_{25}).

The pH of the liquidized aloe gel juice added with 1ml of sodium benzoate showed a pH of 3.5 at zero month after storage followed by 3.72, 5.94, 5.38, 3.80, 2.95 and 2.92 pH on 1 MAS, 2 MAS, 3 MAS, 4 MAS, 5 MAS and 6 MAS respectively.

Bor and Jasper (1988) reported that the pH which supports the growth of most microbes ranged from 6.6 - 7.5. This is in tune with the present study that in the herbal extracts added aloe juice, pH was between this range and it showed microbial population from first month after storage. Under poor handling/storage conditions, in the presence of bacteria, malic acid can be broken down to form lactic acid, which might have resulted in the increase in pH of storage. The increase in pH during first and second month of storage in most of the samples might be because of this. Ruggeri *et al*. (2008) reported reduction in pH of fermented milk due to the persistant growth and metabolic activity of lactic acid bacteria. Yadav *et al*. (2010) reported that variations in pH during storage are due to the change in chemical properties which were affected by storage conditions. The study also reported that significant decrease in pH of the samples during storage might be due to increase in their titratable acidity. From second month after storage the pH of the samples showed a decreasing trend. Pawar *et al*. (2011) reported that the pH of the beverage was found to decrease with increase in storage and could be correlated inversely with the acidity of product. Decrease in pH with storage time, affected the organoleptic qualities of fruits blends (Awsi and Dorcus, 2012). Decrease in pH had resulted in decrease in sensory parameters.

4.3.2.5 Total solids

Effect of herbal extracts and essential oils on total solids of liquidized aloe gel juice are presented in table 56.

The total solids were found to be significantly different among the treatment during zero month after storage. The percentage of total solids was significantly higher in G_2 (*Gymnema sylvestre* tincture 1 ml) and G_5 (*Centella asiatica* tincture 2ml) which recorded a value of 0.81 per cent followed by G_{22} (basil oil 1 ml) and G_{25} (clove oil 1 ml) which recorded a value of 0.78 per cent. The total solids noticed was the least during zero month after storage in G_4 (*Centella asiatica* aqueous 1 ml), G_{10} (*Tridax procumbens* aqueous 2 ml), G_{13} (*Terminalia chebula* aqueous1 ml), and G_{19} (green tea aqueous 2ml) which recorded 0.61per cent.

As observed in Table 56, the treatments varied significantly with respect to total solids during first month after storage. Among the treatments, higher per centage of total solids (0.79 per cent) were noticed in G_2 (*Gymnema sylvestre* tincture 1 ml) which was followed by G_5 (*Centella asiatica* tincture 2 ml) which recorded a value of 0.77 per cent. In the case of total solids the least value were observed in G_{19} (green tea aqueous 2ml) with 0.51 per cent followed by G_{13} (*Terminalia chebula* aqueous 1 ml) and G_{21} (green tea decoction 2 ml) with 0.57 per cent.

Significant variation in total solids was observed among the treatments in second month after storage. Highest value of 0.72 per cent was noticed in G_2

(*Gymnema sylvestre* tincture 1ml), which was followed by G_{20} (green tea tincture 1 ml) and G_{25} (clove oil 1 ml) with a value of 0.68 per cent. Least value of total soilds aftersecond month of storage having a value of 0.41 per cent was obtained from G_{13} (*Terminalia chebula* aqueous 1 ml) followed by G_{12} (*Tridax procumbens* decoction 1 ml) and (0.43 per cent in G_{19} (green tea aqueous 2 ml).

The total solid was significantly different under different treatments during third month of storage. Highest value was noticed in G_{23} (lemon grass oil 1 ml) and G_{25} (clove oil 1 ml) having 0.63 per cent and was significantly different from other treatments and which was followed by G_2 (*Gymnema sylvestre* tincture 1 ml), G_{20} (green tea tincture 1 ml), G_{22} (basil oil 1 ml) and G_{26} (cardamom oil 1ml) was 0.61 per cent. Among the treatments the least value was shown by G_{19} (green tea aqueous 2ml) with 0.32 per cent followed by G_{13} (*Terminalia chebula* aqueous 1 ml) with 0.35 per cent.

After the fourth month of storage significant variation in total solids was noticed among the treatments. The percentage of total solids was significantly higher for G_{25} (clove oil 1ml) with 0.60 per cent which was followed by G_{23} (lemon grass oil 1 ml with 0.57 per cent. The least total solid among the treatments was showed by G_{13} (*Terminalia chebula* aqueous 1 ml) (0.22 per cent) which was on par with G_{19} (green tea aqueous 2 ml) with 0.23 per cent.

Total solid content of liquidized aloe gel juice added with herbal extracts and essential oils after five months after storage showed significant variations among the treatments. Treatment with clove oil 1 ml, G_{25} had 0.57 per cent total solid which was followed by G_{23} (lemon grass oil 1ml) with a value of 0.51 per cent. Total solid content of other treatments containing essential oils such as basil oil, G_{22} was 0.41 per cent while that with cardamom oil 1ml, G_{26} and cinnamon bark oil, G_{24} was 0.50 per cent. Least value was shown by the treatment G_{13} (*Terminalia chebula* aqueous 1 ml) and G_{19} (green tea aqueous 2 ml) with 0.17 per cent which was on par with 0.18 per cent in G_{12} (*Tridax procumbens* decoction 1 ml).

After sixth month of storage, the total solid of the liquidized aloe gel juice added with herbal extracts, essential oil and sodium benzoate showed significant variation. Significantly high total solid value among the treatments was shown by $G_{25,}$ clove oil 1 ml (0.55 per cent) followed by 0.47 per cent in G_{23} (lemon grass oil 1 ml). Among the treatments the least value of 0.10 per cent was noticed for G_7 (*Achyranthes aspera* aqueous 2 ml) and G_{10} (*Tridax procumbens* aqueous 2 ml) which was on par with G_{19} (green tea aqueous 2 ml) with 0.11 per cent.

Total solids during zero month after storage ranged from 0.61 to 0.81 per cent, this range was supported by the study of Chandegara and Varshney (2012) that total solids (without preservatives and additives) present in aloe leaves by hand filleting method was 0.45 – 0.65 per cent. The higher content of total solids in the present experiment may be due to the addition of herbal extracts and essential oils. The present result was also supported by Ahlawat *et al*. (2013) that total solids in *Aloe vera* increased from 0.68 to 1.1 per cent with increasing age of up to four years implying accumulation of more bio-molecules in the gel.

Total solid is the amount of dry material remaining after the water is evaporated (Richard and Pierrie, 2006). High total solid content implies that samples were able to retain fibres in the sample (Francis *et al*., 2017). The above two studies supported the decreasing trend shown by total solids during storage, the decrease in total solids implies loss of fibre content and quality of aloe juice on storage.

Table 55. Effect of herbal extracts and essential oils on pH of liquidized aloe gel juice

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	3.50	6.75	7.16	6.10	6.01	5.95	5.30
G_2 (*Gymnema sylvestre* tincture1ml)	3.50	4.27	4.50	4.01	3.99	3.85	2.94
G_3 (*Gymnema sylvestre* decoction 2ml)	3.50	6.62	6.36	6.27	6.04	5.87	5.03
G_4 (*Centella asiatica* aqueous 1ml)	3.50	2.42	2.31	2.27	2.51	2.49	2.05
G_5 (*Centella asiatica* tincture 2ml)	3.50	3.53	3.46	3.35	3.33	3.31	3.29
G_6 (*Centella asiatica* decoction 1ml)	3.50	3.53	3.57	3.38	2.99	2.98	2.91
G_7 (*Achyranthes aspera* aqueous 2 ml)	3.50	3.92	3.83	3.51	3.22	3.18	2.97
G_8 (*Achyranthes aspera* tincture 2ml)	3.50	3.52	3.95	3.90	3.47	3.31	3.26
G_9 (*Achyranthes aspera* decoction 1.50 ml)	3.50	4.53	5.93	4.84	4.75	3.93	3.28
G_{10} (*Tridax procumbens* aqueous 2ml)	3.50	4.85	4.98	3.47	3.30	2.82	2.60
G_{11} (*Tridax procumbens* tincture 2ml)	3.50	4.00	4.73	4.43	4.02	3.70	3.31
G_{12} (*Tridax procumbens* decoction 1ml)	3.50	5.75	6.51	5.86	4.96	4.57	3.47
G_{13} (*Terminalia chebula* aqueous 1ml)	3.50	4.61	4.66	4.51	3.86	2.94	2.49
G_{14} (*Terminalia chebula* tincture 1ml)	3.50	3.88	3.83	3.66	3.61	3.57	3.46
G_{15} (*Terminalia chebula* decoction 1ml)	3.50	3.67	4.20	3.94	3.45	3.21	3.05

Table 55. Effect of herbal extracts and essential oils on pH of liquidized aloe gel juice (continued)

G_{16} (*Punica granatum* aqueous 1ml)	3.50	3.71	3.83	2.46	2.34	2.23	2.04
G_{17} (*Punica granatum* tincture 2ml)	3.50	3.45	3.86	3.48	3.37	3.36	3.22
G_{18} (*Punica granatum* decoction 1ml)	3.50	2.47	3.43	2.52	2.50	2.43	2.39
G_{19} (green tea aqueous 2ml)	3.50	4.24	4.58	4.23	4.05	3.95	3.83
G_{20} (green tea tincture 1ml)	3.50	3.58	3.59	3.56	3.48	3.45	3.43
G_{21} (green tea decoction 2ml)	3.50	3.56	4.99	4.18	3.94	3.83	3.37
G_{22} (basil oil 1ml)	3.50	3.51	3.58	3.54	3.48	3.43	3.41
G_{23} (lemon grass oil 1ml)	3.50	3.51	3.57	3.51	3.37	3.28	3.17
G_{24} (cinnamon bark oil 1ml)	3.50	3.68	3.82	3.35	3.33	3.29	3.28
G_{25} (clove oil 1ml)	3.50	3.50	3.55	3.53	3.52	3.49	3.49
G_{26} (cardamom oil 1ml)	3.50	3.49	3.58	3.31	3.25	2.89	2.82
G_{27} (control - sodium benzoate 1ml)	3.50	3.72	5.94	5.38	3.80	2.95	2.92
SEm (±)	0.000	0.010	0.047	0.009	0.005	0.021	0.039
CD (0.05)	NS	0.028	0.129	0.025	0.013	0.059	0.109

*Each value is the mean of five replications

Table 56. Effect of herbal extracts and essential oils on total solids of liquidized aloe gel juice, percentage

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.62	0.60	0.57	0.49	0.32	0.21	0.17
G_2 (*Gymnema sylvestre* tincture1ml)	0.81	0.79	0.72	0.61	0.50	0.38	0.26
G_3 (*Gymnema sylvestre* decoction 2ml)	0.70	0.67	0.61	0.52	0.41	0.31	0.21
G_4 (*Centella asiatica* aqueous 1ml)	0.61	0.58	0.50	0.41	0.30	0.21	0.15
G_5 (*Centella asiatica* tincture 2ml)	0.81	0.77	0.64	0.59	0.42	0.33	0.21
G_6 (*Centella asiatica* decoction 1ml)	0.71	0.67	0.60	0.51	0.40	0.32	0.21
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.66	0.61	0.56	0.40	0.31	0.20	0.10
G_8 (*Achyranthes aspera* tincture 2ml)	0.71	0.70	0.64	0.60	0.51	0.41	0.34
G_9 (*Achyranthes aspera* decoction 1.50 ml)	0.67	0.60	0.56	0.41	0.32	0.21	0.12
G_{10} (*Tridax procumbens* aqueous 2ml)	0.61	0.59	0.51	0.41	0.31	0.22	0.10
G_{11} (*Tridax procumbens* tincture 2ml)	0.73	0.70	0.61	0.51	0.42	0.32	0.21
G_{12} (*Tridax procumbens* decoction 1ml)	0.63	0.58	0.43	0.37	0.24	0.18	0.12
G_{13} (*Terminalia chebula* aqueous 1ml)	0.61	0.57	0.41	0.35	0.22	0.17	0.12
G_{14} (*Terminalia chebula* tincture 1ml)	0.76	0.71	0.65	0.57	0.41	0.28	0.20
G_{15} (*Terminalia chebula* decoction 1ml)	0.62	0.59	0.54	0.46	0.38	0.24	0.16

Table 56. Effect of herbal extracts and essential oils on total solids of liquidized aloe gel juice, percentage (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.62	0.60	0.51	0.41	0.32	0.21	0.14
G_{17} (*Punica granatum* tincture 2ml)	0.74	0.71	0.62	0.51	0.41	0.31	0.21
G_{18} (*Punica granatum* decoction 1ml)	0.64	0.61	0.53	0.42	0.32	0.23	0.15
G_{19} (green tea aqueous 2ml)	0.61	0.51	0.43	0.32	0.23	0.17	0.11
G_{20} (green tea tincture 1ml)	0.76	0.72	0.68	0.61	0.51	0.41	0.29
G_{21} (green tea decoction 2ml)	0.62	0.57	0.50	0.40	0.32	0.21	0.12
G_{22} (basil oil 1ml)	0.78	0.72	0.66	0.61	0.52	0.41	0.32
G_{23} (lemon grass oil 1ml)	0.75	0.71	0.64	0.63	0.57	0.51	0.47
G_{24} (cinnamon bark oil 1ml)	0.77	0.73	0.64	0.60	0.53	0.50	0.43
G_{25} (clove oil 1ml)	0.78	0.74	0.68	0.63	0.60	0.57	0.55
G_{26} (cardamom oil 1ml)	0.74	0.71	0.67	0.61	0.56	0.50	0.44
G_{27} (control - sodium benzoate 1ml)	0.62	0.60	0.53	0.48	0.41	0.30	0.22
SEm (±)	0.004	0.004	0.004	0.003	0.003	0.003	0.003
CD (0.05)	0.012	0.012	0.010	0.009	0.009	0.009	0.008

*Each value is the mean of five replication

4.3.2.6 Mono and Polysaccharides

4.3.2.6.1 Monosaccharides

Effect of herbal extracts and essential oils on monosaccharide content of liquidized aloe gel juice are presented in table 57.

The monosaccharide was found to be significant among the treatment during zero month after storage. The percentage of monosaccharides was significantly higher in G_{19} (green tea aqueous 2 ml), G_{23} (lemon grass oil 1ml), G_{24} (cinnamon bark oil 1 ml) and G_{27} (control – sodium benzoate) which recorded a value of 0.05 per cent followed by G_{22} (basil oil 1 ml), G_{25} (clove oil 1ml) and G_{26} (cardamom oil 1 ml) which recorded a value of 0.04 per cent. The monosaccharide noticed was the least during zero month after storage in G_1 (*Gymnema sylvestre* aqueous 1 ml), G_3 (*Gymnema sylvestre* decoction 2 ml), G_4 (*Centella asiatica* aqueous 1 ml), G_6 (*Centella asiatica* decoction 1 ml), G_7 (*Achyranthes aspera* aqueous 2 ml), G_9 (*Achyranthes aspera* decoction 1.5 ml), G_{10} (*Tridax procumbens* aqueous 2 ml), G_{11}(*Tridax procumbens* tincture 2 ml), G_{12} (*Tridax procumbens* decoction 1 ml), G_{15} (*Terminalia chebula* decoction 1 ml), G_{16} (*Punica granatum* aqueous 1 ml), G_{17} (*Punica granatum* tincture 2 ml) and G_{18} (*Punica granatum* decoction 1 ml) as 0.01 per cent.

After first month of storage the treatments varied significantly with respect to monosaccharides. Among the treatments, higher percentage of monosaccharides were noticed in in G_{19} (green tea aqueous 2ml), G_{23} (lemon grass oil 1 ml), G_{24} (cinnamon bark oil 1 ml) and G_{27} (control – sodium benzoate) which significantly differed from G_{22}(basil oil 1ml), G_{25} (clove oil 1 ml) and G_{26} (cardamom oil 1 ml) and recorded 0.05 and 0.04 per cent respectively. G_{20} (green tea tincture 1 ml) and G_{21} (green tea decoction 2 ml) showed 0.03 per cent monosaccharides.

Significant variation in monosaccharides was observed among the treatments aftersecond month of storage. Highest value of 0.05 per cent was noticed in G_{19}

(green tea aqueous 2 ml), G_{23} (lemon grass oil 1 ml), G_{24} (cinnamon bark oil 1 ml) G_{26} (cardamom oil 1 ml) and G_{27} (control – sodium benzoate) which was followed by G_{20} (green tea tincture 1 ml), G_{21} (green tea decoction 2 ml), G_{22} (basil oil 1 ml), G_{25} (clove oil 1ml) with a value of 0.04 per cent. Least value of monosaccharide during second month after storage having a value of 0.01 was obtained from G_1 (*Gymnema sylvestre* aqueous 1 ml), G_3 (*Gymnema sylvestre* decoction 2ml), G_{10} (*Tridax procumbens* aqueous 2 ml), G_{11} (*Tridax procumbens* tincture 2 ml) and G_{12} (*Tridax procumbens* decoction 1 ml).

The monosaccharide content of the treatments was significantly different and was showing an increasing trend from third month of storage. Highest value was noticed in G_{19} (green tea aqueous 2 ml) and G_{27} (control – sodium benzoate) having 0.06 per cent and was significantly different from other treatments. Monosaccharides of G_{22} (basil oil 1 ml) G_{23} (lemon grass oil 1 ml), G_{24} (cinnamon bark oil 1 ml) and G_{26} (cardamom oil 1 ml) was 0.05 per cent. Among the treatments the least value was shown by *Tridax procumbens* aqueous 2 ml, G_{10} and *Tridax procumbens* tincture 2 ml, G_{11} with 0.01 per cent.

During the fourth month after storage significant variation in monosaccharide was noticed among the treatments. The percentage of monosaccharide were significantly higher for G_{19} (green tea aqueous 2 ml), G_{23} (lemon grass oil 1 ml) and G_{27} (control – sodium benzoate) with 0.06 per cent which was followed by G_{22} (basil oil 1 ml), G_{24} (cinnamon bark oil 1 ml) G_{26} (cardamom oil 1 ml) with 0.05 per cent. The least monosaccharide among the treatments was shown by treatments from G_1 to G_{18} with a value of 0.02 per cent.

Monosaccharide content of liquidized aloe gel juice added with herbal extracts and essential oils after five months after storage showed significant variations among the treatments. Treatment with green tea aqueous 2 ml, G_{19} had 0.07 per cent of monosaccharide which was followed by G_{22} (basil oil 1 ml), G_{23} (lemon grass oil 1 ml) and G_{27} (control – sodium benzoate) with a value of 0.06 per cent.

Monosaccharide content of other treatments containing essential oils such as clove 1 ml, G_{25} was 0.04 per cent while that for cardamom oil 1 ml, G_{26} and cinnamon bark oil G_{24} was 0.05 per cent. Least value was shown by the treatments from G_1 to G_{18} (0.02 per cent) except G_5.

After sixth month of storage, the monosaccharide of the liquidized aloe gel juice added with herbal extracts, essential oil and sodium benzoate showed significant variation. Significantly high monosaccharide value among the treatments was shown by green tea aqueous 2ml, G_{19} (0.08 per cent) which was significantly different from the control, G_{27} (0.07 per cent). Among the treatments the least value of 0.02 per cent was noticed in G_1 to G_{18} except G_4.

The monosaccharide content of the liquidized aloe gel juice added with 1ml of clove oil showed a value of 0.04 per cent from zero MAS to 6 MAS, without showing any change while for the treatment with 1ml cinnamon bark oil the monosaccharide content was the same from zero MAS to 5 MAS.

4.3.2.6.2 Polysaccharides

The effect of herbal extracts and essential oils on polysaccharide content of liquidized aloe gel juice is presented in table 58. The treatments were found to be significantly different for the polysaccharide content from zero month after storage up to six month after storage.

The polysaccharide content showed significant variation among the treatment during zero month after storage. Polysaccharide content was higher in G_{22} (basil oil 1 ml) with 0.38 per centwhich was significantly higher from G_{23} (lemon grass oil 1 ml) and G_{24} (cinnamon bark oil 1 ml) having a value of 0.27 per cent. The control (sodium benzoate) had polysaccharide content of 0.25 per cent. Among the treatments with essential oils the polysaccharide content ranged from 0.24 per cent (G_{25}, clove oil, 1 ml) to 0.38 per cent ($G_{22,}$ basil oil 1 ml).

On first month after storage the polysaccharide content did not change compared to zero month after storage in treatments G_{22}, basil oil 1 ml (0.37 per cent) which was the highest followed by G_{23}, lemon grass oil 1 ml (0.27 per cent). The treatment G_{27} (control – sodium benzoate) had polysaccharide content of 0.25 per cent. The percentage of polysaccharide among essential oils ranged from 0.25 per cent to 0.37 per cent. The percentage of polysaccharide among herbal extracts ranged from 0.02 per cent to 0.25 per cent.

The treatments were significantly different and showed a decreasing trend in polysaccharide content from second month after storage. Highest polysaccharide content of 0.36 per cent was noticed in G_{22} (basil oil 1 ml) which differed significantly from G_{23} (lemon grass oil 1 ml) having 0.27 per cent. The control, G_{27} (sodium benzoate) showed polysaccharide content of 0.25 per cent. The polysaccharide content among oils ranged from 0.23 per cent in G_{26} (cardamom oil 1ml) to 0.36 per cent in G_{22} (basil oil 1 ml).The polysaccharide content in treatments with herbal extract varied from 0.02 per cent to 0.23 per cent.

The polysaccharide content of the treatments showed significant difference during third month after storage. The polysaccharide content of G_{22} (basil oil 1 ml) was 0.34 per cent which was the significantly high followed by G_{23} (lemon grass oil 1ml) with 0.27 per cent. The polysaccharide content of G_{26} (cardamom oil 1 ml) was 0.23 per cent and that for clove oil 1 ml was 0.24 per cent and cinnamon bark oil 1 ml was 0.26 per cent. The polysaccharide content of G_{27} (control – sodium benzoate) was 0.23 per cent.

During the fourth month after storage significant variation in polysaccharide content was noticed among the treatments. The value of polysaccharide was significantly higher for G_{22} (basil oil 1ml) with 0.30 per cent followed by G_{23} (lemon grass oil 1 ml) and G_{24} (cinnamon bark oil 1 ml) having a value of 0.26 per cent. The control showed a polysaccharide content of 0.23 per cent while for treatments with essential oils polysaccharides ranged from 0.23 per cent in G_{26} (cardamom oil 1 ml) to 0.30 per cent in G_{22} (basil oil 1 ml) .

Polysaccharide content of treatments at fifth month after storage showed significant variation. The highest polysaccharide content noticed in G_{22} (basil oil 1ml) which was 0.30 per cent followed by G_{23} (lemon grass oil 1ml) and G_{24} (cinnamon bark oil 1ml) with a value of 0.26 per cent. The G_{27} (control – sodium benzoate) had a polysaccharide content of 0.20 per cent. The polysaccharide content of G_{26}, cardamom oil 1ml was 0.22 per cent and that for clove oil 1ml, G_{25} was 0.24 per cent and that for cinnamon bark oil 1ml, G_{24} was to 0.26 per cent.

On sixth month after storage, the polysaccharide content of the liquidized aloe gel juice added with herbal extracts, essential oil and sodium benzoate were low and was significantly different. Significantly high polysaccharide content of 0.25 per cent was observed from G_{22} (basil oil 1ml), G_{23} (lemongrass oil 1ml) and G_{24} (cinnamon bark oil 1ml) which was followed by G_{25} (clove oil 1ml) with a value of 0.24 per cent. The control, G_{27} showed a polysaccharide content of 0.20 per cent. The polysaccharide content of treatments with herbal extracts varied from 0.01 to 0.20 per cent while that of essential oils varied from 0.22 per cent to 0.25 per cent.

Carbohydrates of *Aloe vera* are derived from mucilage layer of the plant under the rind, surrounding the inner parenchyma or gel comprising of both mono and polysaccharides. The long chain polysaccharides, comprising glucose and mannose, known as the glucomannans [β (1, 4) - linked acetylated mannan] are the most important. Xylose, rhamnose, galactose and arabinose are also present in trace amounts along with lupeol, cholesterol, campesterol and β-sitosterol. Structural studies on *Aloe vera* gel polysaccharides have shown that the gel is composed of at least four different partially acetylated glucomannans, being linear polymers with no branching and having 1,4 glycosidic linkages with glucose and mannose in the ratio of 1:2:8 (Pandey and Singh, 2016).

Carbohydrates in foods do not appear to protect bacteria from the action of essential oils as much as protein does (Shelef *et al*., 1984). According to Lee and Nagy (1988) the fructose, glucose and sucrose got degraded in grape fruit juice during storage at 37 ^{0}C for 16 weeks. During storage the heat processed juices showed increase in sugar levels which might be due to the inactivation of enzymes, they must be responsible for decreasing acidity and conversion of polysaccharides into simple sugars (Ghorai and Khurdiya 1998). These studies supported the result of increasing trend shown by monosaccharides and decreasing trend in polysaccharides during storage. The polysaccharides found in aloe gel are not stable, especially under stress conditions such as heat, in the presence of acid and enzymatic activities (Hamman, 2008). Kilima *et al*. (2014) reported that increase in reducing sugars was probably due to the hydrolysis of polysaccharides like starch, cellulose, pectin, etc., and conversion into simple sugars (glucose, fructose) during storage. These also supported the present results of the study. Talib *et al*. (2016) reported total, reducing and non reducing sugars of aloe juice as 0.70, 0.22 and 0.48 respectively.

Table 57. Effect of herbal extracts and essential oils on monosaccharides of liquidized aloe gel juice, percentage

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.01	0.01	0.01	0.02	0.02	0.02	0.02
G_2 (*Gymnema sylvestre* tincture1ml)	0.02	0.02	0.02	0.02	0.02	0.02	0.02
G_3 (*Gymnema sylvestre* decoction 2ml)	0.01	0.01	0.01	0.02	0.02	0.02	0.02
G_4 (*Centella asiatica* aqueous 1ml)	0.01	0.02	0.02	0.02	0.02	0.02	0.03
G_5 (*Centella asiatica* tincture 2ml)	0.02	0.02	0.02	0.02	0.02	0.03	0.02
G_6 (*Centella asiatica* decoction 1ml)	0.01	0.01	0.02	0.02	0.02	0.02	0.02
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.01	0.01	0.02	0.02	0.02	0.02	0.02
G_8 (*Achyranthes aspera* tincture 2ml)	0.02	0.02	0.02	0.02	0.02	0.02	0.02
G_9 (*Achyranthes aspera* decoction 1.50 ml)	0.01	0.01	0.02	0.02	0.02	0.02	0.02
G_{10} (*Tridax procumbens* aqueous 2ml)	0.01	0.01	0.01	0.01	0.02	0.02	0.02
G_{11} (*Tridax procumbens* tincture 2ml)	0.01	0.01	0.01	0.01	0.02	0.02	0.02
G_{12} (*Tridax procumbens* decoction 1ml)	0.01	0.01	0.01	0.02	0.02	0.02	0.02
G_{13} (*Terminalia chebula* aqueous 1ml)	0.02	0.02	0.02	0.02	0.02	0.02	0.02
G_{14} (*Terminalia chebula* tincture 1ml)	0.02	0.02	0.02	0.02	0.02	0.02	0.02
G_{15} (*Terminalia chebula* decoction 1ml)	0.01	0.01	0.02	0.02	0.02	0.02	0.02

Table 57. Effect of herbal extracts and essential oils on monosaccharides of liquidized aloe gel juice, percentage (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.01	0.01	0.02	0.02	0.02	0.02	0.02
G_{17} (*Punica granatum* tincture 2ml)	0.01	0.02	0.02	0.02	0.02	0.02	0.02
G_{18} (*Punica granatum* decoction 1ml)	0.01	0.02	0.02	0.02	0.02	0.02	0.02
G_{19} (green tea aqueous 2ml)	0.05	0.05	0.05	0.06	0.06	0.07	0.08
G_{20} (green tea tincture 1ml)	0.03	0.03	0.04	0.04	0.04	0.04	0.05
G_{21} (green tea decoction 2ml)	0.03	0.03	0.04	0.04	0.04	0.05	0.05
G_{22} (basil oil 1ml)	0.04	0.04	0.04	0.05	0.05	0.06	0.06
G_{23} (lemon grass oil 1ml)	0.05	0.05	0.05	0.05	0.06	0.06	0.06
G_{24} (cinnamon bark oil 1ml)	0.05	0.05	0.05	0.05	0.05	0.05	0.06
G_{25} (clove oil 1ml)	0.04	0.04	0.04	0.04	0.04	0.04	0.04
G_{26} (cardamom oil 1ml)	0.04	0.04	0.05	0.05	0.05	0.05	0.06
G_{27} (control - sodium benzoate 1ml)	0.05	0.05	0.05	0.06	0.06	0.06	0.07
SEm (±)	0.000	0.000	0.000	0.000	0.000	0.000	0.000
CD (0.05)	0.001	0.001	0.001	0.001	0.001	0.001	0.001

*Each value is the mean of five replication

Table 58. Effect of herbal extracts and essential oils on polysaccharides of liquidized aloe gel juice, percentage

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.02	0.02	0.02	0.02	0.02	0.01	0.01
G_2 (*Gymnema sylvestre* tincture1ml)	0.02	0.02	0.02	0.02	0.02	0.02	0.01
G_3 (*Gymnema sylvestre* decoction 2ml)	0.02	0.02	0.02	0.02	0.02	0.01	0.01
G_4 (*Centella asiatica* aqueous 1ml)	0.03	0.02	0.02	0.02	0.02	0.01	0.01
G_5 (*Centella asiatica* tincture 2ml)	0.03	0.03	0.03	0.03	0.02	0.02	0.03
G_6 (*Centella asiatica* decoction 1ml)	0.03	0.02	0.02	0.02	0.02	0.02	0.01
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.02	0.02	0.02	0.01	0.01	0.01	0.01
G_8 (*Achyranthes aspera* tincture 2ml)	0.03	0.03	0.03	0.03	0.03	0.02	0.02
G_9 (*Achyranthes aspera* decoction 1.50 ml)	0.02	0.02	0.02	0.01	0.01	0.01	0.01
G_{10} (*Tridax procumbens* aqueous 2ml)	0.02	0.02	0.02	0.01	0.01	0.01	0.01
G_{11} (*Tridax procumbens* tincture 2ml)	0.02	0.02	0.02	0.01	0.02	0.02	0.01
G_{12} (*Tridax procumbens* decoction 1ml)	0.02	0.02	0.02	0.01	0.01	0.01	0.01
G_{13} (*Terminalia chebula* aqueous 1ml)	0.02	0.02	0.02	0.02	0.01	0.01	0.01
G_{14} (*Terminalia chebula* tincture 1ml)	0.03	0.03	0.03	0.02	0.02	0.02	0.02
G_{15} (*Terminalia chebula* decoction 1ml)	0.03	0.02	0.02	0.02	0.01	0.01	0.01

Table 58. Effect of herbal extracts and essential oils on polysaccharides of liquidized aloe gel juice, percentage (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.03	0.02	0.02	0.02	0.01	0.01	0.01
G_{17} (*Punica granatum* tincture 2ml)	0.03	0.03	0.03	0.03	0.02	0.02	0.02
G_{18} (*Punica granatum* decoction 1ml)	0.03	0.02	0.02	0.02	0.02	0.01	0.01
G_{19} (green tea aqueous 2ml)	0.25	0.25	0.23	0.19	0.16	0.12	0.11
G_{20} (green tea tincture 1ml)	0.26	0.24	0.23	0.22	0.20	0.20	0.20
G_{21} (green tea decoction 2ml)	0.26	0.23	0.20	0.16	0.13	0.11	0.08
G_{22} (basil oil 1ml)	0.38	0.37	0.36	0.34	0.30	0.30	0.25
G_{23} (lemon grass oil 1ml)	0.27	0.27	0.27	0.27	0.26	0.26	0.25
G_{24} (cinnamon bark oil 1ml)	0.27	0.26	0.26	0.26	0.26	0.26	0.25
G_{25} (clove oil 1ml)	0.24	0.25	0.24	0.24	0.24	0.24	0.24
G_{26} (cardamom oil 1ml)	0.25	0.25	0.23	0.23	0.23	0.22	0.22
G_{27} (control - sodium benzoate 1ml)	0.25	0.25	0.24	0.23	0.23	0.20	0.20
SEm (±)	0.000	0.000	0.000	0.000	0.000	0.000	0.000
CD (0.05)	0.001	0.001	0.001	0.001	0.001	0.001	0.001

*Each value is the mean of five replication

4.3.2.7 Amino acids

The effect of herbal extracts and essential oils on amino acids content of liquidized aloe gel juice is presented in table 59.

The amino acid content was found to be significantly different among the treatments during the month of storage. Significantly higher amino acid content (0.08 ppm) was noticed in G_7 (*Achyranthes aspera* aqueous, 2 ml), G_8 (*Achyranthes aspera* tincture, 2 ml) and G_9 (*Achyranthes aspera* decoction, 1.50 ml) followed by G_{16} (*Punica granatum* aqueous, 1 ml), G_{17} (*Punica granatum* tincture, 2 ml) and G_{18} (*Punica granatum* decoction, 1 ml) with an aminoacid content of 0.07 ppm. The amino acid content among treatments with essential oils was found to be 0.04 ppm in all essential oils except lemon grass oil 1 ml, G_{23} with 0.05 ppm. The control G_{27} showed an amino acid content of 0.04 ppm.

On first month after storage amino acid content of the treatments showed significant variation. Amino acid content was significantly higher for G_7 (*Achyranthes aspera* aqueous, 2 ml), G_8 (*Achyranthes aspera* tincture, 2 ml) and G_9 (*Achyranthes aspera* decoction, 1.50 ml) having a value of 0.08 ppm which differed significantly from G_{16} (*Punica granatum* aqueous, 1 ml), G_{17} (*Punica granatum* tincture, 2 ml) and G_{18} (*Punica granatum* decoction, 1 ml) with 0.07 ppm. The amino acid content of the control G_{27} was 0.04 ppm. The amino acid content of liquidized aloe gel juice with herbal extracts varied from 0.03 to 0.08 ppm while amino acid content of all essential oils was 0.04 ppm.

Amino acid content of the treatments varied significantly during second month of storage. Highest amino acid of 0.08 ppm was noticed in G_7 (*Achyranthes aspera* aqueous, 2 ml) and G_8 (*Achyranthes aspera* tincture, 2 ml) which was significantly higher than G_9 (*Achyranthes aspera* decoction, 1.50 ml), G_{16} (*Punica granatum* aqueous, 1 ml), G_{17} (*Punica granatum* tincture, 2 ml) and G_{18} (*Punica granatum* decoction, 1 ml) with an amino acid content of 0.07 ppm. The value obtained in control G_{27} was 0.03 ppm. The amino acid content of liquidized aloe gel juice with different essential oils was 0.04 ppm.

The amino acid content of the treatments was significantly different and was showing a decreasing trend from third month after storage compared to earlier periods of storage. Highest amino acid content (0.07 ppm) was noticed in G_{16} (*Punica granatum* aqueous, 1 ml) and G_{17} (*Punica granatum* tincture, 2 ml) which was significantly higher than G_7 (*Achyranthes aspera* aqueous, 2 ml), G_8 (*Achyranthes aspera* tincture, 2 ml) and G_{18} (*Punica granatum* decoction, 1 ml) with 0.06 ppm. The control G_{27} (control) obtained a value of 0.03 ppm. The value obtained in essential oils treated samples were 0.04 ppm.

Amino acid content showed significant difference between treatments after four month of storage. Highest amino acid content of 0.06 ppm was noticed in G_{16} (*Punica granatum* aqueous, 1 ml), G_{17} (*Punica granatum* tincture, 2 ml) and G_{18} (*Punica granatum* decoction, 1 ml) which differed significantly from G_8 (*Achyranthes aspera* tincture, 2 ml), G_{10} (*Tridax procumbens* aqueous 2 ml), G_{11} (*Tridax procumbens* tincture 2 ml), G_{12} (*Tridax procumbens* decoction, 1 ml), G_{23} (lemon grass oil, 1ml) and G_{25} (clove oil, 1 ml) with 0.04 ppm of amino acid content. The control G_{27} had an amino acid content of 0.03 ppm. The amino acid content of essential oils treated samples varied from 0.03 ppm to 0.04 ppm.

The amino acid content of the treatments was significantly different after five month of storage. Highest amino acid content of 0.05 ppm was noticed in G_{16} (*Punica granatum* aqueous, 1ml), G_{17} (*Punica granatum* tincture, 2ml) and G_{18} (*Punica granatum* decoction, 1ml) which was followed by G_{11} (*Tridax procumbens* tincture 2ml), and G_{25} (clove oil, 1ml) with 0.04 ppm amino acid content. Amino acid content of control G_{27} was 0.02 ppm. The value obtained in essential oils treated samples varied from 0.03 ppm to 0.04 ppm.

Amino acid content of liquidized aloe gel juice added with herbal extracts and essential oil at six month after storage showed significant difference among treatments.Amino acid content of 0.06 ppm was observed from G_{16} (*Punica granatum* aqueous, 1 ml), G_{17} (*Punica granatum* tincture, 2 ml) and G_{18} (*Punica granatum* decoction, 1 ml) followed by G_{25} (clove oil, 1 ml) with a value of 0.04 ppm. The amino acid content of control G_{27} was 0.01 ppm. The amino acid content of essential oils treated samples ranged from 0.03 ppm - 0.04 ppm.

The amino acid content of treatment with 1ml clove oil remained same as 0.04 ppm throughout the observation period from zero MAS to 6 MAS.

David and Sudarsanam (2013) reported that the antimicrobial activity of leaf extracts of *Gymnema sylvestre*, which can be attributed to the presence of phytochemicals like flavonoids, terpenoids, amino acids, glycosides, tannins, amino acids and carbohydrates.

The change in free amino acid composition of peach juice concentrate as a result of Maillard reactions, damage the product quality (Buedo *et al*., 2000) and a decrease in total amino acid content was noticed in peach juice concentrate after 112 days of storage at temperatures 15 ^{0}C, 30 ^{0}C and 37 ^{0}C respectively. These studies supported the present result of decreasing trend in amino acids during storage which might be because of the degradation of the samples in those added with herbal extracts. But in samples added with essential oils especially clove oil, the there was no change in amino acid content thus implying less degradation of the product.

4.3.2.8 Vitamins

4.3.2.8.1 Vitamin A

Effect of herbal extracts and essential oils on vitamin A content of liquidized aloe gel juice are presented in table 60.

The vitamin A content was found to be significant among the treatment during zero month after storage. Vitamin A content at zero MAS was 0.11 mg/100 ml for all liquidized aloe gel juice added with herbal extracts and essential oils except G_{19} (green tea aqueous, 2 ml), G_{20} (green tea tincture, 1 ml), G_{24} (cinnamon bark oil, 1 ml) and G_{27} (control – sodium benzoate) which recorded 0.12 mg/100ml.

On first month after storage the treatments showed significant variation in vitamin A content. The highest value of 0.11 mg/100 ml was noticed in all the treatments except G_9 (*Achyranthes aspera* decoction, 1.50 ml), G_{10} (*Tridax procumbens* aqueous, 2 ml), G_{16} (*Punica granatum* aqueous, 1 ml) and G_{22} (basil oil, 1 ml). The G_9 (*Achyranthes aspera* decoction, 1.50 ml), G_{16} (*Punica granatum* aqueous, 1 ml) and G_{22} (basil oil, 1 ml) obtained a vitamin A content of 0.10 mg per 100 ml and G_{10} (*Tridax procumbens* aqueous, 2 ml) 0.09 mg/100 ml.

The vitamin A content was found to be significant among the treatment during second month after storage. Vitamin A content of liquidized aloe gel juice added with herbal extracts and essential oil ranged from 0.07 to 0.11 mg/100 ml.Vitamin A content of liquidized aloe gel juice added with 1ml clove oil, G_{25} did not show any variation from zero MAS upto 3 MAS (0.11 mg/100 ml). In 2 MAS significantly higher vitamin A content was observed in liquidized aloe gel juice added with 1 ml clove oil (0.11 mg/100 ml) followed by G_2 (*Gymnema sylvestre* tincture, 1 ml), G_{20} (green tea tincture, 1 ml), G_{22} (basil oil, 1 ml) and G_{23} (lemon grass oil, 1 ml) with a vitamin A content of 0.10 mg/100 ml. The vitamin A content obtained for the control was found to be 0.09 mg/100 ml.

On third month after storage, the liquidized aloe gel juice added with herbal extracts and essential oil showed significant variation. The highest vitamin A content in the third month in were in treatments G_8 (*Achyranthes aspera* tincture 2 ml) G_{19} (green tea aqueous, 2 ml), G_{20} (green tea tincture, 1 ml), G_{22} (basil oil 1 ml) and G_{25} (clove oil 1 ml) followed by G_1 (*Gymnema sylvestre* aqueous 1 ml), G_2 (*Gymnema sylvestre* tincture 1 ml) , G_5 (*Centella asiatica* tincture 2 ml), G_{11} (*Tridax procumbens* tincture 2 ml), G_{12} (*Tridax procumbens* decoction 1 ml), G_{17} (*Punica granatum* tincture,2 ml), G_{18} (*Punica granatum* decoction,1 ml), G_{21} (green tea decoction, 2 ml), G_{23} (lemon grass oil, 1 ml), G_{24} (cinnamon bark oil, 1 ml) and G_{27} (control – sodium benzoate) with 0.09 and 0.08 mg/100 ml respectively.

The vitamin A content was found to be significantly different among the treatment during fourth month after storage. The vitamin A content of the treatment G_{25} (clove oil 1 ml) was 0.09 mg/100 ml which was significantly higher than G_{24} (cinnamon bark oil, 1 ml) with a vitamin content of 0.08 mg/100 ml. The vitamin A content obtained for the control was found to be 0.06 mg/100 ml.

On fifth month after storage the treatments were significantly variable in vitamin A content. The highest value of 0.07 mg/100 ml was noticed in the treatments G_{19} (green tea aqueous, 2 ml), G_{20} (green tea tincture, 1 ml) and G_{25} (clove oil, 1 ml) which differed significantly from G_5 (*Centella asiatica* tincture 2 ml), G_8 (*Achyranthes aspera* tincture, 2 ml), G_{12} (*Tridax procumbens* decoction 1 ml) and G_{24} (cinnamon bark oil, 1 ml) with a value of 0.06 mg/100 ml and the control G_{27} showed a value of 0.05 mg/100 ml.

The vitamin A content was found to be significant among the treatments during sixth month after storage. Significantly higher vitamin A content was noticed in G_{25} (clove oil, 1 ml) with vitamin A content of 0.07 mg/100 ml followed by, G_{19} (green tea aqueous 2 ml) and G_{20} (green tea tincture 1 ml) with a vitamin A content of 0.06 mg per 100 ml. The vitamin A content obtained for the control was found to be 0.03 mg/100 ml.

High oxygen concentrations have been associated with high levels of carotenoid degradation during storage in dried sweet potato flakes (Emenhiser *et al*., 1999), semi-preserved tomato sauces (Baiano *et al*., 2005) and in pasteurised mango puree (Caicedo *et al*., 2007). According to Penicaud *et al*. (2011) β - carotene was more sensitive to isomerisation. The heat treatment imparted changes or deformation of the physical ultrastructure of the cell wall and organelles, and β - carotene with its two bulky β-ionone rings could hardly reorganise and bear these changes. The occurrence of oxygen during processing has also been investigated by Hiranvarachat *et al*. (2008). It was found that low-pressure superheated steam drying and vacuum drying led to less degradation of β-carotene in carrot than hot air drying because this last process was the most aerobic.

The reduction in vitamin A over a period of storage shows the degradation of vitamin A on storage which might have taken place due to structural changes and due to the reduction of oxygen level around the sample.

Effect of herbal extracts and essential oils on vitamin C content of liquidized aloe gel juice are presented in table 61.

The vitamin C content was found to be significant among the treatments during zero month after storage. Significantly higher vitamin C content was observed in G_{19} (green tea aqueous, 2 ml) with vitamin C content of 40.07 mg/100 ml followed by G_{21} (green tea decoction, 2 ml) with vitamin C content of 37.84 mg/100 ml. The vitamin C content obtained for the control was found to be 23.79 mg/100 ml.

Vitamin C content of liquidized aloe gel juice added with herbal extracts and essential oil on the first month after storage showed significant variation. Significantly higher vitamin C content of 38.17 mg per 100 ml was noticed in the treatment G_{19} (green tea aqueous 2 ml) followed by G_{21} (green tea decoction 2 ml) with vitamin C content of 35.42 mg/100 ml. The vitamin C content of G_{27} (control – sodium benzoate) recorded was 22.72 mg/100 ml.

On second month after storage the vitamin C content of treatments were significantly variable among themselves. Vitamin C content was significantly higher (35.11 mg per 100 ml) in the treatment G_{19} (green tea aqueous 2 ml) followed by G_{21} (green tea decoction 2 ml) with a value of 30.92 mg/100 ml. The vitamin C content of control G_{27} (sodium benzoate) was 20.74 mg/100 ml.

The vitamin C content was found to be significantly varying among the treatments during third month after storage. G_{19} (green tea aqueous, 2 ml) recorded higher vitamin C content of 30.17 mg/100 ml followed by G_{21} (green tea decoction 2 ml) containing 28.63 mg per 100 ml of vitamin C. The vitamin C content obtained for the control was 19.64 mg/100 ml.

During fourth month after storage the vitamin C content was found to be significant among treatments. In the treatment G_{21} (green tea decoction, 2ml) the vitamin C content recorded was 25.43 mg/100 ml followed by G_{19} (green tea aqueous, 2 ml) with vitamin C content of 25.12 mg/100 ml. The vitamin C content in control was 15.24 mg/100 ml.

The vitamin C content of the treatments at fifth month after storage varied significantly between treatments. Vitamin C content was significantly higher in G_{25} (clove oil, 1 ml) which recorded 20.83 mg/100ml followed by G_{19} (green tea aqueous, 2ml) having a value of 17.17 mg/100 ml while the vitamin C content for the control was 13.87 mg/100 ml.

The vitamin C content was found to be significantly decreasing during sixth month after storage. Liquidized aloe gel juice with 1 ml clove oil (G_{25}) showed vitamin C content of 20.78 mg/100 ml followed by G_{22} (basil oil, 1ml) with 14.87 mg/100 ml. The vitamin C content obtained for the control was found to be 11.90 mg/100 ml.

Among the treatments, G_{25} (clove oil, 1ml) showed least variation in vitamin C content between the months showing vitamin C content of 23.78, 23.77, 22.18, 21.72, 21.64, 20.83 and 20.78 during zero MAS, 1 MAS, 2 MAS, 3 MAS, 4 MAS, 5 MAS and 6 MAS respectively.

In the present study the vitamin C content showed a decreasing trend during storage. The following studies supported the present result about vitamin content and degradation of aloe juice during storage. Ascorbic acid transformed to diketoglutaric acid due to reaction with air and metal ions may contribute to the losses of ascorbic acid (Harris, 1975 and Addo, 1981). Ascorbic acid is water soluble and it is readily lost via leaching from cut or bruised surfaces of raw material which results in chemical degradation of the product (Tannenbaum *et al*., 1985). Vitamin C content of all roselle-fruit blends decreased during storage with the advancement of storage period, which was probably due to the sensitivity of vitamin C to oxygen, light and heat (Ziena, 2000). The retention of vitamin C used as an estimate for the overall nutrient retention of food products because it is a least stable nutrient and is highly sensitive to oxidation and leach into water soluble media during storage (Franke *et al*., 2004). Jain and Khurdiya, (2009) observed the loss in vitamin C during storage of aonla juice. Vitamin C content of *Aloe* gel was 47 – 61 mg per 100 ml (Chandegara and Varshney, 2013). In the present study the vitamin C content ranged from 17.56 mg/100ml to 40.07 mg/100 ml.

Table 59. Effect of herbal extracts and essential oils on amino acids of liquidized aloe gel juice, ppm

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.03	0.03	0.03	0.03	0.02	0.02	0.01
G_2 (*Gymnema sylvestre* tincture1ml)	0.04	0.03	0.03	0.03	0.03	0.02	0.02
G_3 (*Gymnema sylvestre* decoction 2ml)	0.03	0.03	0.03	0.02	0.02	0.02	0.01
G_4 (*Centella asiatica* aqueous 1ml)	0.04	0.04	0.04	0.03	0.03	0.03	0.03
G_5 (*Centella asiatica* tincture 2ml)	0.04	0.04	0.04	0.04	0.03	0.03	0.03
G_6 (*Centella asiatica* decoction 1ml)	0.04	0.04	0.04	0.03	0.03	0.03	0.03
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.08	0.08	0.08	0.06	0.03	0.02	0.01
G_8 (*Achyranthes aspera* tincture 2ml)	0.08	0.08	0.08	0.06	0.04	0.03	0.01
G_9 (*Achyranthes aspera* decoction 1.50 ml)	0.08	0.08	0.07	0.04	0.03	0.03	0.01
G_{10} (*Tridax procumbens* aqueous 2ml)	0.05	0.05	0.04	0.04	0.04	0.03	0.02
G_{11} (*Tridax procumbens* tincture 2ml)	0.05	0.05	0.05	0.04	0.04	0.04	0.03
G_{12} (*Tridax procumbens* decoction 1ml)	0.05	0.05	0.04	0.04	0.04	0.03	0.02
G_{13} (*Terminalia chebula* aqueous 1ml)	0.05	0.04	0.04	0.04	0.03	0.03	0.02
G_{14} (*Terminalia chebula* tincture 1ml)	0.05	0.05	0.05	0.04	0.03	0.03	0.03
G_{15} (*Terminalia chebula* decoction 1ml)	0.05	0.05	0.04	0.04	0.03	0.03	0.02

Table 59. Effect of herbal extracts and essential oils on amino acids of liquidized aloe gel juice, ppm (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.07	0.07	0.07	0.07	0.06	0.05	0.05
G_{17} (*Punica granatum* tincture 2ml)	0.07	0.07	0.07	0.07	0.06	0.05	0.05
G_{18} (*Punica granatum* decoction 1ml)	0.07	0.07	0.07	0.06	0.06	0.05	0.05
G_{19} (green tea aqueous 2ml)	0.03	0.03	0.02	0.02	0.01	0.01	0.01
G_{20} (green tea tincture 1ml)	0.03	0.03	0.02	0.02	0.02	0.02	0.01
G_{21} (green tea decoction 2ml)	0.03	0.03	0.02	0.02	0.02	0.01	0.01
G_{22} (basil oil 1ml)	0.04	0.04	0.04	0.04	0.03	0.03	0.03
G_{23} (lemon grass oil 1ml)	0.05	0.04	0.04	0.04	0.04	0.03	0.03
G_{24} (cinnamon bark oil 1ml)	0.04	0.04	0.04	0.04	0.03	0.03	0.03
G_{25} (clove oil 1ml)	0.04	0.04	0.04	0.04	0.04	0.04	0.04
G_{26} (cardamom oil 1ml)	0.04	0.04	0.04	0.04	0.03	0.03	0.03
G_{27} (control - sodium benzoate 1ml)	0.04	0.04	0.03	0.03	0.03	0.02	0.01
SEm (±)	0.000	0.000	0.000	0.000	0.000	0.000	0.000
CD (0.05)	0.001	0.001	0.001	0.001	0.001	0.001	0.001

*Each value is the mean of five replication

Table 60. Effect of herbal extracts and essential oils on vitamin A of liquidized aloe gel juice, mg/100 ml

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.11	0.11	0.09	0.08	0.05	0.03	0.02
G_2 (*Gymnema sylvestre* tincture1ml)	0.11	0.11	0.10	0.08	0.06	0.05	0.03
G_3 (*Gymnema sylvestre* decoction 2ml)	0.11	0.11	0.08	0.06	0.04	0.03	0.02
G_4 (*Centella asiatica* aqueous 1ml)	0.11	0.11	0.08	0.05	0.04	0.03	0.02
G_5 (*Centella asiatica* tincture 2ml)	0.11	0.11	0.09	0.08	0.07	0.06	0.05
G_6 (*Centella asiatica* decoction 1ml)	0.11	0.11	0.08	0.07	0.06	0.05	0.02
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.11	0.11	0.09	0.06	0.05	0.05	0.03
G_8 (*Achyranthes aspera* tincture 2ml)	0.11	0.11	0.09	0.09	0.07	0.06	0.05
G_9 (*Achyranthes aspera* decoction 1.50 ml)	0.11	0.10	0.08	0.07	0.06	0.05	0.04
G_{10} (*Tridax procumbens* aqueous 2ml)	0.11	0.09	0.08	0.06	0.05	0.04	0.03
G_{11} (*Tridax procumbens* tincture 2ml)	0.11	0.11	0.09	0.08	0.06	0.05	0.05
G_{12} (*Tridax procumbens* decoction 1ml)	0.11	0.11	0.09	0.08	0.06	0.06	0.04
G_{13} (*Terminalia chebula* aqueous 1ml)	0.11	0.11	0.09	0.07	0.06	0.04	0.02
G_{14} (*Terminalia chebula* tincture 1ml)	0.11	0.11	0.09	0.07	0.06	0.05	0.04
G_{15} (*Terminalia chebula* decoction 1ml)	0.11	0.11	0.09	0.07	0.06	0.04	0.03

Table 60. Effect of herbal extracts and essential oils on vitamin A content of liquidized aloe gel juice, mg/100 ml (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.11	0.10	0.07	0.05	0.04	0.02	0.01
G_{17} (*Punica granatum* tincture 2ml)	0.11	0.11	0.09	0.08	0.07	0.05	0.04
G_{18} (*Punica granatum* decoction 1ml)	0.11	0.11	0.09	0.08	0.06	0.04	0.03
G_{19} (green tea aqueous 2ml)	0.12	0.11	0.09	0.09	0.07	0.07	0.06
G_{20} (green tea tincture 1ml)	0.12	0.11	0.10	0.09	0.07	0.07	0.06
G_{21} (green tea decoction 2ml)	0.11	0.11	0.09	0.08	0.07	0.05	0.03
G_{22} (basil oil 1ml)	0.11	0.10	0.10	0.09	0.06	0.04	0.03
G_{23} (lemon grass oil 1ml)	0.11	0.11	0.10	0.08	0.06	0.04	0.02
G_{24} (cinnamon bark oil 1ml)	0.12	0.11	0.09	0.08	0.08	0.06	0.03
G_{25} (clove oil 1ml)	0.11	0.11	0.11	0.09	0.09	0.07	0.07
G_{26} (cardamom oil 1ml)	0.11	0.11	0.09	0.07	0.06	0.04	0.02
G_{27} (control - sodium benzoate 1ml)	0.12	0.11	0.09	0.08	0.06	0.05	0.03
SEm (±)	0.000	0.000	0.000	0.000	0.000	0.000	0.000
CD (0.05)	0.001	0.001	0.001	0.001	0.001	0.001	0.001

*Each value is the mean of five replication

Table 61. Effect of herbal extracts and essential oils on vitamin C content of liquidized aloe gel juice, mg/100 ml

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	17.56	15.57	13.54	11.21	9.76	8.93	7.77
G_2 (*Gymnema sylvestre* tincture1ml)	20.82	18.75	16.71	15.41	13.91	12.72	11.84
G_3 (*Gymnema sylvestre* decoction 2ml)	18.83	17.24	15.81	11.34	9.57	8.82	7.13
G_4 (*Centella asiatica* aqueous 1ml)	17.84	15.83	13.24	10.04	8.27	6.94	5.32
G_5 (*Centella asiatica* tincture 2ml)	23.48	21.95	19.24	18.12	17.30	15.22	14.86
G_6 (*Centella asiatica* decoction 1ml)	19.87	18.91	15.62	13.32	10.22	8.92	6.82
G_7 (*Achyranthes aspera* aqueous 2 ml)	17.75	15.81	14.24	12.92	10.25	8.82	7.21
G_8 (*Achyranthes aspera* tincture 2ml)	23.80	21.21	19.15	17.11	15.16	14.81	13.81
G_9 (*Achyranthes aspera* decoction 1.50 ml)	18.89	17.79	16.78	14.31	10.23	8.25	7.42
G_{10} (*Tridax procumbens* aqueous 2ml)	19.71	18.82	17.16	15.51	10.17	8.17	6.91
G_{11} (*Tridax procumbens* tincture 2ml)	20.64	19.81	18.16	16.12	13.17	11.16	9.32
G_{12} (*Tridax procumbens* decoction 1ml)	19.40	18.41	17.19	16.15	14.51	10.27	7.96
G_{13} (*Terminalia chebula* aqueous 1ml)	20.80	19.15	18.43	16.11	15.31	13.16	11.90
G_{14} (*Terminalia chebula* tincture 1ml)	29.76	28.11	27.14	23.11	20.52	16.17	14.86
G_{15} (*Terminalia chebula* decoction 1ml)	19.79	18.71	17.13	16.15	14.12	12.97	10.41

Table 61. Effect of herbal extracts and essential oils on vitamin C content of liquidized aloe gel juice, mg/100 ml (continued)

G_{16} (*Punica granatum* aqueous 1ml)	21.43	20.18	18.72	15.54	12.11	10.13	7.93
G_{17} (*Punica granatum* tincture 2ml)	24.80	23.11	22.18	20.71	18.64	15.32	12.78
G_{18} (*Punica granatum* decoction 1ml)	20.44	19.31	18.15	15.13	12.11	10.34	8.12
G_{19} (green tea aqueous 2ml)	40.07	38.17	35.11	30.17	25.12	17.17	7.93
G_{20} (green tea tincture 1ml)	24.60	22.17	20.18	18.36	15.12	13.24	11.89
G_{21} (green tea decoction 2ml)	37.84	35.42	30.92	28.63	25.43	16.15	8.96
G_{22} (basil oil 1ml)	23.80	22.12	20.44	18.24	17.32	15.18	14.87[b]
G_{23} (lemon grass oil 1ml)	29.75	28.22	27.15	23.22	20.14	17.12	14.80[b]
G_{24} (cinnamon bark oil 1ml)	23.80	22.78	21.25	19.14	17.63	14.14	11.89
G_{25} (clove oil 1ml)	23.78	23.77	22.18	21.72	21.64	20.83	20.78
G_{26} (cardamom oil 1ml)	23.80	22.64	21.91	19.81	16.54	13.51	11.93
G_{27} (control - sodium benzoate 1ml)	23.79	22.72	20.74	19.64	15.24	13.87	11.90
SEm (±)	0.177	0.054	0.056	0.040	0.016	0.012	0.028
CD (0.05)	0.490	0.148	0.154	0.110	0.044	0.033	0.081

*Each value is the mean of five replication

4.3.2.9 Enzymes

4.3.2.9.1 Amylase

Effect of herbal extracts and essential oils on amylase (unit per 100 ml) of liquidized aloe gel juice are presented in table 62.

The amylase activity was found to be significant among the liquidized aloe gel juice added with herbal extracts and essential oils during zero month after storage. Amylase activity was the highest in G_{25} with 1 ml clove oil (1560.60 unit per 100 ml) which was significantly different from G_{23} (lemon grass oil, 1ml) which possessed 1556.00 unit amylase per 100 ml. The amylase activity among treatments with essential oils ranged from 1541.60 unit per 100 ml in G_{26} (cardamom oil, 1 ml) to 1560.60 in G_{25} (clove oil, 1 ml).The amylase activity among treatments with herbal extracts ranged from 1315.20 in G_7 (*Achyranthes aspera* aqueous, 2 ml) to 1498.00 in G_{20} (green tea tincture, 1 ml). The control G_{27} showed amylase activity of 1435.40 unit per 100 ml.

On first month after storage the amylase activity varied significantly between treatments. The highest amylase activity was found in G_{25} (clove oil, 1 ml) with 1559.80 unit per 100 ml followed by G_{23} (lemon grass oil, 1 ml) with an amylase activity of 1550.40 unit per 100 ml. The control G_{27} recorded an amylase activity of 1429.60 unit per 100ml. Amylase activity of liquidized aloe gel juice added with essential oil ranged from 1539.40 to 1559.80 unit per 100ml.

Significant difference among the treatments was noticed in amylase activity during second month after storage. Significantly higher amylase activity was noticed in G_{25} treatment with 1 ml clove oil (1541.40 unit per 100 ml) followed by G_{26} treatment with 1ml cardamom oil (1528.40 unit per 100 ml). The amylase activity of liquidized aloe gel juice added with essential oils such as lemon grass oil, 1ml ,G_{23} was 1491.20 while that for basil oil,1 ml (G_{22}) was 1499.49 unit per 100 ml, cinnamon bark oil, 1 ml (G_{24})was 1520.60, cardamom oil, 1 ml (G_{26}) was 1539.40

and that for clove oil, 1ml (G_{25}) was 1541.40.The control G_{27} had an amylase activity of 1400.80 unit per 100 ml.

On third month after storage the treatments showed significant variation with respect to amylase activity. The highest amylase content among the treatments was found in G_{25} (clove oil 1 ml) with 1533.80 unit per 100 ml followed by G_{26} (cardamom oil, 1ml) with 1517.60 unit per 100 ml. The control G_{27} showed an amylase activity of 1324.20 unit per 100 ml. The essential oil treated samples showed a range from 1485.40 in G_{23} (lemon grass oil, 1 ml) to 1533.80 in G_{25} (clove oil 1 ml).

Liquidized aloe gel juice added with herbal extracts and essential oil did not show significant difference in amylase activity between treatments during fourth month after storage.

On fifth month after storage the treatments were found to be significantly variable. Significantly higher amylase activity was found in G_{25} (clove oil 1 ml) with 1499.60 unit per 100 ml followed by G_{26} (cardamom oil 1 ml) with 1491.40 unit per 100 ml. The control G_{27} showed amylase content of 1210.80 unit per 100 ml. The essential oil treated samples showed a range of 1401.60 unit per 100 ml in G_{22} (basil oil, 1 ml) to 1499.60 in G_{25} (clove oil, 1 ml).

Significant difference among the treatments was noticed in amylase activity during sixth month after storage. Significantly higher amylase activity of 1491.20 unit per 100 ml was noticed in G_{25} (clove oil, 1 ml) followed by G_{23} (lemon grass oil, 1ml) with 1460.40 unit amylase per 100 ml. The amylase activity among treatments with essential oils was ranged from 1338.80 in G_{24} (cinnamon bark oil, 1 ml) to 1491.20 in G_{25} (clove oil, 1 ml). The control G_{27} showed an amylase activity of 1135.40 units per 100 ml.

4.3.2.9.2 Lipase

Effect of herbal extracts and essential oils on lipase content of liquidized aloe gel juice are presented in table 63.

The lipase activity was found to be significant among the herbal extracts and essential oil added to liquidized aloe gel juice during zero month after storage. Lipase activity in G_{22} (basil oil, 2 ml) was 839.60 unit per 100 ml followed by G_{20} ,1 ml green tea tincture (809.40 unit per 100ml). The lipase content in the treatments with essential oils were ranged from 689.60 unit per 100ml in G_{24}, cinnamon bark oil, 1ml to 839.60 per 100 ml in G_{22} (basil oil, 1ml). The lipase activity of liquidized aloe gel juice added with herbal extracts ranged from 559.60 to 809.40 unit per 100 ml. The lipase activity in control was found to be 659.60 unit per 100 ml.

On first month after storage the treatments showed significant variation in lipase activity. The highest lipase activity of 837.60 unit per 100 ml was noticed in G_{22} (basil oil, 2 ml) followed by G_{20} (green tea tincture, 1 ml) having 799.20 unit per 100ml. The essential oils treated samples showed the lipase content ranging from 686.20 unit per 100ml in G_{24} (cinnamon bark oil,1 ml) to G_{22}, basil oil, 1 ml (837.60 unit per 100 ml).The lipase activity of control treatment was 656.40 unit per 100 ml.

The lipase content during second month after storage was found to be significantly different. Significantly higher lipase activity was noticed in G_{22} (basil oil, 2ml) with a value of 826.60 unit per 100 ml which was followed by G_{20} (green tea tincture, 1 ml) with 788.20 unit per 100 ml and G_{25} (clove oil, 1 ml) with 775.80 unit per 100 ml. The lipase content in the treatments with essential oils were ranged from 681.60 unit per 100ml in G_{24}, cinnamon bark oil, 1ml to 826.60 unit per 100 ml in G_{22} (basil oil, 1 ml). The lipase activity per 100 ml in control was found to be 618.60.

On third month after storage the treatments differed significantly for lipase activity. Lipase activity on third month after storage was the highest (800.80 unit per 100 ml) in G_{22} (basil oil, 2 ml) followed by1 mlclove oil, G_{25} (759.60 unit per 100 ml). The essential oils treated samples showed the lipase activity of 671.60 unit per 100 ml in G_{24} (cinnamon bark oil, 1ml) to 800.80 unit per 100ml in G_{22}, basil oil, 1 ml. The control with sodium benzoate showed lipase activity of 588.60 unit per 100 ml.

The lipase activity among the treatments was found to be significantly different during fourth month after storage. Highest lipase activity of 772.60 unit per 100 ml was noticed in G_{22} (basil oil, 2 ml) followed by G_{25} with a value of 734.40 unit per 100 ml. The lipase activity in the treatments with essential oils ranged from 600.80 unit per 100 ml in G_{24}, cinnamon bark oil, 1 ml to 772.60 unit per 100 ml in G_{22} (basil oil, 1 ml). The lipase activity in control was found to be 551.40 unit per 100 ml.

On fifth month after storage also the treatments showed significant variation. The lipase activity was highest (700.60 unit per 100 ml) in G_{22} (basil oil, 2 ml) which was on par with G_{25} (clove oil, 1 ml) having 700.40 unit per 100 ml. The lipase activity of essential oils treated samples were 571.40 unit per 100 ml in G_{24} (cinnamon bark oil, 1 ml), 616.60 unit per 100 ml in G_{23} (lemon grass oil, 1 ml), 651.60 unit per 100 ml in G_{26} (cardamom oil,1 ml), 700.40 unit per 100 ml in G_{25} (clove oil 1 ml) and G_{22}, basil oil, 1 ml (700.60 unit per 100 ml). Lipase activity of control was 500.60 unit per 100 ml.

During the final sixth month after storage the values decreased and recorded significant variations. Lipase activity of the treatment G_{25} (clove oil, 1 ml) was 679.60 unit per 100 ml which was followed by G_{22} (basil oil, 1ml) with 678.20 unit per 100ml. The essential oils treated samples showed a range from 509.60 unit per 100ml in G_{24} (cinnamon bark oil, 1ml) to G_{25}, clove oil, 1ml (679.60 unit per 100ml). The lipase activity of control was 479.40 unit per 100 ml.

Gowda *et al*. (1979) opined that viscous pseudoplastic nature of *Aloe vera* gel lost shortly after extraction due to enzymatic degradation. Wide variation in enzyme reduction was shown when herbal extracts were added and it reduced the nutritional quality of the samples. Amylases have a wide range of application in various industries such as in the food, bread making, paper industries, textiles, sweeteners, glucose and fructose syrups, fruit juices, detergents, fuel ethanol from starches, alcoholic beverages, digestive aid and spot remover in dry cleaning and also α - amylases are being used in clinical, medicinal, and analytical chemistry (Pandey *et al*., 2000). Chandegara and Varshney (2013) reported amylase content of 1100 – 1600 units and lipase 600 – 800 units in aloe gel. The amylase content reported to vary from 1315.20 to 1560.60 unit per 100 ml and lipase ranged from 559.60 to 789.60 unit per 100ml during zero month after storage during present study among the samples.

Table 62. Effect of herbal extracts and essential oils on amylase of liquidized aloe gel juice, unit per 100ml

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	1381.80	1379.60	1361.20	1295.60	1248.60	1196.20	1151.20
G_2 (*Gymnema sylvestre* tincture1ml)	1387.60	1385.60	1368.60	1298.20	1254.20	1198.40	1165.60
G_3 (*Gymnema sylvestre* decoction 2ml)	1383.60	1378.80	1357.60	1293.60	1249.60	1194.20	1155.60
G_4 (*Centella asiatica* aqueous 1ml)	1396.20	1389.60	1344.20	1284.40	1241.60	1181.60	1129.60
G_5 (*Centella asiatica* tincture 2ml)	1436.40	1415.60	1353.60	1298.60	1252.40	1195.60	1134.40
G_6 (*Centella asiatica* decoction 1ml)	1398.00	1381.60	1346.40	1286.80	1244.20	1188.20	1130.80
G_7 (*Achyranthes aspera* aqueous 2 ml)	1315.20	1292.80	1284.60	1251.20	1184.60	1191.60	1148.40
G_8 (*Achyranthes aspera* tincture 2ml)	1324.40	1313.60	1297.60	1283.60	1213.60	1198.60	1152.40
G_9 (*Achyranthes aspera* decoction 1.50 ml)	1319.60	1299.80	1286.60	1258.20	1208.20	1193.60	1150.60
G_{10} (*Tridax procumbens* aqueous 2ml)	1338.60	1300.40	1290.60	1262.60	1211.00	1193.60	1161.00
G_{11} (*Tridax procumbens* tincture 2ml)	1347.60	1339.60	1300.40	1284.60	1220.80	1189.40	1171.00
G_{12} (*Tridax procumbens* decoction 1ml)	1339.60	1300.40	1291.00	1263.60	1213.60	1194.80	1162.80
G_{13} (*Terminalia chebula* aqueous 1ml)	1371.60	1370.00	1340.60	1291.20	1212.60	1180.60	1161.60
G_{14} (*Terminalia chebula* tincture 1ml)	1389.40	1384.40	1381.20	1312.80	1289.60	1203.60	1196.40
G_{15} (*Terminalia chebula* decoction 1ml)	1370.40	1368.60	1339.40	1292.60	1210.00	1182.00	1163.20

Table 62. Effect of herbal extracts and essential oils on amylase of liquidized aloe gel juice, unit per 100ml (continued)

G_{16} (*Punica granatum* aqueous 1ml)	1389.60	1380.60	1341.60	1280.60	1238.60	1201.00	1153.60
G_{17} (*Punica granatum* tincture 2ml)	1410.80	1409.40	1400.20	1382.60	1270.40	1219.60	1173.20
G_{18} (*Punica granatum* decoction 1ml)	1393.80	1389.60	1361.20	1290.60	1239.60	1202.80	1154.80
G_{19} (green tea aqueous 2ml)	1480.80	1477.60	1419.20	1359.80	3637.20	1297.80	1167.00
G_{20} (green tea tincture 1ml)	1498.00	1494.00	1428.20	1371.00	1318.60	1251.60	1192.80
G_{21} (green tea decoction 2ml)	1493.40	1479.60	1424.40	1364.80	1306.20	1230.80	1170.60
G_{22} (basil oil 1ml)	1548.60	1539.60	1499.40	1488.20	1436.40	1401.60	1345.80
G_{23} (lemon grass oil 1ml)	1556.00	1550.40	1491.20	1485.40	1479.60	1471.60	1460.40
G_{24} (cinnamon bark oil 1ml)	1554.40	1549.40	1520.60	1499.20	1461.80	1402.00	1338.80
G_{25} (clove oil 1ml)	1560.60	1559.80	1541.40	1533.80	1518.40	1499.60	1491.20
G_{26} (cardamom oil 1ml)	1541.60	1539.40	1528.40	1517.60	1502.60	1491.40[b]	1449.60
G_{27} (control - sodium benzoate 1ml)	1435.40	1429.60	1400.80	1324.20	1292.60	1210.80	1135.40
SEm (±)	0.340	0.351	0.332	0.306	0.000	0.315	0.303
CD (0.05)	0.942	0.972	0.921	0.847	NS	0.873	0.850

*Each value is the mean of five replication

Table 63. Effect of herbal extracts and essential oils on lipase of liquidized aloe gel juice, unit per 100ml

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	559.60	569.20	499.60	474.40	424.60	309.20	301.00
G_2 (*Gymnema sylvestre* tincture1ml)	600.60	597.60	581.60	564.60	542.60	489.60	459.40
G_3 (*Gymnema sylvestre* decoction 2ml)	564.60	562.60	553.60	509.60	449.60	401.60	311.20
G_4 (*Centella asiatica* aqueous 1ml)	579.60	575.60	559.80	499.40	469.60	404.60	359.60
G_5 (*Centella asiatica* tincture 2ml)	600.00	597.40	589.60	574.60	544.80	504.60	479.00
G_6 (*Centella asiatica* decoction 1ml)	593.80	590.60	519.60	449.60	424.60	396.80	365.00
G_7 (*Achyranthes aspera* aqueous 2 ml)	699.20	696.80	670.60	623.60	550.40	409.00	397.40
G_8 (*Achyranthes aspera* tincture 2ml)	719.60	717.40	681.60	623.60	581.60	509.20	418.20
G_9 (*Achyranthes aspera* decoction 1.50 ml)	704.60	700.80	694.40	619.60	542.60	497.60	401.40
G_{10} (*Tridax procumbens* aqueous 2ml)	621.40	619.60	599.80	561.60	509.40	464.60	396.60
G_{11} (*Tridax procumbens* tincture 2ml)	639.60	637.60	616.80	582.60	524.60	474.60	409.20
G_{12} (*Tridax procumbens* decoction 1ml)	624.60	621.20	597.60	562.80	521.20	471.60	401.60
G_{13} (*Terminalia chebula* aqueous 1ml)	651.60	649.60	627.40	594.60	544.20	496.80	424.60
G_{14} (*Terminalia chebula* tincture 1ml)	659.60	657.60	646.60	598.20	574.60	517.80	439.60
G_{15} (*Terminalia chebula* decoction 1ml)	649.20	646.60	627.60	595.00	541.60	498.20	426.60

Table 63. Effect of herbal extracts and essential oils on lipase of liquidized aloe gel juice, unit per 100ml (continued)

G_{16} (*Punica granatum* aqueous 1ml)	587.80	584.60	511.40	500.20	481.60	444.20	409.60
G_{17} (*Punica granatum* tincture 2ml)	599.20	597.60	581.60	533.80	496.60	451.60	419.40
G_{18} (*Punica granatum* decoction 1ml)	589.60	585.60	102.00	498.40	484.60	448.40	411.20
G_{19} (green tea aqueous 2ml)	749.40	746.60	709.60	680.40	607.80	562.20	429.60
G_{20} (green tea tincture 1ml)	809.40	799.20	788.20	754.60	696.60	600.80	509.00
G_{21} (green tea decoction 2ml)	749.60	746.60	711.60	686.60	609.40	572.40	478.60
G_{22} (basil oil 1ml)	839.60	837.60	826.60	800.80	772.60	700.60	678.20
G_{23} (lemon grass oil 1ml)	739.40	737.80	724.20	703.00	661.40	616.60	569.60
G_{24} (cinnamon bark oil 1ml)	689.60	686.20	681.60	671.60	600.80	571.40	509.60
G_{25} (clove oil 1ml)	789.60	787.40	775.80	759.60	734.40	700.40	679.60
G_{26} (cardamom oil 1ml)	779.40	775.20	762.80	727.60	700.80	651.60	569.60
G_{27} (control - sodium benzoate 1ml)	659.60	656.40	618.60	588.60	551.40	500.60	479.40
SEm (±)	0.283	0.302	0.312	0.380	0.304	0.374	0.315
CD (0.05)	0.784	0.837	0.874	1.05	0.843	1.037	0.889

*Each value is the mean of five replication

4.3.2.10 Protein

The effect of herbal extracts and essential oils on amino acids content of liquidized aloe gel juice is presented in table 64.

The protein content was found to be significant among the treatments during zero month after storage. Significantly higher protein content (0.26 per cent) was noticed in G_{27} (sodium benzoate 1 ml) followed G_{11} (*Tridax procumbens* tincture, 2 ml), G_{19} (green tea aqueous, 2 ml), G_{20} (green tea tincture, 1 ml), G_{22} (basil oil 1 ml), G_{23}(lemon grass oil 1 ml) and G_{25} (clove oil1 ml) with protien content of 0.24 per cent. The least value among the treatments were shown by G_4 (*Centella asiatica* aqueous 1ml) and G_6 (*Centella asiatica* decoction 1ml) with 0.20 per cent protein content which was on par with 0.21 per cent protein content in G_1 (*Gymnema sylvestre* aqueous 1 ml), G_5 (*Centella asiatica* tincture 2 ml), G_{10} (*Tridax procumbens* aqueous 2 ml) and G_{13} (*Terminalia chebula* aqueous 1 ml).

On first month after storage protein content of the treatments showed significant variation. Protien content was significantly higher for G_{27} (sodium benzoate 1 ml) with 0.25 per cent which was on par with G_{20} (green tea tincture, 1 ml) and G_{25} (clove oil1 ml) having a value of 0.24 per cent. The lowest protein content among the treatments were observed in G_4 (*Centella asiatica* aqueous 1 ml) and G_6 (*Centella asiatica* decoction 1 ml) having 0.19 per cent which was on par with G_5 (*Centella asiatica* tincture 1 ml), G_{10} (*Tridax procumbens* aqueous 2 ml) and G_{13} (*Terminalia chebula* aqueous 1 ml) with 0.20 per cent. The protein content of liquidized aloe gel juice with herbal extracts varied from 0.19 to 0.24 per cent while protein content of all essential oils were 0.23 and 0.24 per cent.

Protien content of the treatments varied significantly during second month of storage. Highest protein content of 0.24 per cent was noticed in G_{24} (cinnamon bark oil1 ml), G_{25} (clove oil1 ml) and G_{27} (sodium benzoate 1 ml) which was on par with G_{20} (green tea tincture, 1 ml), G_{23} (lemon grass oil, 1 ml) and G_{26} (cardamom oil 1 ml) with protein content of 0.23 per cent. The least value was noticed in G_4 (*Centella asiatica* aqueous 1 ml) with 0.16 per cent which was on par with G_6 (*Centella asiatica* decoction 1 ml) recording 0.15 per cent.

There was significant difference among the treatments during third month after storage. Highest protein content (0.24 per cent) was noticed in G_{25} (clove oil 1 ml) which was on par with G_{20} (green tea tincture 1ml), G_{23} (lemon grass oil 1ml) G_{24} (cinnamon bark oil 1 ml) and G_{26} (cardamom oil 1ml) with 0.23 per cent and G_{27} (control). The protein content was noticed least for G_4 (*Centella asiatica* aqueous 1 ml) with 0.13 per cent which was followed by G_5 (*Centella asiatica* tincture 2 ml) and G_6 (*Centella asiatica* decoction 1 ml).

Protien content showed significant difference between treatments at four month after storage. Highest protein content of 0.24 per cent was noticed inG_{25} (clove oil, 1 ml) which was on par with protein content of 0.23 per cent in G_{23} (lemon grass oil, 1 ml), G_{24} (cinnamon bark oil, 1 ml) and G_{26} (cardamom oil, 1 ml). The least protein content was recorded in G_4 (*Centella asiatica* aqueous 1 ml) and G_5 (*Centella asiatica* tincture 2 ml) with 0.12 per cent and which was on par with protein content of 0.13 per cent in G_1 (*Gymnema sylvestre aqueous* 1 ml) and G_6 (*Centella asiatica* decoction 1 ml). The control G_{27} had protein content of 0.22 per cent. The amino acid content of essential oil treated samples varied from 0.22 - 0.24 per cent.

The protein content of the treatments was significantly different at five month after storage. Highest protein content of 0.23 per cent was noticed in G_{23} (lemon grass oil1 ml), G_{25} (clove oil 1 ml) and G_{26} (cardamom oil1 ml) which was on par with G_{22} (basil oil 1 ml) and G_{24} (cinnamon bark oil 1 ml) with 0.22 per cent protein content. Protein content of control G_{27} was 0.21 per cent. The value obtained in essential oil treated samples varied from 0.22 per cent - 0.23 per cent.

Protein content of liquidized aloe gel juice added with herbal extracts and essential oil at six month after storage showed significant difference among treatments. Protien content of 0.23 per cent was observed from G_{25} (clove oil 1ml) and G_{26} (cardamom oil 1ml) which was on par with 0.22 per cent in G_{22} (basil oil 1 ml), G_{23} (lemon grass oil 1 ml) and G_{24} (cinnamon bark oil, 1 ml). The least content were observed in G_4 (*Centella asiatica* aqueous 1 ml) having a value of 0.08 per cent which was followed by G_1 (*Gymnema sylvestre* aqueous 1 ml), G_5 (*Centella asiatica* tincture 2 ml), G_6 (*Centella asiatica* decoction 1 ml) and G_{10} (*Tridax procumbens* aqueous 2 ml) with 0.10 per cent. The protein content of control G_{27} was 0.17 per cent. The protein content of essential oil treated samples ranged from 0.22 – 0.23 per cent. The protein content of treatment G_{25} clove oil 1 ml did not change and was 0.24 per cent upto fourth month after storage.

Protein content of 0.11g per 100g of aloe gel was reported by Chandegara and Varshney (2013), the protein content in present study varied due to the addition of herbal extracts and essential oils.

Protein serve as enzymatic catalyst, third highest parameter in aloe leaves (Whimey and Rolfes, 2005). Based on this study, the protein content in aloe juice samples decreased on storage with herbal extracts thus affecting the juice quality. But variation in protein content was less in aloe juice samples with essential oils.

4.3.2.11 Ash

Effect of herbal extracts and essential oils on ash content of liquidized aloe gel juice are presented in table 65.

The ash content was found to be significantly different among the treatment during zero month after storage. Significantly higher ash content was noticed in G_{23} (lemon grass oil, 1 ml) and G_{25} (clove oil, 1 ml) with 0.89 per cent followed by $G_{2,}$ *Gymnema sylvestre,* tincture, 1 ml and G_{26}, cardamom oil, 1 ml (0.82 per cent). The ash content among treatments with essential oils were found range from 0.81 per cent, G_{22} (basil oil, 1 ml) to 0.89 per cent in G_{25} (clove oil, 1 ml) while that for herbal extracts ranged from 0.60 per cent in G_{19} (green tea aqueous 2 ml) to 0.82 per cent in

G_2 (*Gymnema sylvestre* tincture, 1 ml). The control G_{27} showed an ash content of 0.81 per cent.

On first month after storage the treatments showed significant variation. The highest value among the treatments was found in G_{25} (clove oil, 1 ml) with an ash content of 0.87 per cent which was followed by G_{23} (lemon grass oil, 1 ml) with 0.86 per cent. The control G_{27} showed an ash content of 0.71 per cent. The ash content of treatments with essential oils ranged from 0.71 per cent in G_{22}, basil oil, 1 ml to 0.87 per cent in G_{25}, clove oil, 1 ml, while that for herbal extracts ranged from 0.51 per cent, G_{19} (green tea aqueous 2 ml) to 0.79 per cent, G_2 (*Gymnema sylvestre* tincture, 1 ml).

The ash content during second month after storage was found to be significant among the treatments. Significantly higher ash content was noticed in G_{25} (clove oil, 1 ml) with 0.84 per cent which was followed by $G_{23,}$ lemon grass oil, 1 ml with an ash content of 0.81 per cent. The ash content among treatments with essential oils were found range from 0.63 per cent in G_{22} (basil oil, 1 ml) to 0.84 per cent in G_{25} (clove oil, 1 ml). The ash content of herbal extracts range from 0.50 per cent, G_1 (*Gymnema sylvestre* aqueous 1 ml green tea aqueous 2ml) to 0.77 per cent, G_2 (*Gymnema sylvestre* tincture, 1 ml). The control G_{27} showed an ash content of 0.59 per cent.

The ash content on third month after storage showed significant variation among treatments. The highest ash content among the treatments was found in G_{25} (clove oil 1 ml) with 0.83 per cent followed by G_{26} (cardamom oil 1 ml) with 0.78 per cent ash content. The control G_{27} (control – sodium benzoate) showed ash content of 0.51 per cent. The liquidized aloe gel juice with essential oil showed an ash content ranging from 0.51 per cent in G_{22}, basil oil 1ml to 0.83 per cent in G_{25}, clove oil 1ml. The ash content of treatments with herbal extracts ranged from 0.31 per cent in G_4 (*Centella asiatica* aqueous 1ml) to 0.67 per cent in G_2 (*Gymnema sylvestre* tincture, 1ml).

On fourth month after storage the treatments recorded significant difference in ash content. Significantly higher ash content was recorded from G_{25} (clove oil 1 ml) with 0.80 per cent followed by G_{26} (cardamom oil, 1 ml) with 0.75 per cent. The control G_{27} recorded an ash content of 0.37 per cent. The treatments with essential oils ranged from 0.39 per cent of ash content in G_{22}, basil oil, 1 ml to 0.80 per cent in G_{25}, clove oil, 1 ml. The treatments with herbal extracts ranged from 0.25 per cent in G_4 (*Centella asiatica* aqueous 1 ml*)* to 0.52 per cent in G_{11} (*Tridax procumbens* tincture, 2 ml).

The ash content during fifth month after storage was significantly different among the treatments. Highest ash content was noticed in G_{25} (clove oil, 1 ml) with 0.77 per cent followed by G_{26} (cardamom oil, 1 ml) with 0.71 per cent. The ash content among treatments with essential oils were found to range from 0.36 per cent in G_{22} (basil oil, 1 ml) to 0.77 per cent in G_{25} (clove oil 1 ml) and that for herbal extracts ranged from 0.18 per cent in G_4 (*Centella asiatica* aqueous 1ml*)* to 0.39 per cent in G_2 (*Gymnema sylvestre* tincture 1ml). The ash content of control was 0.29 per cent.

The ash content varied significantly among treatments during sixth month after storage. Highest ash content was noticed in G_{25} (clove oil, 1ml) with 0.73 per cent followed by G_{24}, cinnamon bark oil, 1ml (0.62 per cent). The ash content in treatments with essential oils ranged from 0.19 per cent in G_{22} (basil oil, 1ml) to 0.73 per cent in G_{25} (clove oil, 1ml) while for herbal extracts the ash content ranged from 0.11 per cent in G_{10} (*Tridax procumbens* aqueous 2ml*)* to 0.22 per cent in G_5 (*Centella asiatica* tincture, 2ml and G_2 (*Gymnema sylvestre* tincture 1ml). The control G_{27} had an ash content of about 0.14 per cent.

Antia *et al*. (2006) reported that ash content is a reflection of mineral preserved in the sample, it represents total mineral content which is essential for proper functioning of tissues and act as second messengers in biological cascade mechanisms and important part of proximate analysis for nutrient evaluation. In present study ash content showed decreasing trend on storage representing degradation of the nutrients. Ash content of 0.25 per cent was reported in *Aloe vera* gel by Chandegara and Varshney (2013). Ash content in blanched aloe gel was 0.26 percent and unblanched aloe gel was 0.25 per cent as reported by Shubhra *et al* (2014). An ash content of 0.23 percent was reported by Talib *et al*. (2016) in aloe gel. Ash content in present study ranged from 0.60 to 0.89 percent during zero month after storage. The variations in ash content in the present study are due to the presence of herbal extracts and essential oils.

Table 64. Effect of herbal extracts and essential oils on protein content of liquidized aloe gel juice, percentage

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.21	0.21	0.19	0.17	0.13	0.12	0.10
G_2 (*Gymnema sylvestre* tincture1ml)	0.23	0.23	0.21	0.19	0.15	0.15	0.13
G_3 (*Gymnema sylvestre* decoction 2ml)	0.23	0.21	0.21	0.19	0.16	0.13	0.11
G_4 (*Centella asiatica* aqueous 1ml)	0.20	0.19	0.16	0.13	0.12	0.10	0.08
G_5 (*Centella asiatica* tincture 2ml)	0.21	0.20	0.19	0.15	0.12	0.12	0.10
G_6 (*Centella asiatica* decoction 1ml)	0.20	0.19	0.17	0.15	0.13	0.11	0.10
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.22	0.21	0.20	0.19	0.16	0.13	0.11
G_8 (*Achyranthes aspera* tincture 2ml)	0.23	0.22	0.21	0.18	0.17	0.14	0.12
G_9 (*Achyranthes aspera* decoction 1.50 ml)	0.22	0.21	0.20	0.19	0.17	0.13	0.11
G_{10} (*Tridax procumbens* aqueous 2ml)	0.21	0.20	0.19	0.17	0.14	0.12	0.10
G_{11} (*Tridax procumbens* tincture 2ml)	0.24	0.22	0.18	0.17	0.17	0.13	0.13
G_{12} (*Tridax procumbens* decoction 1ml)	0.23	0.22	0.19	0.17	0.16	0.14	0.12
G_{13} (*Terminalia chebula* aqueous 1ml)	0.21	0.20	0.18	0.16	0.15	0.13	0.12
G_{14} (*Terminalia chebula* tincture 1ml)	0.23	0.22	0.21	0.20	0.18	0.17	0.16
G_{15} (*Terminalia chebula* decoction 1ml)	0.22	0.21	0.21	0.19	0.16	0.14	0.12

Table 64. Effect of herbal extracts and essential oils on protein content of liquidized aloe gel juice, percentage (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.22	0.21	0.19	0.18	0.17	0.16	0.13
G_{17} (*Punica granatum* tincture 2ml)	0.23	0.22	0.21	0.19	0.19	0.16	0.15
G_{18} (*Punica granatum* decoction 1ml)	0.23	0.22	0.19	0.19	0.18	0.17	0.14
G_{19} (green tea aqueous 2ml)	0.24	0.22	0.21	0.21	0.20	0.18	0.16
G_{20} (green tea tincture 1ml)	0.24	0.24	0.23	0.23	0.20	0.19	0.18
G_{21} (green tea decoction 2ml)	0.23	0.23	0.21	0.19	0.19	0.17	0.14
G_{22} (basil oil 1ml)	0.24	0.23	0.21	0.21	0.22	0.22	0.22
G_{23} (lemon grass oil 1ml)	0.24	0.23	0.23	0.23	0.23	0.23	0.22
G_{24} (cinnamon bark oil 1ml)	0.23	0.23	0.24	0.23	0.23	0.22	0.22
G_{25} (clove oil 1ml)	0.24	0.24	0.24	0.24	0.24	0.23	0.23
G_{26} (cardamom oil 1ml)	0.23	0.23	0.23	0.23	0.23	0.23	0.23
G_{27} (control - sodium benzoate 1ml)	0.26	0.25	0.24	0.23	0.22	0.21	0.17
SEm (±)	0.003	0.002	0.002	0.002	0.002	0.002	0.002
CD (0.05)	0.008	0.006	0.006	0.007	0.007	0.007	0.007

*Each value is the mean of five replication

Table 65. Effect of herbal extracts and essential oils on ash content of liquidized aloe gel juice, percentage

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.61	0.58	0.50	0.42	0.30	0.22	0.19
G_2 (*Gymnema sylvestre* tincture1ml)	0.82	0.79	0.77	0.67	0.50	0.39	0.22
G_3 (*Gymnema sylvestre* decoction 2ml)	0.71	0.70	0.67	0.51	0.36	0.25	0.19
G_4 (*Centella asiatica* aqueous 1ml)	0.63	0.61	0.56	0.31	0.25	0.18	0.15
G_5 (*Centella asiatica* tincture 2ml)	0.81	0.80	0.75	0.61	0.41	0.34	0.22
G_6 (*Centella asiatica* decoction 1ml)	0.71	0.69	0.61	0.50	0.39	0.24	0.20
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.66	0.63	0.59	0.40	0.34	0.20	0.12
G_8 (*Achyranthes aspera* tincture 2ml)	0.70	0.70	0.67	0.62	0.43	0.32	0.20
G_9 (*Achyranthes aspera* decoction 1.50 ml)	0.69	0.64	0.59	0.43	0.37	0.21	0.14
G_{10} (*Tridax procumbens* aqueous 2ml)	0.62	0.60	0.57	0.49	0.33	0.23	0.11
G_{11} (*Tridax procumbens* tincture 2ml)	0.80	0.78	0.75	0.61	0.53	0.38	0.21
G_{12} (*Tridax procumbens* decoction 1ml)	0.65	0.63	0.59	0.47	0.31	0.20	0.13
G_{13} (*Terminalia chebula* aqueous 1ml)	0.63	0.61	0.57	0.41	0.37	0.19	0.14
G_{14} (*Terminalia chebula* tincture 1ml)	0.78	0.78	0.64	0.50	0.42	0.25	0.12
G_{15} (*Terminalia chebula* decoction 1ml)	0.67	0.62	0.59	0.41	0.32	0.23	0.16

Table 65. Effect of herbal extracts and essential oils on ash content of liquidized aloe gel juice, percentage (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.65	0.61	0.57	0.47	0.38	0.22	0.17
G_{17} (*Punica granatum* tincture 2ml)	0.75	0.75	0.68	0.59	0.42	0.29	0.15
G_{18} (*Punica granatum* decoction 1ml)	0.68	0.66	0.61	0.53	0.37	0.21	0.12
G_{19} (green tea aqueous 2ml)	0.60	0.51	0.44	0.33	0.30	0.24	0.12
G_{20} (green tea tincture 1ml)	0.78	0.77	0.71	0.62	0.43	0.29	0.16
G_{21} (green tea decoction 2ml)	0.62	0.60	0.54	0.41	0.37	0.27	0.13
G_{22} (basil oil 1ml)	0.81	0.71	0.63	0.51	0.39	0.36	0.19
G_{23} (lemon grass oil 1ml)	0.89	0.86	0.81	0.75	0.70	0.65	0.60
G_{24} (cinnamon bark oil 1ml)	0.81	0.79	0.74	0.72	0.69	0.65	0.62
G_{25} (clove oil 1ml)	0.89	0.87	0.84	0.83	0.80	0.77	0.73
G_{26} (cardamom oil 1ml)	0.82	0.80	0.79	0.78	0.75	0.71	0.60
G_{27} (control - sodium benzoate 1ml)	0.81	0.71	0.59	0.51	0.37	0.29	0.14
SEm (±)	0.003	0.002	0.003	0.002	0.002	0.002	0.002
CD (0.05)	0.008	0.007	0.008	0.007	0.007	0.007	0.007

*Each value is the mean of five replication

4.3.2.12 Calories

Effect of herbal extracts and essential oils on calorie content of liquidized aloe gel juice are presented in table 66.

The calorie content was found to be significant among the treatment during zero month after storage. Significantly higher calorie content was noticed in G_{22} (basil oil, 1 ml) with 2.88 kcal/100 g followed by 2.53 kcal/100 g in G_{27} (control). The least calorific value of 1.16 kcal/100 g was noticed in G_1 (*Gymnema sylvestre* aqueous, 1 ml) which was followed by G_6, *Centella asiatica* decoction 1 ml with 1.21 kcal/100 g. The calorie content among treatments with essential oils was found range from 2.38 kcal/100 g in G_{25} (clove oil 1 ml) to 2.88 kcal/100 g in G_{22} (basil oil, 1 ml) while that for herbal extracts ranged from 1.16 kcal/100 g in G_1 (*Gymnema sylvestre* aqueous, 1 ml) to 2.39 kcal/100 g in G_{19} (green tea aqueous 2 ml).

On first month after storage the treatments showed significant variation. The highest value among the treatments was found in G_{22} (basil oil 1 ml) with calorie content of 2.84 kcal/100 g which was followed by G_{23} (lemon grass oil, 1 ml) with 2.48 kcal/100 g. The least value was noticed in G_3 (*Gymnema sylvestre* decoction 2 ml) with 1.14 kcal/100 g which was on par with G_{10} (*Tridax procumbens* aqueous 2 ml) with 1.16 kcal/100 g. The control G_{27} showed calorie content of 2.44 kcal/100 g. The calorie content of treatments with essential oils ranged from 2.38 kcal/100 g in G_{26} (cardamom oil 1 ml) to 2.84 kcal/100g in G_{22}, basil oil, 1 ml.

The calorie content during second month after storage was found to be significant among the treatments. Significantly higher calorie content was noticed in G_{22} (basil oil 1 ml) with 2.74 kcal/100 g which was followed by $G_{23,}$ lemon grass oil, 1 ml with calorie content of 2.48 kcal/100 g. The calorie content among treatments with essential oils was found to range from 2.31 kcal/100 g in G_{26} (cardamom oil 1ml) to 2.74 kcal/100 g in G_{22} (basil oil 1ml). The calorie content of herbal extracts ranged from 0.96 kcal/100 g, G_4 (*Centella asiatica* aqueous 1ml) to 2.23 kcal/100 g in G_{20} (green tea tincture 1 ml). The control G_{27} showed calorie content of 2.46 kcal/100 g.

There was significant difference among the treatments during third month after storage. The highest calorie content among the treatments was found in G_{22} (basil oil 1 ml) with 2.68 kcal/100 g followed by G_{23} (lemon grass oil 1 ml) and G_{24} (cinnamon bark oil, 1 ml) with 2.45 kcal/100 g calorific value. The control G_{27} (control – sodium benzoate) showed calorie content of 2.33 kcal/100 g. The liquidized aloe gel juice with essential oil showed calorie content ranging from 2.31 kcal/100g in G_{26} (cardamom oil 1ml) to 2.68 kcal/100 g in G_{22}, basil oil, 1ml. The calorie content of treatments with herbal extracts ranged from 0.86 kcal/100 g in G_4 (*Centella asiatica* aqueous, 1ml) to 2.05 kcal/100 g in G_{20} (green tea tincture, 1ml).

On fourth month after storage the treatments recorded significant difference in calorie content. Significantly higher ash content was recorded from G_{22} (basil oil 1 ml) with 2.50 kcal/100 g followed by 2.42 kcal/100 g in G_{23} (lemon grass oil 1 ml). The control G_{27} recorded calorie content of 2.26 kcal/100 g. The treatments with essential oils ranged from 2.33 kcal/100 g in G_{26} (cardamom oil 1 ml) to 2.50 kcal/100 g in G_{22}, basil oil 1 ml. The treatments with herbal extracts ranged from 0.80 kcal/100 g in G_{10} (*Tridax procumbens* aqueous 2 ml) to 1.91 kcal/100 g in G_{20} (green tea tincture, 1 ml).

The calorie content during fifth month after storage was significantly different among the treatments. Highest calorie content was noticed in G_{22} (basil oil 1 ml) with 2.51 kcal/100 g followed by G_{23} (lemon grass oil, 1 ml) with 2.45 kcal/100 g. The least value of 0.62 kcal/100 g was noticed in G_4 (*Centella asiatica* aqueous, 1 ml) which was on par with G_{10} (*Tridax procumbens* aqueous, 2 ml) with 0.65 kcal/100 g. The calorie content among treatments with essential oils were found to be ranged from 2.26 kcal/100 g in G_{26} (cardamom oil 1 ml) to 2.51 kcal/100 g in G_{22} (basil oil 1 ml) and that for herbal extracts ranged from 0.62 kcal/100 g in G_4 (*Centella asiatica* aqueous, 1 ml*)* to 1.82 in G_{20} (green tea tincture, 1 ml). The calorie content of control was 2.08 kcal/100 g.

The calorie content varied significantly among treatments during sixth month after storage. Highest calorie content was noticed in G_{23} (lemon grass oil, 1 ml) with 2.44 kcal/100 g followed by G_{24}, cinnamon bark oil 1 ml (2.30 kcal/100 g). The calorie content in treatments with essential oils ranged from 2.27 kcal/100 g in G_{26} (cardamom oil 1ml) to 2.44 kcal/100 g in G_{22} (basil oil, 1 ml) while for herbal extracts the calorie content ranged from 0.51 kcal/100 g in G_4 (*Centella asiatica* aqueous, 1 ml) to 1.78 kcal/100 g in G_{20} (green tea tincture, 1 ml). The control G_{27} had calorie content of about 1.90 kcal/100 g.

The use of sonicated melon juice as substrate for *Lactobacillus casei* growth in cantaloupe melon juice and its effect on product quality revealed that the caloric value was reduced during the storage period due to sugar consumption by the remaining microorganisms (Fonteles *et al*., 2013). The study supports the present results of reduced calorific value during storage of six months. Chandegara and Varshney (2013) reported calorie content of 3.3/100 g in aloe gel.

4.3.2.13 Fat

Effect of herbal extracts and essential oils on fat content of liquidized aloe gel juice are presented in table 67.

The fat content of liquidized aloe gel juice added with herbal extracts and essential oil showed significant difference during zero month after storage. Highest fat content was noticed in G_{27} (control), G_{25} (clove oil 1 ml), G_{17} (*Punica granatum* tincture, 2 ml) and G_{11} (*Tridax procumbens* tincture 2 ml) with a value of 0.04 per cent which was followed by all the other treatments (0.03 per cent) except G_1 (*Gymnema sylvestre* aqueous 1 ml) and G_2 (*Gymnema sylvestre* tincture 1 ml) with 0.02 per cent of fat content.

The fat content of liquidized aloe gel juice with herbal extracts and essential oils were significantly different during first month after storage. Significantly higher fat content was noticed in G_{17} (*Punica granatum*, tincture 2 ml) and G_{25} (clove oil 1ml) with 0.04 per cent followed by all the other treatments with 0.03 per cent except G_3 and G_{19} with 0.02 per cent.

From second month after storage to six month after storage the fat content among the treatments was found to be non significant.

Crude fat content are major structural elements of biological membranes as phospholipids and sterols (Nelson *et al*., 2008). According to Chandegara and Varshney (2013) fat content in aloe gel was 0.09 g per 100 g. In the present study fat content reported was within the range of 0.02 per cent to 0.04 per cent. The proximate composition analysed in *Aloe barbadensis* by Haque *et al*. (2014) revealed lowest value for average crude fat content (0.27 per cent) which are universally stored forms of energy in living organisms.

Table 66. Effect of herbal extracts and essential oils on calories of liquidized aloe gel juice, kcal per 100 g

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	1.16	1.23	1.06	0.98	0.72	0.68	0.60
G_2 (*Gymnema sylvestre* tincture1ml)	1.29	1.33	1.25	1.07	0.94	0.90	0.75
G_3 (*Gymnema sylvestre* decoction 2ml)	1.33	1.14	1.14	1.04	0.97	0.74	0.62
G_4 (*Centella asiatica* aqueous 1ml)	1.22	1.18	0.96	0.86	0.81	0.62	0.51
G_5 (*Centella asiatica* tincture 2ml)	1.29	1.24	1.20	0.95	0.86	0.74	0.64
G_6 (*Centella asiatica* decoction 1ml)	1.21	1.19	1.09	0.91	0.82	0.68	0.62
G_7 (*Achyranthes aspera* aqueous 2ml)	1.28	1.23	1.18	1.05	0.94	0.73	0.65
G_8 (*Achyranthes aspera* tincture 2ml)	1.40	1.32	1.27	1.09	0.99	0.80	0.72
G_9 (*Achyranthes aspera* decoction 1.50ml)	1.30	1.24	1.20	1.04	0.91	0.74	0.66
G_{10} (*Tridax procumbens* aqueous 2ml)	1.22	1.16	1.12	0.95	0.80	0.65	0.60
G_{11} (*Tridax procumbens* tincture 2ml)	1.38	1.25	1.10	0.97	0.98	0.75	0.72
G_{12} (*Tridax procumbens* decoction 1ml)	1.31	1.24	1.14	0.99	0.90	0.74	0.66
G_{13} (*Terminalia chebula* aqueous 1ml)	1.28	1.20	1.03	0.96	0.87	0.73	0.67
G_{14} (*Terminalia chebula* tincture 1ml)	1.37	1.31	1.27	1.13	1.04	0.92	0.89
G_{15} (*Terminalia chebula* decoction 1ml)	1.32	1.25	1.14	1.06	0.90	0.76	0.69

Table 66. Effect of herbal extracts and essential oils on calories of liquidized aloe gel juice, kcal per 100 g (continued)

G_{16} (*Punica granatum* aqueous 1ml)	1.32	1.25	1.08	1.01	0.89	0.83	0.73
G_{17} (*Punica granatum* tincture 2ml)	1.42	1.40	1.28	1.13	1.05	0.87	0.84
G_{18} (*Punica granatum* decoction 1ml)	1.37	1.30	1.11	1.07	1.00	0.89	0.77
G_{19} (green tea aqueous 2ml)	2.39	2.27	2.16	1.99	1.76	1.59	1.49
G_{20} (green tea tincture 1ml)	2.32	2.31	2.23	2.05	1.91	1.82	1.78
G_{21} (green tea decoction 2ml)	2.34	2.23	1.97	1.75	1.63	1.37	1.17
G_{22} (basil oil 1ml)	2.88	2.84	2.74	2.68	2.50	2.51	2.30
G_{23} (lemon grass oil 1ml)	2.49	2.48	2.48	2.45	2.42	2.45	2.44
G_{24} (cinnamon bark oil 1ml)	2.46	2.45	2.46	2.45	2.39	2.27	2.30
G_{25} (clove oil 1ml)	2.38	2.42	2.44	2.42	2.39	2.44	2.31
G_{26} (cardamom oil 1ml)	2.39	2.38	2.31	2.31	2.33	2.26	2.27
G_{27} (control - sodium benzoate 1ml)	2.53	2.44	2.46	2.33	2.26	2.08	1.90
SEm±	0.013	0.011	0.009	0.010	0.010	0.010	0.010
CD (0.05)	0.036	0.030	0.024	0.028	0.028	0.028	0.029

*Each value is the mean of five replication

Table 67. Effect of herbal extracts and essential oils on fat content of liquidized aloe gel juice, percentage

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.02	0.03	0.03	0.02	0.01	0.01	0.01
G_2 (*Gymnema sylvestre* tincture1ml)	0.02	0.03	0.03	0.02	0.02	0.02	0.01
G_3 (*Gymnema sylvestre* decoction 2ml)	0.03	0.02	0.02	0.02	0.02	0.01	0.01
G_4 (*Centella asiatica* aqueous 1ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_5 (*Centella asiatica* tincture 2ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_6 (*Centella asiatica* decoction 1ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_8 (*Achyranthes aspera* tincture 2ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_9 (*Achyranthes aspera* decoction 1.50 ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_{10} (*Tridax procumbens* aqueous 2ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_{11} (*Tridax procumbens* tincture 2ml)	0.04	0.03	0.03	0.02	0.02	0.01	0.01
G_{12} (*Tridax procumbens* decoction 1ml)	0.03	0.03	0.03	0.02	0.01	0.01	0.01
G_{13} (*Terminalia chebula* aqueous 1ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_{14} (*Terminalia chebula* tincture 1ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_{15} (*Terminalia chebula* decoction 1ml)	0.03	0.03	0.03	0.02	0.01	0.01	0.01

Table 67. Effect of herbal extracts and essential oils on fat content of liquidized aloe gel juice, percentage (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.03	0.03	0.03	0.02	0.01	0.01	0.01
G_{17} (*Punica granatum* tincture 2ml)	0.04	0.04	0.04	0.02	0.02	0.01	0.01
G_{18} (*Punica granatum* decoction 1ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_{19} (green tea aqueous 2ml)	0.03	0.02	0.02	0.02	0.01	0.01	0.01
G_{20} (green tea tincture 1ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_{21} (green tea decoction 2ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_{22} (basil oil 1ml)	0.03	0.03	0.03	0.02	0.02	0.02	0.02
G_{23} (lemon grass oil 1ml)	0.03	0.03	0.03	0.03	0.03	0.03	0.03
G_{24} (cinnamon bark oil 1ml)	0.03	0.03	0.03	0.03	0.03	0.02	0.02
G_{25} (clove oil 1ml)	0.04	0.04	0.04	0.04	0.04	0.04	0.03
G_{26} (cardamom oil 1ml)	0.03	0.03	0.03	0.03	0.03	0.03	0.03
G_{27} (control - sodium benzoate 1ml)	0.04	0.03	0.03	0.03	0.03	0.02	0.02
SEm±	0.001	0.000	0.000	0.000	0.000	0.000	0.000
CD (0.05)	0.001	0.001	N S	N S	N S	N S	N S

*Each value is the mean of five replication

4.3.2.14. Crude fiber

Effect of herbal extracts and essential oils on crude fibre content of liquidized aloe gel juice are presented in table 68.

The crude fibre content was found to be significant among the treatment during the month of storage. Highest value among the treatments were obtained in G_{17} (*Punica granatum* tincture 2 ml) as 0.08 per cent which was significantly different from $G_{16,}$ *Punica granatum* aqueous 1 ml and G_{18} *Punica granatum* decoction 1 ml having a value of 0.06 per cent. The crude fibre content among treatments with essential oils was found to be same as 0.04 per cent. The control G_{27} (control – sodium benzoate) showed a crude fibre content of about 0.04 per cent.

On first month after storage the treatments were significantly different. The highest crude fibre among the treatments were found out in G_{17} (*Punica granatum* tincture 2 ml) with 0.08 per cent of crude fibre content which was significantly different from $G_{16,}$ *Punica granatum* aqueous, 1 ml and G_{18} *Punica granatum* decoction, 1 ml with crude fiber content of 0.06 per cent. The control G_{27} had recorded crude fibre content of 0.03 per cent. The crude fiber content of treatments with essential oils ranged from 0.03 per cent to 0.04 per cent while that for herbal extracts ranged from 0.02 to 0.08 per cent.

The significant difference in crude fiber content was noticed among the treatments during second month after storage. Significantly higher crude fiber content of 0.06 per cent was reported from G_{17} (*Punica granatum* tincture, 2 ml) which was followed by G_2 (*Gymnema sylvestre* tincture 1 ml), $G_{16,}$ *Punica granatum* aqueous 1ml and G_{18}, *Punica granatum* decoction 1 ml with 0.05 per cent. The crude fibre content among the treatments with essential oils was found to vary from 0.03 per cent to 0.04 per cent. Treatments with herbal extracts showed crude fiber content from 0.01 to 0.06 per cent. The control, G_{27} had crude fibre content of 0.03 per cent.

The treatments were found to be significantly different in crude fiber content during third month after storage. Significantly higher crude fibre were noticed in $G_{17,}$

Punica granatum tincture 2ml (0.06 per cent) which was different from G_{18}, *Punica granatum* decoction 1ml (0.05 per cent). The crude fibre content obtained in control G_{27} (control – sodium benzoate) was 0.03 per cent. The crude fiber content of treatments with essential oils varied from 0.03 per cent to 0.04 per cent and that for herbal extracts varied from 0.01- 0.06 per cent.

Significant variation in crude fibre content was noticed among liquidized aloe gel juice with herbal extracts and essential oils during fourth month after storage. Highest crude fibre content of 0.06 per cent was reported from G_{17} (*Punica granatum* tincture, 2 ml) which differed significantly from G_{18}, *Punica granatum* decoction, 1ml with crude fibre content of 0.05 per cent. The crude fiber content among treatments with essential oils was found range from 0.02 per cent to 0.03 per cent. Treatments with herbal extracts showed a variation from 0.01 per cent to 0.06 per cent. The control G_{27} recorded a crude fibre content of 0.02 per cent.

Significant difference in crude fiber content was noticed among the treatments during fifth month after storage. Crude fibre content was the highest in G_{17} (*Punica granatum* tincture 2 ml) and G_{18} (*Punica granatum* decoction 1ml) 0.05 per cent which was significantly different from by G_{14} (*Terminalia chebula* tincture 1 ml), $G_{16,}$ *Punica granatum* aqueous, 1 ml, G_{23} (lemon grass oil 1 ml) and G_{25} (clove oil 1 ml) which recorded 0.03 per cent. The control G_{27} showed crude fibre content of about 0.02 per cent. The crude fibre content of treatments with essential oils ranged from 0.02 to 0.03 per cent while that for herbal extracts varied from 0.01 to 0.05 per cent.

On sixth month after storage the treatments showed significant variation in crude fiber content. The crude fiber content of 0.05 per cent of was noticed in G_{17} (*Punica granatum* tincture 2ml) which was significantly higher than the next best treatment $G_{18,}$ *Punica granatum* decoction 1ml with 0.04 per cent. The control G_{27} showed crude fibre content of about 0.02 per cent. The crude fiber content of treatments with essential oils ranged from 0.02 per cent to 0.03 per cent while that added with herbal extracts ranged from.0.01 to 0.05 per cent.

Chandegara and Varshney (2013) reported that fiber content is directly related to purity of aloe gel and reported crude fiber content as 0.10 percent. In present study the crude fibre decreased on storage showing that it affected the purity of juice. Shubra *et al*. (2014) reported a crude fibre content of 0.26 per cent in aloe gel and study also revealed that blanching effect has negligible effect in crude fibre content.

The fibre content of aloe gel ranged from 0.074 to 0.088 per cent of fresh weight of pulp (Wang and Strong, 1993). The study of Wang and Strong (1993) supported the present study as a range of 0.02 percent to 0.08 per cent, less quantity may be due to the addition of extracts.

Table 68. Effect of herbal extracts and essential oils on crude fiber content of liquidized aloe gel juice, percentage

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.04	0.03	0.02	0.02	0.01	0.01	0.01
G_2 (*Gymnema sylvestre* tincture1ml)	0.04	0.05	0.05	0.03	0.02	0.02	0.02
G_3 (*Gymnema sylvestre* decoction 2ml)	0.03	0.03	0.01	0.01	0.02	0.01	0.01
G_4 (*Centella asiatica* aqueous 1ml)	0.05	0.03	0.01	0.01	0.01	0.01	0.01
G_5 (*Centella asiatica* tincture 2ml)	0.05	0.05	0.03	0.02	0.02	0.02	0.02
G_6 (*Centella asiatica* decoction 1ml)	0.02	0.02	0.01	0.01	0.01	0.01	0.01
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.04	0.03	0.02	0.02	0.01	0.01	0.01
G_8 (*Achyranthes aspera* tincture 2ml)	0.04	0.04	0.04	0.03	0.03	0.02	0.02
G_9 (*Achyranthes aspera* decoction 1.50ml)	0.04	0.03	0.03	0.03	0.02	0.02	0.02
G_{10} (*Tridax procumbens* aqueous 2ml)	0.03	0.02	0.02	0.01	0.01	0.01	0.01
G_{11} (*Tridax procumbens* tincture 2ml)	0.03	0.02	0.02	0.02	0.01	0.01	0.01
G_{12} (*Tridax procumbens* decoction 1ml)	0.02	0.02	0.02	0.02	0.01	0.01	0.01
G_{13} (*Terminalia chebula* aqueous 1ml)	0.05	0.03	0.03	0.03	0.03	0.02	0.01
G_{14} (*Terminalia chebula* tincture 1ml)	0.05	0.04	0.04	0.04	0.03	0.03	0.02
G_{15} (*Terminalia chebula* decoction 1ml)	0.04	0.03	0.03	0.03	0.03	0.02	0.02

Table 68. Effect of herbal extracts and essential oils on crude fiber content of liquidized aloe gel juice, percentage (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.06	0.06	0.05	0.04	0.04	0.03	0.02
G_{17} (*Punica granatum* tincture 2ml)	0.08	0.08	0.06	0.06	0.06	0.05	0.05
G_{18} (*Punica granatum* decoction 1ml)	0.06	0.06	0.05	0.05	0.05	0.05	0.04
G_{19} (green tea aqueous 2ml)	0.03	0.02	0.02	0.01	0.01	0.01	0.01
G_{20} (green tea tincture 1ml)	0.04	0.04	0.03	0.02	0.01	0.01	0.01
G_{21} (green tea decoction 2ml)	0.03	0.03	0.03	0.02	0.02	0.01	0.01
G_{22} (basil oil 1ml)	0.04	0.03	0.03	0.03	0.03	0.02	0.02
G_{23} (lemon grass oil 1ml)	0.04	0.04	0.03	0.03	0.03	0.03	0.02
G_{24} (cinnamon bark oil 1ml)	0.04	0.03	0.03	0.03	0.03	0.02	0.02
G_{25} (clove oil 1ml)	0.04	0.04	0.04	0.04	0.03	0.03	0.03
G_{26} (cardamom oil 1ml)	0.04	0.03	0.03	0.03	0.02	0.02	0.02
G_{27} (control - sodium benzoate 1ml)	0.04	0.03	0.03	0.03	0.02	0.02	0.02
SEm±	0.002	0.002	0.002	0.002	0.002	0.002	0.002
C D (0.05)	0.007	0.006	0.006	0.006	0.006	0.005	0.005

*Each value is the mean of five replication

4.3.2.15. Microbial population

4.3.2.15.1 Mean fungal population

Effect of herbal extracts and essential oils on mean fungal population of liquidized aloe gel juice is presented in table 69.

The mean fungal population were found to be non significant during zero month after storage (0 MAS), the mean fungal population was absent through out the treatments.

During first month after storage the treatments were found to be significantly different. The fungal population increased during first month after storage. The mean fungal population was highest for G_{10} (*Tridax procumbens* aqueous, 2 ml) having a population of 3.66 log cfu ml^{-1} which was on par with all the treatments except G_1 (1.80 log cfu ml^{-1}), G_2 (1.20 log cfu ml^{-1}), G_5 (0.60 log cfu ml^{-1}), G_{11} (0.00 log cfu ml^{-1}), G_{14} (0.00 log cfu ml^{-1}), G_{17} (0.60 log cfu ml^{-1}) and all essential oil treated samples (0.00 log cfu ml^{-1}). The control showed mean fungal population of 3.24 log cfu ml^{-1}.

The mean fungal population of liquidised aloe gel juice during second month after storage also increased significantly. The treatment G_{10} *Tridax procumbens* aqueous 2 ml, (3.75 log cfu ml^{-1}) had the highest fungal population and was on par with G_{21}, green tea decoction, 2 ml (3.73 log cfu ml^{-1}) and G_{13}, *Terminalia chebula*, aqueous, 1ml (3.68 log cfu ml^{-1}). The treatments with essential oils showed no fungal population (0.00 log cfu ml^{-1}). The mean fungal population observed in G_{27} (control) was 3.44 log cfu ml^{-1}.

Significant difference was noticed in mean fungal population during third month after storage. The mean fungal population was highest for G_{21}, green tea decoction, 2 ml added with liquidized aloe gel juice, which had a population of 3.82 log cfu ml^{-1} and was on par with $G_{10,}$ *Tridax procumbens* aqueous, 2 ml (3.78 log cfu ml^{-1}) and G_6, *Centella asiatica* decoction 1 ml (3.76 log cfu ml^{-1}). There was no fungal population in the case of essential oil treated samples. The mean fungal population of control was 3.68 log cfu ml^{-1}.

The mean fungal population during fourth month after storage was significantly different among the treatments. The highest population was noticed in G_{16}, *Punica granatum* aqueous 1ml and G_{17}, *Punica granatum* tincture 2 ml (3.93 log cfu ml^{-1}) which was on par with all the treatments except the samples treated with essential oils. There was no fungal population in clove oil treated sample. The treatments G_{22} (basil oil 1ml), G_{23} (lemon grass oil 1 ml), G_{24} (cinnamon bark oil 1ml) and G_{26} (cardamom oil 1ml) showed mean fungal population of 1.80 log cfu ml^{-1}. Mean fungal population of the control was 3.82 log cfu ml^{-1}.

Significant variations in fungal populations were shown among the treatments during fifth month after storage. The fungal population was noticed highest for G_{16}, *Punica granatum* aqueous, 1ml, showing 4.19 log cfu ml^{-1} which was on par with all the treatments except essential oils treated samples. Absence of population was noticed in G_{25} (clove oil, 1ml). The highest population shown among essential oils in G_{22} (1.98 log cfu ml^{-1}) which was on par with G_{23} (1.86 log cfu ml^{-1}), G_{24} (1.86 log cfu ml^{-1}), and G_{26} (1.86 log cfu ml^{-1}). The treatment G_{27} (sodium benzoate, 1ml) showed a mean fungal population of about 4.17 log cfu ml^{-1}.

The mean fungal population shown significant increase during sixth month after storage. The mean fungal population was noticed highest for G_{16}, *Punica granatum* aqueous, 1ml (4.25 log cfu ml^{-1}) which was on par with all treatments except all oils, G_1, *Gymnema sylvestre* aqueous 1ml (4.12 log cfu ml^{-1}), G_2, *Gymnema sylvestre* tincture 1 ml (4.08 log cfu ml^{-1}), G_3, *Gymnema sylvestre* decoction 2ml (4.13 log cfu ml^{-1}) and G_5, *Centella asiatica* tincture 2 ml (4.13 log cfu ml^{-1}). Absence of population was noticed in G_{25} (clove oil, 1 ml). The mean fungal population of control was 4.24 log cfu ml^{-1}.

4.3.2.15.2 Mean bacterial population

Effect of herbal extracts and essential oils on mean bacterial population of liquidized aloe gel juice is presented in table 70.

The mean bacterial population were found to be non – significant during zero month after storage, the mean bacterial population was absent through out the treatments.

During first month after storage the treatments were found to be significantly different. The mean bacterial population was getting increasing during first month after storage. The highest mean bacterial population was found among the treatments in G_{13} (*Terminalia chebula* aqueous, 1ml) having a population of 6.82 log cfu ml^{-1} which was on par with all the treatments except $G_{8,}$ *Achyranthes aspera* tincture 2ml (4.80 log cfu ml^{-1}), G_{11}, *Tridax procumbens* tincture 2 ml (0.00 log cfu ml^{-1}), G_{14}, *Terminalia chebula* tincture 1ml (2.40 log cfu ml^{-1}) and $G_{17,}$ *Punica granatum* tincture 2ml (4.80 log cfu ml^{-1}) and all essential oil treated samples (0.00 log cfu ml^{-1}). The mean bacterial population observed in control were 6.49 log cfu ml^{-1}.

The mean bacterial population of liquidised aloe gel juice during second month after storage was significantly increasing. The treatment G_{13}, *Terminalia chebula* aqueous 1ml having highest population of 6.88 log cfu ml^{-1} which was on par with G_{21}, green tea decoction, 2ml (6.78 log cfu ml^{-1}). The treatments with essential oils showed no bacterial population (0.00 log cfu ml^{-1}). The mean bacterial population observed in G_{27} (control) was 6.62 log cfu ml^{-1}.

Significant increase was noticed in mean bacterial population during third month after storage. The mean bacterial population was highest for G_{13}, *Terminalia chebula* aqueous 1ml recording a population of 7.01 log cfu ml^{-1} which was on par with $G_{6,}$ *Centella asiatica* decoction 1ml (6.95 log cfu ml^{-1}), $G_{7,}$ *Achyranthes aspera* aqueous, 2 ml (6.95 log cfu ml^{-1}), $G_{15,}$ *Terminalia chebula* decoction,1 ml (6.97 log cfu ml^{-1}) $G_{16,}$ *Punica granatum* aqueous,1 ml (6.99 log cfu ml^{-1}) and G_{17} *Punica granatum* tincture 2 ml (6.95 log cfu ml^{-1}). The zero population was noticed in the case of essential oil treated samples. The control showed a mean bacterial population of 6.72 log cfu ml^{-1}.

The mean bacterial population during fourth month after storage was significantly different among the treatments. The highest population was noticed in G_{16}, *Punica granatum* aqueous 1 ml (7.30 log cfu ml^{-1}) which was on par with all the treatments except the samples treated with essential oils. The clove oil treated sample showed absence of bacterial population. Among the essential oils the highest population was observed in G_{22} (basil oil, 1 ml) with a population of 4.80 log cfu ml^{-1} and was on par with G_{23}, lemon grass oil, 1 ml (3.60 log cfu ml^{-1}) G_{24} , cinnamon bark oil, 1ml (3.60 log cfu ml^{-1}) and $G_{26,}$ cardamom oil 1ml (3.60 log cfu ml^{-1}). Mean bacterial population in the control was 7.27 log cfu ml^{-1}.

Significant variations were shown in an increasing trend among the treatments during fifth month after storage. The mean bacterial population was noticed highest for G_{16}, *Punica granatum* aqueous 1ml and G_{19}, green tea aqueous 2ml with a population of 7.90 log cfu $ml^{-1}$and was on par with all the treatments except essential oil treated samples. Absence of population was noticed in G_{25} (clove oil, 1ml).The highest bacterial population among essential oils used was in $G_{22,}$ basil oil 1ml (5.09 log cfu ml^{-1}) which was on par with $G_{23,}$ lemon grass oil, 1ml (3.89 log cfu ml^{-1}), G_{24} cinnamon bark oil ,1ml (3.72 log cfu ml^{-1}), and $G_{26,}$ cardamom oil 1ml (3.79 log cfu ml^{-1}). The treatment G_{27} (sodium benzoate, 1ml) showed a mean bacterial population of about 7.83 log cfu ml^{-1}.

The mean bacterial population showed a significant increase during six month after storage. The mean bacterial population was highest for G_{13} (*Terminalia chebula* aqueous 1 ml), G_{16}, *Punica granatum* aqueous, 1 ml and G_{19}, green tea aqueous 2 ml (8.37 log cfu ml^{-1}) and was on par with all treatments except all essential oil treated samples. Among essential oil treated samples highest population was noticed in G_{24} (cinnamon bark oil, 1ml) recording a value of 6.71 log cfu ml^{-1} which was on par with G_{22}, basil oil 1ml 6.62 log cfu ml^{-1}, $G_{23,}$ lemon grass oil 1ml 6.62 log cfu ml^{-1}

and G_{26} cardamom oil, 1ml 6.55 log cfu ml^{-1} . The least bacterial population during six month after storage was seen in G_{25}, clove oil, 1ml (2.40 log cfu ml^{-1}). The control recorded a mean bacterial population of 8.31 log cfu ml^{-1}.

Ethanolic extracts of *Aloe vera* showed advanced antibacterial property than aqueous extracts (Choi and Chung, 2003). The result of present study showed that the most of the alcoholic extracts of herbal material added samples had more antimicrobial property than aqueous extracts.

The most active constituents (essential oils) of many spices having wide spectra of antimicrobial activity are aromatic phenolic compounds, such as thymol and carvacrol in oregano and thyme, eugenol in clove and cinnamon and cinnamic aldehyde in cinnamon (Karapinar and Aktug, 1987; Beuchat and Golden, 1989).

Researchers have reported, phenolic compounds from different plant sources could inhibit various foodborne pathogens (Nychas, 1995; Smid and Gorris, 1999; Prashanth *et al*., 2001; Kim *et al*., 2005).

Flavonoids are a major group of phenolic compounds reported for their antiviral (Chiang *et al*., 2003), antimicrobial (Bastos *et al*., 2009) and spasmolytic properties (Amor *et al*., 2005). Many plant flavonoids are reported to (e.g., epigallocatechin, catechin, myricetin, quercetin) have antimicrobial activity (Cushnie and Lamb, 2005).These studies supported the the use of different herbal plant extracts in the study. Bassole and Juliani (2012) repoted that essential oils containing aldehydes or phenols, such as cinnamaldehyde, citral, carvacrol, eugenol or thymol as major components has highest antibacterial activity, followed by essential oils containing terpene alcohols.

According to Kolesnikova *et al*. (1976) essential oils are natural plant products containing complex mixture of components and thus having multiple antimicrobial properties. In basil, the strongest antimicrobial activity of sweet basil was attributed to eugenol (19 per cent) and linalool (54 per cent) content and a synergistic effect was observed. Juliani *et al*. (2004) reported that cinnamaldehyde containing oils (non-phenolic) showed lower antimicrobial activities than eugenol containing oils. The antimicrobial activity of the essential oil of *Cinnamomum zeylanicum* has been related to its cinnamaldehyde content (Prabuseenivasan *et al*., 2006).

According to Wendakoon and Sakaguchi (1995) the antibacterial activity of essential oil of clove may be attributed to eugenol, the antimicrobial compound of clove. The microbial inhibition of eugenol might be related to either membrane disruption or by inactivation of enzymes and genetic material. Sulieman *et al*. (2007) reported that clove oil has a potential antimicrobial activity against *E.coli, Staphylococcus aureus* and *Salmonella lyphimirium*. According to Gulcin *et al*. (2012), the antioxidant activity of clove oil compared with synthetic antioxidants measured as the scavenging of the DPPH radical decreased in the following order: clove oil > BHT > alfa-tocopherol > butylated hydroxyanisole > Trolox. The clove essential oil is widely used for its antimicrobial, antifungal and antioxidant action, these characteristics are strongly attributed to its major compound, eugenol, present in the oil in concentrations that can reach up to 90 per cent (Hongfang, *et al*., 2017, Radunz *et al*., 2019).

Helander *et al*. (1998) reported the antibacterial activity of essential oil of cinnamon may be attributed to cinnamaldehyde the antimicrobial compound of cinnamon and observed the inhibition of *E.coli* 0157:H7 and *Salmonella lyphimirium*. Hoque *et al.* (2007) reported that essential oil of cinnamon at 5 per cent showed the highest antibacterial activity against *Staphylococcus aureus.* Among spices, clove showed the higher content of polyphenols and antioxidant compounds (Rojas *et al*., 2014). These studies about clove oil supports the present study of highest antibacterial properties of clove oil. At a concentration ≤ 2%, lemon grass essential oil inhibits the growth of several kinds of microorganisms including periodontal pathogens, especially the reference strains *Actinomyces naeslundii* and *Porphyromonas gingivalis*, which were resistant to tetracycline hydrochloride. Increased tissue healing response seen in the experimental group can be explained by the antioxidant activity of lemongrass essential oil gel components (Warad *et al*, 2013).

Cymbopogon citratus contains minor component; limonene, which was found to possess an efficient antibacterial property and that may contribute to improve the observed antimicrobial activity (Lin *et al*., 1986). Cardamom (*Eletteria cardamomum*), belonging to the family of *Zingiberaceae* and has been reported to possess several pharmacological properties, including antimicrobial activity (Vaidya and Rathod, 2014).

The above studies about essential oils reported the antimicrobial and antioxidant properties of essential oils. Due to these properties the essential oils of clove, lemon grass, cinnamon, cardamom and basil oil added samples exhibited high antimicrobial properties.

From the gel stabilization studies of liquidized aloe juice with selected dosage of three forms of herbal extracts and essential oils it was concluded that 25 ml of liquidized aloe juice added with 1ml of clove oil possessed maximum storage life and quality during storage studies.

Table 69. Effect of herbal extracts and essential oils on mean fungal population of liquidized aloe gel juice, log cfu ml^{-1}

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.00×10^3 (0.00)	0.60×10^3 (1.80)	1.80×10^3 (3.24)	3.60×10^3 (3.54)	7.00×10^3 (3.85)	11.00×10^3 (4.04)	13.20×10^3 (4.12)
G_2 (*Gymnema sylvestre* tincture1ml)	0.00×10^3 (0.00)	0.40×10^3 (1.20)	2.00×10^3 (3.30)	4.00×10^3 (3.60)	7.60×10^3 (3.88)	11.00×10^3 (4.04)	12.20×10^3 (4.08)
G_3 (*Gymnema sylvestre* decoction 2ml)	0.00×10^3 (0.00)	1.20×10^3 (3.06)	2.2×10^3 (3.34)	4.20×10^3 (3.62)	6.20×10^3 (3.79)	12.00×10^3 (4.08)	13.60×10^3 (4.13)
G_4 (*Centella asiatica* aqueous 1ml)	0.00×10^3 (0.00)	2.60×10^3 (3.40)	3.60×10^3 (3.55)	5.20×10^3 (3.71)	7.20×10^3 (3.86)	13.60×10^3 (4.13)	15.40×10^3 (4.19)
G_5 (*Centella asiatica* tincture 2ml)	0.00×10^3 (0.00)	0.20×10^3 (0.60)	1.20×10^3 (3.06)	3.20×10^3 (3.50)	7.60×10^3 (3.88)	12.00×10^3 (4.08)	13.60×10^3 (4.13)
G_6 (*Centella asiatica* decoction 1ml)	0.00×10^3 (0.00)	3.20×10^3 (3.49)	4.20×10^3 (3.62)	5.80×10^3 (3.76)	7.40×10^3 (3.87)	13.60×10^3 (4.13)	15.20×10^3 (4.18)
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.00×10^3 (0.00)	1.00×10^3 (3.00)	2.00×10^3 (3.30)	4.00×10^3 (3.60)	7.40×10^3 (3.87)	14.00×10^3 (4.15)	17.40×10^3 (4.24)
G_8 (*Achyranthes aspera* tincture 2ml)	0.00×10^3 (0.00)	1.00×10^3 (3.00)	2.00×10^3 (3.30)	4.00×10^3 (3.60)	6.20×10^3 (3.79)	11.60×10^3 (4.06)	14.00×10^3 (4.14)
G_9 (*Achyranthes aspera* decoction 1.50 ml)	0.00×10^3 (0.00)	1.40×10^3 (3.12)	2.40×10^3 (3.37)	4.60×10^3 (3.66)	7.60×10^3 (3.88)	11.80×10^3 (4.07)	14.60×10^3 (4.16)
G_{10} (*Tridax procumbens* aqueous 2ml)	0.00×10^3 (0.00)	4.60×10^3 (3.66)	5.60×10^3 (3.75)	6.00×10^3 (3.78)	7.40×10^3 (3.87)	13.60×10^3 (4.13)	16.20×10^3 (4.21)
G_{11} (*Tridax procumbens* tincture 2ml)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	1.00×10^3 (3.00)	3.00×10^3 (3.48)	7.00×10^3 (3.85)	11.60×10^3 (4.06)	13.80×10^3 (4.14)
G_{12} (*Tridax procumbens* decoction 1ml)	0.00×10^3 (0.00)	1.60×10^3 (3.18)	2.60×10^3 (3.41)	4.60×10^3 (3.66)	8.00×10^3 (3.90)	11.80×10^3 (4.07)	13.80×10^3 (4.14)
G_{13} (*Terminalia chebula* aqueous 1ml)	0.00×10^3 (0.00)	3.80×10^3 (3.57)	4.80×10^3 (3.68)	5.60×10^3 (3.74)	7.40×10^3 (3.87)	15.00×10^3 (4.18)	16.40×10^3 (4.21)
G_{14} (*Terminalia chebula* tincture 1ml)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	1.00×10^3 (3.00)	4.00×10^3 (3.60)	7.80×10^3 (3.89)	13.40×10^3 (4.13)	14.20×10^3 (4.15)
G_{15} (*Terminalia chebula* decoction 1ml)	0.00×10^3 (0.00)	3.40×10^3 (3.52)	4.40×10^3 (3.64)	5.20×10^3 (3.72)	7.40×10^3 (3.87)	15.00×10^3 (4.18)	17.00×10^3 (4.23)

Table 69. Effect of herbal extracts and essential oils on mean fungal population of liquidized aloe gel juice, log cfu ml^{-1} (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.00×10^3 (0.00)	2.40×10^3 (3.37)	3.40×10^3 (3.53)	5.60×10^3 (3.74)	8.60×10^3 (3.93)	15.40×10^3 (4.19)	17.80×10^3 (4.25)
G_{17} (*Punica granatum* tincture 2ml)	0.00×10^3 (0.00)	0.20×10^3 (0.60)	1.20×10^3 (3.06)	3.20×10^3 (3.50)	8.60×10^3 (3.93)	12.40×10^3 (4.09)	14.80×10^3 (4.17)
G_{18} (*Punica granatum* decoction 1ml)	0.00×10^3 (0.00)	2.40×10^3 (3.37)	3.40×10^3 (3.53)	5.20×10^3 (3.71)	7.20×10^3 (3.86)	14.00×10^3 (4.15)	15.60×10^3 (4.19)
G_{19} (green tea aqueous 2ml)	0.00×10^3 (0.00)	1.20×10^3 (3.06)	2.20×10^3 (3.34)	4.80×10^3 (3.67)	7.60×10^3 (3.88)	15.00×10^3 (4.18)	16.80×10^3 (4.23)
G_{20} (green tea tincture 1ml)	0.00×10^3 (0.00)	1.80×10^3 (3.24)	2.80×10^3 (3.44)	4.60×10^3 (3.66)	7.00×10^3 (3.85)	13.00×10^3 (4.11)	14.80×10^3 (4.17)
G_{21} (green tea decoction 2ml)	0.00×10^3 (0.00)	4.40×10^3 (3.64)	5.40×10^3 (3.73)	6.80×10^3 (3.82)	6.80×10^3 (3.83)	14.40×10^3 (4.16)	15.40×10^3 (4.19)
G_{22} (basil oil 1ml)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.60×10^3 (1.80)	1.20×10^3 (1.98)	3.60×10^3 (3.54)
G_{23} (lemon grass oil 1ml)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.60×10^3 (1.80)	0.80×10^3 (1.86)	2.20×10^3 (3.29)
G_{24} (cinnamon bark oil 1ml)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.60×10^3 (1.80)	0.80×10^3 (1.86)	1.60×10^3 (3.18)
G_{25} (clove oil 1ml)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)
G_{26} (cardamom oil 1ml)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.00×10^3 (0.00)	0.60×10^3 (1.80)	0.80×10^3 (1.86)	2.20×10^3 (3.29)
G_{27} (control - sodium benzoate 1ml)	0.00×10^3 (0.00)	1.80×10^3 (3.24)	2.80×10^3 (3.44)	4.80×10^3 (3.68)	6.60×10^3 (3.82)	14.80×10^3 (4.17)	17.20×10^3 (4.24)
SEm (±)	0.000	0.261	0.031	0.026	0.283	0.298	0.039
CD (0.05)	NS	0.723	0.087	0.071	0.785	0.826	0.107

*Figures in parantheses indicate transformed values *Each value is the mean of five replication

Table 70. Effect of herbal extracts and essential oils on mean bacterial population of liquidized aloe gel juice, log cfu ml^{-1}

Herbal extracts/essential oils	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS	5 MAS	6 MAS
G_1 (*Gymnema sylvestre* aqueous 1ml)	0.00 x10^6 (0.00)	2.60 x10^6 (6.41)	3.60 x10^6 (6.55)	6.80 x10^6 (6.83)	11.80 x10^6 (7.07)	72.00 x10^6 (7.86)	173.20 x10^6 (8.23)
G_2 (*Gymnema sylvestre* tincture1ml)	0.00 x10^6 (0.00)	1.20 x10^6 (6.06)	2.20 x10^6 (6.34)	5.40 x10^6 (6.73)	10.40 x10^6 (7.02)	46.20 x10^6 (7.66)	113.80 x10^6 (8.06)
G_3 (*Gymnema sylvestre* decoction 2ml)	0.00 x10^6 (0.00)	3.60 x10^6 (6.55)	4.60 x10^6 (6.66)	7.80 x10^6 (6.89)	12.60 x10^6 (7.10)	63.80 x10^6 (7.81)	133.80 x10^6 (8.13)
G_4 (*Centella asiatica* aqueous 1ml)	0.00 x10^6 (0.00)	4.40 x10^6 (6.64)	5.40 x10^6 (6.73)	7.60 x10^6 (6.88)	13.60 x10^6 (7.13)	75.6 x10^6 (7.88)	196.00 x10^6 (8.29)
G_5 (*Centella asiatica* tincture 2ml)	0.00 x10^6 (0.00)	1.60 x10^6 (6.16)	2.60 x10^6 (6.40)	8.40 x10^6 (6.92)	10.80 x10^6 (7.03)	41.60 x10^6 (7.62)	141.40 x10^6 (8.15)
G_6 (*Centella asiatica* decoction 1ml)	0.00 x10^6 (0.00)	3.40 x10^6 (6.53)	4.40 x10^6 (6.64)	9.00 x10^6 (6.95)	14.60 x10^6 (7.16)	65.20 x10^6 (7.81)	156.60 x10^6 (8.19)
G_7 (*Achyranthes aspera* aqueous 2 ml)	0.00 x10^6 (0.00)	1.40 x10^6 (6.12)	2.40 x10^6 (6.37)	9.00 x10^6 (6.95)	16.20 x10^6 (7.21)	68.60 x10^6 (7.84)	189.00 x10^6 (8.25)
G_8 (*Achyranthes aspera* tincture 2ml)	0.00 x10^6 (0.00)	0.80 x10^6 (4.80)	1.80 x10^6 (6.24)	8.80 x10^6 (6.94)	12.80 x10^6 (7.10)	48.00 x10^6 (7.68)	150.00 x10^6 (8.18)
G_9 (*Achyranthes aspera* decoction 1.50 ml)	0.00 x10^6 (0.00)	1.40 x10^6 (6.12)	2.40 x10^6 (6.37)	6.80 x10^6 (6.83)	16.20 x10^6 (7.21)	54.40 x10^6 (7.73)	214.20 x10^6 (8.32)
G_{10} (*Tridax procumbens* aqueous 2ml)	0.00 x10^6 (0.00)	1.20 x10^6 (6.06)	2.20 x10^6 (6.34)	7.20 x10^6 (6.85)	16.60 x10^6 (7.22)	66.60 x10^6 (7.82)	218.60 x10^6 (8.32)
G_{11} (*Tridax procumbens* tincture 2ml)	0.00 x10^6 (0.00)	0.00 x10^6 (0.00)	1.00 x10^6 (6.00)	7.80 x10^6 (6.89)	14.00 x10^6 (7.15)	43.60 x10^6 (7.64)	144.40 x10^6 (8.16)
G_{12} (*Tridax procumbens* decoction 1ml)	0.00 x10^6 (0.00)	1.40 x10^6 (6.12)	2.40 x10^6 (6.37)	7.80 x10^6 (6.88)	16.00 x10^6 (7.20)	66.40 x10^6 (7.82)	230.00 x10^6 (8.35)
G_{13} (*Terminalia chebula* aqueous 1ml)	0.00 x10^6 (0.00)	6.60 x10^6 (6.82)	7.60 x10^6 (6.88)	10.20 x10^6 (7.01)	17.60 x10^6 (7.25)	77.80 x10^6 (7.89)	238.00 x10^6 (8.37)
G_{14} (*Terminalia chebula* tincture 1ml)	0.00 x10^6 (0.00)	0.40 x10^6 (2.40)	1.40 x10^6 (6.12)	8.20 x10^6 (6.91)	14.60 x10^6 (7.16)	47.80 x10^6 (7.68)	147.80 x10^6 (8.17)
G_{15} (*Terminalia chebula* decoction 1ml)	0.00 x10^6 (0.00)	3.20 x10^6 (6.49)	4.20 x10^6 (6.62)	9.40 x10^6 (6.97)	17.80 x10^6 (7.25)	61.00 x10^6 (7.78)	222.00 x10^6 (8.33)

Table 70. Effect of herbal extracts and essential oils on mean bacterial population of liquidized aloe gel juice, log cfu ml^{-1} (continued)

G_{16} (*Punica granatum* aqueous 1ml)	0.00×10^6 (0.00)	2.00×10^6 (6.28)	3.00×10^6 (6.47)	9.80×10^6 (6.99)	19.80×10^6 (7.30)	78.60×10^6 (7.90)	238.60×10^6 (8.37)
G_{17} (*Punica granatum* tincture 2ml)	0.00×10^6 (0.00)	0.80×10^6 (4.80)	1.80×10^6 (6.24)	9.00×10^6 (6.95)	16.00×10^6 (7.20)	56.40×10^6 (7.75)	156.40×10^6 (8.19)
G_{18} (*Punica granatum* decoction 1ml)	0.00×10^6 (0.00)	1.60×10^6 (6.18)	2.60×10^6 (6.41)	7.80×10^6 (6.88)	16.80×10^6 (7.22)	66.60×10^6 (7.82)	226.40×10^6 (8.34)
G_{19} (green tea aqueous 2ml)	0.00×10^6 (0.00)	3.60×10^6 (6.55)	4.60×10^6 (6.66)	6.40×10^6 (6.80)	18.60×10^6 (7.27)	78.60×10^6 (7.90)	237.80×10^6 (8.37)
G_{20} (green tea tincture 1ml)	0.00×10^6 (0.00)	2.20×10^6 (6.34)	3.20×10^6 (6.50)	8.80×10^6 (6.94)	16.00×10^6 (7.20)	52.00×10^6 (7.71)	163.20×10^6 (8.21)
G_{21} (green tea decoction 2ml)	0.00×10^6 (0.00)	4.60×10^6 (6.66)	6.00×10^6 (6.78)	7.60×10^6 (6.88)	18.40×10^6 (7.27)	68.40×10^6 (7.84)	228.40×10^6 (8.35)
G_{22} (basil oil 1ml)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.80×10^6 (4.80)	2.00×10^6 (5.09)	4.40×10^6 (6.62)
G_{23} (lemon grass oil 1ml)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.60×10^6 (3.60)	1.80×10^6 (3.89)	4.20×10^6 (6.62)
G_{24} (cinnamon bark oil 1ml)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.60×10^6 (3.60)	1.20×10^6 (3.72)	5.20×10^6 (6.71)
G_{25} (clove oil 1ml)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.40×10^6 (2.40)
G_{26} (cardamom oil 1ml)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.00×10^6 (0.00)	0.60×10^6 (3.60)	1.40×10^6 (3.79)	3.60×10^6 (6.55)
G_{27} (control - sodium benzoate 1ml)	0.00×10^6 (0.00)	3.20×10^6 (6.49)	4.20×10^6 (6.62)	5.20×10^6 (6.72)	18.40×10^6 (7.27)	68.20×10^6 (7.83)	208.00×10^6 (8.31)
SE_m (±)	0.000	0.435	0.036	0.021	0.542	0.573	0.286
CD (0.05)	0.000	1.204	0.101	0.058	1.502	1.588	0.792

* Figures in parantheses indicate transformed values *Each value is the mean of five replication

4.4 PREPARATION OF NUTRACEUTICALS

The stabilized liquidized aloe gel juice mixed with clove oil was used for the preparation of nutraceuticals such as aloe herbal health drink and aloe herbal powder.

4.4.1. Aloe – herbal health drink

Aloe herbal health drink were prepared by adding lemon juice, orange juice and honey to the stabilized liquidized aloe gel juice in different proportions *viz.*, 50:50, 25:75 and 10:90 followed by pasteurization and flash cooling and were stored in 30 ml glass bottles. Observations on sensory/organoleptic parameters, chemical/nutrient composition and shelf life were recorded on these nutraceuticals prepared. The nutraceuticals prepared during the study are presented in plate 13.

4.4.1.1. Sensory/Organoleptic parameters

4.4.1.1.1 Appearance

The appearance of stabilized aloe juice with different proportions of lemon juice, orange juice and honey is presented in table 71.

The appearance of aloe herbal health drink stabilized with clove oil was significantly different during the month of storage. The highest mean rank for appearance among the treatments was scored by T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) which showed a mean rank value of 162.48 which was followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with an appearance of about 141.11.The least rank value was shown by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with an appearance of 77.30 which was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) with 80.32.

During first month after storage the mean rank value for appearance was significantly higher for T_1 - Lemon juice + Liquidized aloe gel juice (50: 50) with a mean rank value of 161.12 which was followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with a mean rank value of about 146.74. The lowest mean rank value among the treatments was noticed in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with mean rank value of 76.40 which was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) scoring a mean rank value of 78.34.

The treatments significantly varied during second month after storage. The treatments which shown highest mean rank value was T_1- Lemon juice + Liquidized aloe gel juice (50 : 50) with 160.34 which was followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with a mean rank value of about 146.24.The least rank value among the treatments was noticed in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with a mean rank value of 78.04 and was on par with T_5- Orange juice + Liquidized aloe gel juice (25 : 75) recording 80.08.

During third month after storage the treatments showed significant variation among themselves. Highest mean rank value among the treatments was noticed in T_1- Lemon juice + Liquidized aloe gel juice (50 : 50) with a value of 157.54 which was followed by T_2 - Lemon juice + Liquidized aloe gel juice (25: 75) with a mean rank value of about 142.76. Among the treatments the least mean rank value was noticed in T_6- Orange juice + Liquidized aloe gel juice (10 : 90) with a mean rank value of 79.34 and was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) with 83.64.

The treatments showed significant difference among themselves during fourth month after storage. The highest mean rank value among the treatments was shown by T_1 - Lemon juice + Liquidized aloe gel juice (50 :50) which was having a value of 158.30 followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with a mean rank value of appearance as 139.46. Among the treatments the least rank value was noticed in T_6- Orange juice + Liquidized aloe gel juice (10: 90) with a mean rank value of 80.56 which was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) with 86.66 mean rank value.

4.4.1.1.2 Colour

Colour of aloe herbal health drink mixed with clove oil is presented in table 72.

The colour of treatments of aloe herbal health drink with clove oil was significantly different during the month of storage. The highest value among the treatments was shown by T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) which had a mean rank value of 154.62 which was followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with colour mean rank value of about 134.70. The least value was shown by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with mean rank value of 81.30 which was on par with T_9 - Honey + Liquidized aloe gel juice (10 : 90) with 90.48 mean rank value.

During first month after storage the mean rank value for colour was significantly higher for T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) record ing 153.92 which was followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with 134.24. The lowest mean rank value among the treatments was noticed in T_6- Orange juice + Liquidized aloe gel juice (10 : 90) with mean rank value of about 81.12 which was followed by T_9 - Honey + Liquidized aloe gel juice (10 : 90) with 93.88.

There was significant variation among treatments during second month after storage. The treatments which showed highest mean rank value was 153.18 for T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) which was followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with a mean rank value of about 133.74. The least mean rank value among the treatments (80.80) was noticed in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90). The mean rank value for T_9 - Honey + Liquidized aloe gel juice (10 : 90) was 93.78.

Treatments during third month after storage showed significant variation in mean rank value for colour. Highest mean rank value among the treatments was noticed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) with a value of

150.72 followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) which recorded a mean rank value of 135.34 for colour. Among the treatments the least mean rank value was noticed in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) recording a score of 84.14 which was on par with T_9 - Honey + Liquidized aloe gel juice (10 : 90) recording a mean rank value of 93.44.

The treatments showed significant difference in mean rank value for colour during fourth month after storage. The highest value among the treatments were shown by T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) which had a mean rank value of 151.20 followed by T_2 -Lemon juice + Liquidized aloe gel juice (25 : 75) with an appearance of about 136.34. The least value among the treatments was noticed in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with a mean rank value for colour as 85.88 and was on par with T_9 – Honey + Liquidized aloe gel juice (10 : 90) scoring a mean rank value of 95.02.

4.4.1.1.3 Taste

The taste of stabilized aloe juice with different proportions of lemon juice, orange juice and honey is presented in table 73.

The taste of treatments of aloe herbal health drink with clove oil was significantly different during zero month after storage. The highest mean rank value for taste was shown by T_7 - Honey + Liquidized aloe gel juice (50 : 50) with a rank value of 186.76 followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with a mean taste rank value of about 161.72. The least mean rank value was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with value of 76.96 and was on par with T_1 - Lemon juice (50ml) + Liquidized aloe gel juice (50 : 50), (85.44), T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) both recording a mean rank value of 85.44 and T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with a mean rank value of 87.56.

During first month after storage the treatments were significantly higher for T_7 – Honey + Liquidized aloe gel juice (50:50) with a mean rank value of 185.42 followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with a mean rank value of 160.34 for taste. The lowest mean rank value (78.10) among the treatments was noticed in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) which was on par with T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) recording a mean rank value of 88.42, T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with a mean rank value of 86.10 and T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with a mean rank value of 88.10.

The treatments significantly varied during second month after storage. The treatments which showed highest mean rank value of 185.42 was in T_7 - Honey + Liquidized aloe gel juice (50 : 50) which was followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with a value of 160.38. The least value among the treatments was noticed in T_3 - Lemon juice + Liquidized aloe gel juice (10:90) with a taste value of 78.00 which was on par with T_1 - Lemon juice + Liquidized aloe gel juice (50:50) with a mean rank value of 86.94, T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with a mean rank value of 85.84 and T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with a mean rank value of 88.66.

During third month after storage the mean rank value obtained for taste showed significant variation among them. Highest mean rank value for taste was noticed in T_7 - Honey + Liquidized aloe gel juice (50 : 50) which was having a value of 182.54 followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with a mean rank value of about 158.16. Among the treatments the least rank value was noticed in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)with a mean rank value of 80.98 which was on par with T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) with a mean rank value of 86.08, T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75), with a mean rank value of 87.20, T_5 - Orange juice + Liquidized aloe gel juice (25 :

75) with a mean rank value of 91.28 and T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with a mean rank value of 91.80.

The treatments showed significant difference among themselves during fourth month after storage. The highest mean rank value (179.32) among the treatments was shown in T_7 - Honey + Liquidized aloe gel juice (50 : 50) followed by T_8 – Honey + Liquidized aloe gel juice (25 : 75) with a mean rank value of about 155.92. Among the treatments the least mean rank value was noticed in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with a mean rank value of 83.78 and was on par with T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) which had a mean rank value of 87.50, T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) which had a mean rank value of 86.50, T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) with a mean rank value of 94.78 and T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with a mean rank value of 90.64.

4.4.1.1.4 Odour

The odour of stabilized aloe juice with different proportions of lemon juice, orange juice and honey is presented in table 74.

The odour of stabilized aloe herbal health drink with different proportions of lemon juice, orange juice and honey showed significant difference among the treatments during zero month after storage. The highest mean rank value among the treatments were observed in T_7 - Honey + Liquidized aloe gel juice (50 : 50) recorded a value of 151.46 which was on par with T_8 – Honey + Liquidized aloe gel juice (25 : 75) with a mean rank value of 145.50. The least value was shown by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with value of 58.32 which was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) recording a mean rank value of 68.78.

During first month after storage the mean rank odour scored was highest for T_7 - Honey + Liquidized aloe gel juice (50 : 50) with an odour value of 151.56 which was followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) having a value of about 144.92. Among the treatments, lowest mean rank value was noticed in T_6 - Orange juice + Liquidized aloe gel juice (10:90), scoring 57.98 which was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) with a mean rank value of 68.60.

The treatments significantly varied during second month after storage. The treatments which showed highest mean rank value of 151.14 was from T_7 - Honey + Liquidized aloe gel juice (50 : 50) which was on par with T_8 – Honey + Liquidized aloe gel juice (25 : 75) with a value of 144.44. The least mean rank value among the treatments were noticed in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with a mean rank value of 59.34 which was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) scoring 67.92.

During third month after storage the treatments showed significantly higher mean rank value for T_7 – Honey + Liquidized aloe gel juice (50 : 50) which was having a value of 152.12 but was on par with T_8 – Honey + Liquidized aloe gel juice (25 : 75) with value of about 145.62. Among the treatments the least mean rank value of 60.14 was noticed in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) which was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) with a mean rank value of 71.24.

Significant difference was observed among the treatments during fourth month after storage also. The highest mean rank value for odour was noticed in T_7 - Honey + Liquidized aloe gel juice (50 : 50) recording 149.14 which was on par with T_8 - Honey + Liquidized aloe gel juice (25 : 75) with having a mean rank value of 142.88. The lowest mean rank value of 63.26 among the treatments was observed in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and this was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) with a mean rank value of 73.64.

4.4.1.1.5 Overall acceptability

The overall acceptability of stabilized aloe juice with different proportions of lemon juice, orange juice and honey is presented in table 75.

The mean rank value for overall acceptability of different types of aloe health drink showed significant difference among the treatments during zero month after storage. The highest overall acceptability among the treatments was recorded from T_7 - Honey + Liquidized aloe gel juice (50 : 50) with a mean rank value of 185.12 followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with overall acceptability of about 164.12. The least mean rank value was shown by T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) which was on par with T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with 59.10 and 60.56 respectively.

During first month after storage, the different treatments of aloe herbal health drink showed significantly higher value for T_7 – Honey + Liquidized aloe gel juice (50 : 50) with an acceptability value of 185.88 followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with a value of about 163.72. Among the treatments, lowest mean rank value of 58.28 was observed in T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) and was on par with T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) recording a mean rank value of 59.62.

The mean rank value for overall acceptability showed significant variation during second month after storage. Among the treatments highest mean rank value was recorded in T_7 – Honey + Liquidized aloe gel juice (50 : 50) having overall acceptability of 186.68 which was followed by T_8 -Honey + Liquidized aloe gel juice (25 : 75) with a value of 164.76. The least value among the treatments was noticed in T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) with a mean rank score of 57.04 and was on par with T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) having a mean rank value of 58.38.

Significantly higher mean rank value during third month of storage was noticed in T_7 - Honey + Liquidized aloe gel juice (50 : 50) with a mean rank value of 180.64 followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with a value of about 159.12. Among the treatments the lowest mean rank value was noticed in T_6 Orange juice + Liquidized aloe gel juice (10 : 90) recording 61.16 which was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) recording a mean rank value of 61.22.

In fourth month after storage the treatments showed significantly high variation in the mean rank value of overall acceptability. The highest mean rank value was observed during the period in T_7 - Honey + Liquidized aloe gel juice (50 : 50) with acceptability value of 177.40 which was followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) having a value of 156.84. The least mean rank value for overall acceptability among the treatments was noticed in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and was on par with T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) showing 62.66 and 65.78 respectively.

In the study the mean rank value for the liquidized aloe juice with clove oil blended with honey, orange juice and lemon juice showed less variation during four month of storage. These findings were supported by the following studies of different juices blended with aloe juice. Orange – *Aloe vera* blended RTS revealed that the treatments were found to be suitable at 45 days of storage by the analysis of sensory attributes (Kausar *et al*., 2020).

Boghani *et al*. (2012) made efforts to develop blended RTS beverage using papaya and aloe juice and found that papaya and aloe juice blended juice could be stored for period of three months without change in chemical and organoleptic qualities also reported that appearance of the blended papaya – *Aloe vera* RTS beverage enhanced with increase in the concentration of *Aloe vera* juice up to a level of 10 percent, further increase in *Aloe vera* juice content reduced the appearance profile. Sasikumar (2015) reported the highest score and acceptability for aloe gel incorporated fruit beverage. Honey, *Aloe vera* pulp and soya milk were blended in nine different proportions and evaluated for sensory attributes by Nath *et al*. (2015). Out of the nine combinations, three combinations were adjudged to be the best based on the rank means obtained for sensory qualities with a combination of 2.5:1.0:2.5:4.0. with honey, *Aloe vera* pulp, soya milk and water. The best combination, was the one having honey and soya milk in equal amount with 10 per cent *Aloe vera* pulp. The study of *Aloe vera* fortified beverage with lemon oil revealed the combined juice have better sensory and nutritional characters than when used individually and most recommended formulation is 90 percent lemon and 10 percent *Aloe vera* (Shailaja *et al*., 2018).

Rahman *et al*. (2015) recorded sensory attributes of the fixed fruit jam made from *Aloe vera*, pineapple and mango and Tiwari and Deen (2015) recorded the observation on preparation and storage of blended RTS beverage from bael and *Aloe vera*. Amongst all the blends of *Aloe vera* juice blended with pineapple beverage, blend T_1 with pineapple - *Aloe vera* juice (90:10) was found to be the most acceptable and the organoleptic parameters like color, flavor and taste of the juice were acceptable up to 14 days of storage (Biswas *et al*., 2016). In the present study less variation in sensory parameters upto fourth month after storage was recorded and acceptability was found to be highest for Honey: Aloe juice (50:50) due to less polysaccharide degradation. Sensory parameters like appearance, colour, taste, odour and over all acceptability of aloe herbal health drink presented in figure 8, figure 9, figure 10, figure 11 and figure 12 respectively.

Plate 13 A. Stabilized aloe gel juice + honey (50 : 50)

Plate 13 B. Stabilized aloe gel juice oil + honey (90 : 10)

Plate 13C. Stabilized aloe gel juice + orange juice (50 : 50)

Plate 13 D. Stabilized aloe gel juice + orange juice (90 : 10)

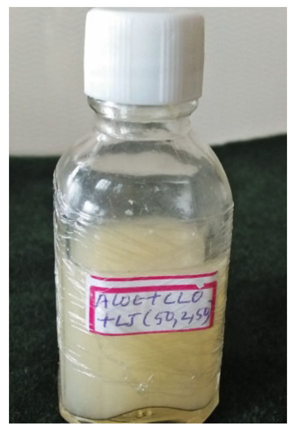


Plate 13 E. Stabilized aloe gel juice + lemon juice (50 : 50)

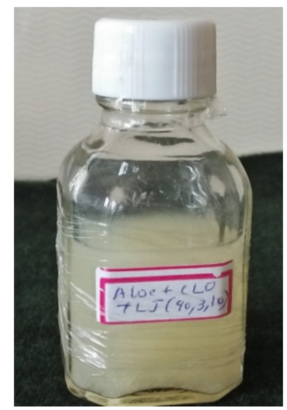


Plate 13 F. Stabilized aloe gel juice + lemon juice (90 : 10)

Plate 13. Stabilized *Aloe vera* herbal health drink

Table 71. Appearance of aloe herbal health drink during storage

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	6.86 (162.48)	6.92 (161.12)	6.88 (160.34)	6.79 (157.54)	6.79 (158.30)
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	6.71 (141.11)	6.75 (146.74)	6.71 (146.24)	6.63 (142.76)	6.63 (139.46)
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	5.79 (126.70)	6.42 (125.12)	6.38 (124.90)	6.29 (124.72)	6.29 (124.34)
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	5.83 (102.98)	6.08 (103.62)	6.04 (102.90)	5.96 (103.34)	5.96 (103.64)
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	6.08 (80.32)	5.75 (78.34)	5.71 (80.08)	5.63 (83.64)	5.63 (86.66)
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	6.42 (77.30)	5.79 (76.40)	5.75 (78.04)	5.67 (79.34)	5.67 (80.56)
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	6.38 (123.00)	6.38 (121.50)	6.33 (121.28)	6.25 (121.22)	6.25 (120.98)
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	6.08 (111.14)	6.21 (109.78)	6.17 (109.26)	6.08 (109.62)	6.08 (107.44)
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	6.21 (95.36)	6.04 (94.38)	6 (93.96)	5.92 (94.82)	5.92 (95.62)
KW value	43.20	42.93	43.75	37.31	34.04
χ^2	15.507				
CD value	11.400				

*Figures in parenthesis represent mean rank value

Table 72. Colour of aloe herbal health drink during storage

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	8.2 (154.62)	8.17 (153.92)	8.12 (153.18)	8.04 (150.72)	8.04 (151.20)
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	7.96 (134.70)	7.96 (134.24)	7.92 (133.74)	7.83 (135.34)	7.83 (136.34)
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	7.2 (123.12)	7.75 (120.64)	7.71 (120.76)	7.63 (120.04)	7.63 (115.82)
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	7.48 (107.82)	7.54 (108.00)	7.5 (108.10)	7.42 (107.64)	7.42 (108.84)
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	7.56 (102.30)	7.46 (102.32)	7.42 (102.24)	7.33 (101.86)	7.33 (103.18)
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	7.8 (81.30)	7.17 (81.12)	7.12 (80.80)	7.04 (84.14)	7.04 (85.88)
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	7.64 (112.14)	7.67 (112.08)	7.63 (113.36)	7.54 (113.00)	7.54 (108.86)
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	7.32 (110.52)	7.62 (110.80)	7.58 (111.04)	7.5 (110.82)	7.5 (111.86)
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	7.68 (90.48)	7.29 (93.88)	7.25 (93.78)	7.17 (93.44)	7.17 (95.02)
KW value	27.33	25.45	25.10	22.50	21.45
χ^2	15.507				
CD value	11.400				

*Figures in parenthesis represent mean rank value

Table 73. Taste of aloe herbal health drink during storage

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	6.12 (85.44)	5.96 (88.42)	5.92 (86.94)	5.83 (86.08)	5.83 (87.50)
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	5.88 (85.44)	6 (86.10)	5.96 (85.84)	5.88 (87.20)	5.88 (86.50)
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	6.08 (76.96)	5.88 (78.10)	5.83 (78.00)	5.75 (80.98)	5.75 (83.78)
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	6.04 (119.68)	6.54 (119.86)	6.54 (120.14)	6.46 (121.10)	6.46 (119.78)
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	6.04 (91.16)	6.08 (91.96)	6.08 (92.58)	6 (91.28)	6 (94.78)
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	6.56 (87.56)	6.04 (88.10)	6.04 (88.66)	5.96 (91.80)	5.96 (90.64)
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	6.56 (186.76)	7.54 (185.42)	7.54 (185.42)	7.46 (182.54)	7.46 (179.32)
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	7.56 (161.72)	7.13 (160.34)	7.12 (160.38)	7.04 (158.16)	7.04 (155.92)
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	7.16 (119.68)	6.5 (118.70)	6.5 (119.04)	6.42 (117.86)	6.42 (118.78)
KW value	76.70	73.29	73.53	66.98	60.27
χ^2	15.507				
CD value	11.400				

*Figures in parenthesis represent mean rank value

Table 74. Odour of aloe herbal health drink during storage

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 M AS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	7.4 (138.22)	7.38 (137.76)	7.33 (137.32)	7.2 (133.52)	7.25 (133.58)
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	7.16 (117.70)	7.13 (117.32)	7.08 (116.80)	7 (113.60)	7 (114.26)
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	7.16 (116.38)	7.13 (116.28)	7.08 (115.96)	7 (115.26)	7 (115.68)
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	6.76 (86.72)	6.71 (88.92)	6.67 (90.74)	6.58 (90.68)	6.58 (92.42)
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	6.48 (68.78)	6.46 (68.60)	6.42 (67.92)	6.33 (71.24)	6.33 (73.64)
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	6.32 (58.32)	6.25 (57.98)	6.21 (59.34)	6.13 (60.14)	6.13 (63.26)
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	7.56 (151.46)	7.54 (151.56)	7.5 (151.14)	7.42 (152.12)	7.42 (149.14)
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	7.48 (145.50)	7.46 (144.92)	7.42 (144.44)	7.33 (145.62)	7.33 (142.88)
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	7.36 (133.92)	7.33 (133.66)	7.29 (133.34)	7.21 (134.82)	7.21 (132.14)
KW value	65.28	64.56	63.57	60.11	51.70
χ^2	15.507				
CD value	11.400				

*Figures in parenthesis represent mean rank value

Table 75. Over all acceptability of aloe herbal health drink during storage

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 M AS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	6.16 (114.54)	6.96 (114.20)	6.92 (113.56)	6.83 (114.46)	6.83 (114.22)
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	6.88 (104.42)	6.83 (104.16)	6.79 (103.50)	6.71 (104.54)	6.71 (105.10)
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	6.76 (95.84)	6.71 (96.98)	6.67 (97.66)	6.58 (99.10)	6.58 (99.58)
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	6.08 (86.16)	6.54 (85.70)	6.5 (87.40)	6.42 (88.90)	6.42 (89.54)
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	7 (59.10)	6.13 (58.28)	6.08 (57.04)	6.04 (61.22)	6.04 (65.78)
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	7.52 (60.56)	6 (59.62)	5.96 (58.38)	5.88 (61.16)	5.88 (62.66)
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	8.2 (185.12)	8.17 (185.88)	8.13 (186.68)	8 (180.64)	8 (177.40)
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	7.8 (164.12)	7.79 (163.72)	7.75 (164.76)	7.62 (159.12)	7.63 (156.84)
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	6.56 (147.14)	7.5 (148.46)	7.46 (148.02)	7.38 (147.86)	7.38 (145.88)
Kw value	102.10	104.40	107.03	91.52	80.87
χ^2	15.507				
CD value	11.400				

*Figures in parenthesis represent mean rank value

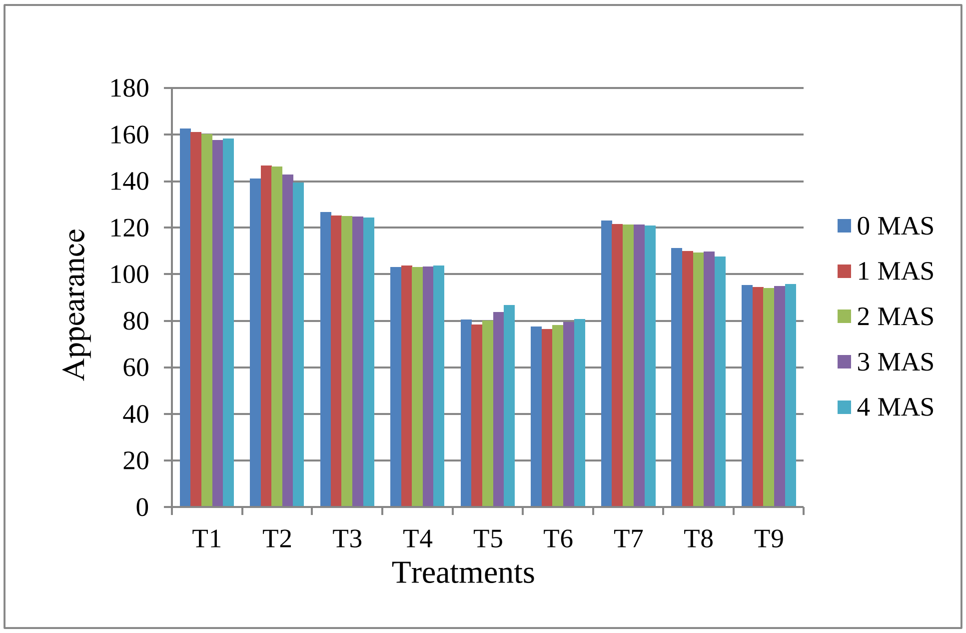


Figure 8. Appearance of aloe herbal health drink with during storage

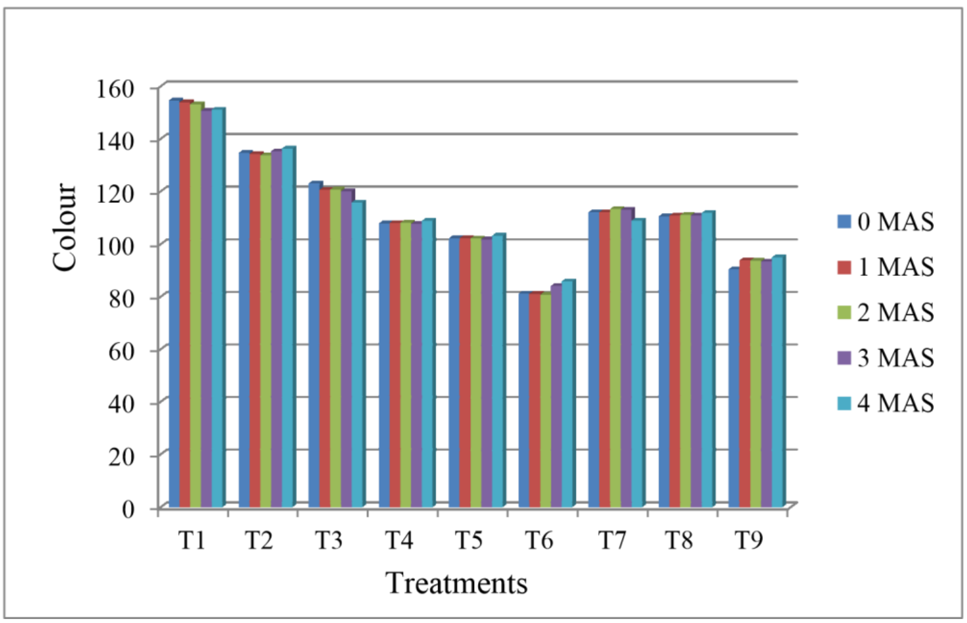


Figure 9. Colour of aloe herbal health drink with during storage

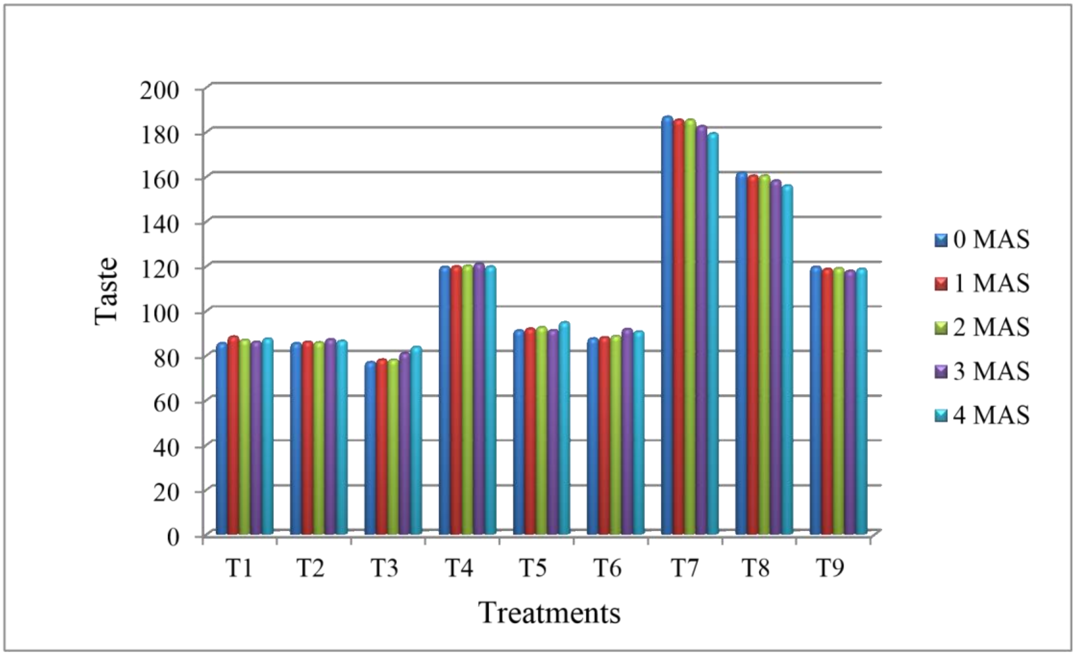


Figure 10. Taste of aloe herbal health drink with during storage

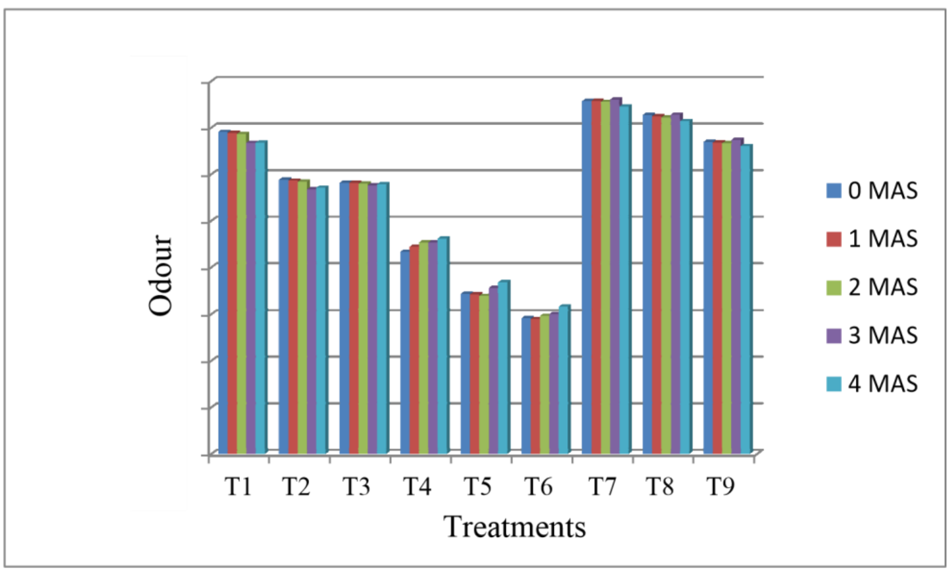


Figure 11. Odour of aloe herbal health drink during storage

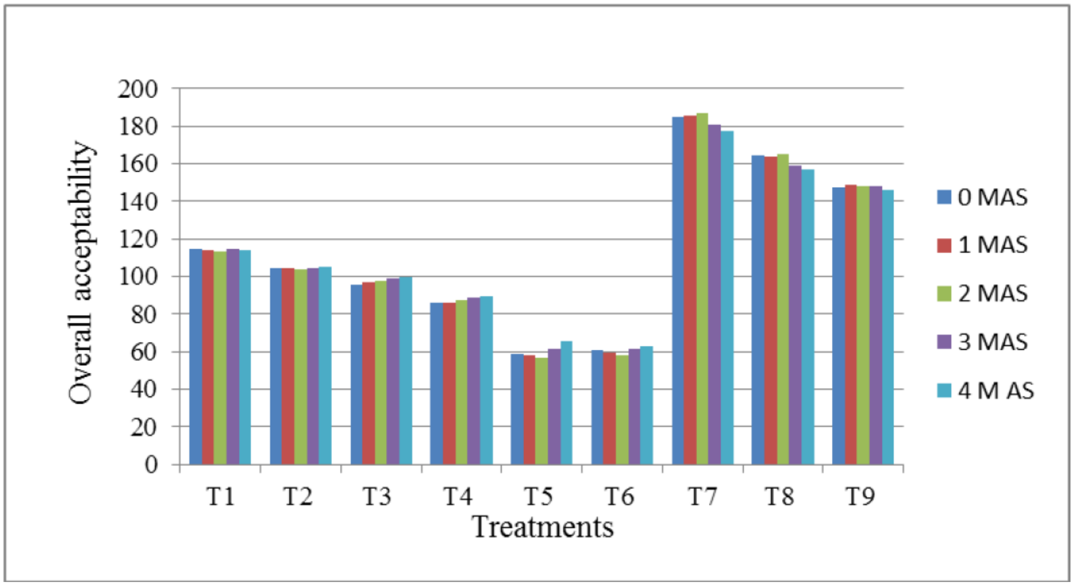


Figure 12. Over all acceptability of aloe herbal health drink during storage

4.4.1.2. Chemical /nutrient composition

4.4.1.2.1 Acidity

Acidity of stabilized Liquidized aloe gel juice with lemon juice, orange juice and honey on varying proportions is presented in table 76. The acidity value increased during first month after storage then decreased during second month after storage and once again increased during third month and fourth month after storage.

The acidity was found to be significant among treatments during zero month after preparation. The highest value among the treatments was noticed inT_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) with an acidity of 0.65meq/ml followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) having an acidity of about 0.37 meq/ml. Acidity was found to be lowest in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90), recording 0.04 meq/ml while that of T_9 - Honey + Liquidized aloe gel juice (10 : 90) was 0.05 meq/ml.

On first month after storage there was significant difference in acidity among various treatments. The highest acidity content of 0.66 meq/ml was recorded in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50), followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) having an acidity of about 0.35 meq/ml. Lowest acidity of 0.03 meq/ml was recorded from T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and acidity of 0.05 meq/ml was observed from T_9 -Honey + Liquidized aloe gel juice (10 : 90). Acidity was found to be same for T_9 during zero month and first month after preparation.

During second month after preparation significantly higher value of 0.65 meq/ml was noticed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) which was followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) having an acidity of about 0.38 meq/ml. Lowest acidity content among the health drinks was observed in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with an acidity of 0.04 meq/ml followed by T_9 - Honey + Liquidized aloe gel juice (10 : 90) showing an acidity of 0.06 meq/ml. The acidity value ranged from 0.65 meq/ml to 0.04 meq/ml during second month after storage among the various treatments.

Significant variation was noticed among the treatments during third month after storage and it showed an increasing trend in acidity. Highest acidity among the treatments was noticed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) which recorded a value of about 0.67 meq/ml followed by T_2- Lemon juice + Liquidized aloe gel juice (25 : 75) with an acidity of about 0.39 meq/ml. The least value was shown by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and T_9 - Honey + Liquidized aloe gel juice (10 : 90) with an acidity of 0.06 meq/ml followed by T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) with 0.08 meq/ml.

There was significant difference in acidity among various treatments of aloe herbal health drink on fourth month of storage. The highest acidity content was noticed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) with 0.68 meq/ml followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with an acidity of about 0.42 meq/ml. Acidity content was lowest for T_6 -Orange juice + Liquidized aloe gel juice (10 : 90) and T_9 - Honey + Liquidized aloe gel juice (10: 90) both recording 0.07 meq/ml which was followed by T_5- Orange juice + Liquidized aloe gel juice (25 : 75) with 0.10 meq/ml and T_7 - Honey + Liquidized aloe gel juice (50 : 50) with 0.10 meq/ml acidity. Acidity content of the neutraceuticals during fourth month after preparation thus ranged from 0.07 meq/ml to 0.68 meq/ml.

The pH of the beverage was found to decrease with increase in storage and could be correlated inversely with the acidity of product reported by Pawar *et al*. (2011), the acidity of the neutraceutical in the present study also supports the same. Singh *et al*. (2012) reported that during manufacturing of lassi, containing aloe juice titrable acidity increased with increasing the level of *Aloe vera* juice from 0 to 15 per cent. In the present study 50 per cent aloe juice with 50 per cent lemon juice reported highest acidity which might be due to the added acidity of lemon juice.

Increasing trend in acidity due to the formation of various organic acids in the fruits such as sulphurous acid was reported by Baramanary *et al*. (1995). Sandhu *et al*. (2001) observed increasing trends in acidity with increasing storage period. According to Rodrigo *et al.,* (2003) increase in acidity indicated the start of spoilage or fermentation of the sample. Singh *et al*. (2014) observed acidity of *bael* RTS decrease during storage period of 6 months. Kumar *et al*. (2013) and Sasikumar (2015) conducted study about acidity of aloe juice and reported acidity of aloe juice as 1.2 percent.

Elbandy *et al*. (2014) reported an acidity of 0.10 percent and Talib *et al*. (2016) reported an acidity of 1.15 percent in aloe gel.The variation in acidity of present study may be due to blending of aloe juice, orange juice, lemon juice and honey with different proportions.

In probiotic honey beverage formulated with sterilized ingredients, honey, soya milk and 1 per cent *Lactobacillus acidophilus* with 6 h of incubation acidity increased considerably from 0.07 per cent - 0.32 per cent (Nath *et al*., 2015). Variation in acidity of aloe herbal health drink were presented in figure 13.

Table 76. Acidity of aloe herbal health drink during storage, meq/ml

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	0.65	0.66	0.65	0.67	0.68
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	0.37	0.35	0.38	0.39	0.42
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	0.09	0.08	0.10	0.12	0.15
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	0.09	0.09	0.10	0.11	0.13
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	0.06	0.06	0.07	0.08	0.10
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	0.04	0.03	0.04	0.06	0.07
T_7- Honey + Liquidized aloe gel juice (50 : 50)	0.09	0.08	0.09	0.09	0.10
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	0.08	0.08	0.09	0.10	0.11
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	0.05	0.05	0.06	0.06	0.07
SEm (±)	0.002	0.002	0.002	0.002	0.002
CD (0.05)	0.005	0.005	0.005	0.005	0.006

*Each value is the mean of six replication

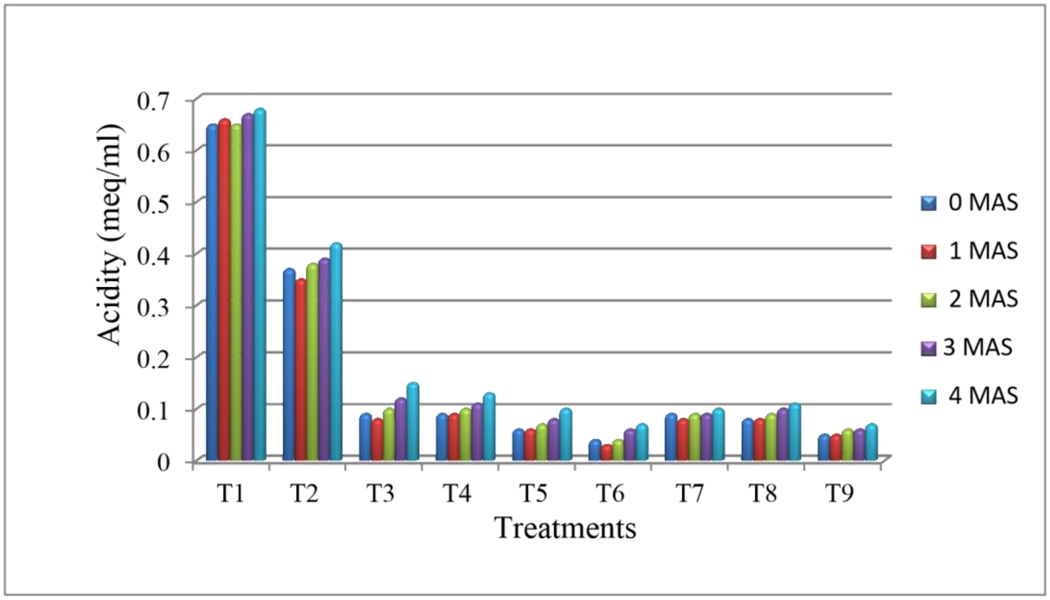


Figure 13. Acidity of aloe herbal health drink during storage

4.4.1.2.2 pH

Effect of different combinations of lemon juice, orange juice and honey with stabilized liquidized aloe gel juice on pH is presented in table 77.

The pH was found to be significant among the treatment during zero month after storage. The highest pH among the treatments was noticed in T_7 - Honey + Liquidized aloe gel juice (50 : 50) with 4.10 pH followed by treatment T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with 3.99 pH. Lowest pH value was noticed for T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) recording 2.21 which was followed by T_2- Lemon juice + Liquidized aloe gel juice (25 : 75) having a pH of about 2.51. The pH ranged from 2.21 to 4.10 during zero month after storage.

On first month after storage the treatments showed significant difference in pH. Treatment T_7 - Honey + Liquidized aloe gel juice (50 : 50) recorded a higher pH of 4.10 followed by treatment T_8 - Honey + Liquidized aloe gel juice (25 : 75) and T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) both with a pH of 3.99. The treatment T_7 and T_4 showed same pH for zero month after n and first month after storage. The least value among the treatments was shown by T_1 - Lemon juice + Liquidized aloe gel juice (50: 50) with 2.2 and treatment T_2 - Lemon juice + Liquidized aloe gel juice (25: 75) had a pH of about 2.53.

Significant variation was noticed among the treatments during second month after storage. Highest pH was noticed in treatment T_7 - Honey + Liquidized aloe gel juice (50 : 50) which had a pH of 4.09 followed by T_8 – Honey + Liquidized aloe gel juice (25 : 75) with 3.98 pH and T_4 Orange juice + Liquidized aloe gel juice (50 : 50) with 3.98pH. Among the treatments, least value was shown by T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) which recorded a pH of 2.21.Treatment T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) had a pH of about 2.51 at 2 MAS. The pH ranged from 2.21 to 4.09 among the various combinations during second month after storage.

Significant change was noticed among the treatments during third month after storageand pH decreased during the month. Among the various combinations, highest pH was noticed in T_7 - Honey + Liquidized aloe gel juice (50 : 50) which recorded a pH of 4.08 and this was followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with pH of 3.97 and T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with a pH of 3.97. The lowest pH among the treatments was observed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) with a pH of 2.20. Treatment, T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) had a pH of about 2.50.

pH of stabilized liquidized aloe gel juice added with different combinations of lemon juice, orange juice and honey during fourth month after storage varied significantly. The highest pH was noticed in T_7 - Honey + Liquidized aloe gel juice (50 : 50) with a recorded pH of 4.08 followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) and T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) both with a recorded pH of 3.95. Least value among the treatments was noticed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) which has a pH of 2.18 while treatment T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) recorded a pH of about 2.47. The pH of various combinations ranged from 2.18 to 4.08.

Adubofuor *et al*. (2010) reported that the pH of the studied samples ranged from 3.83 – 3.90 which was within the expected range of 3 - 5 for fruits and vegetable juices. According to the present study pH and acidity are correlated inversely, which was also supported by the study of Pawar *et al*. (2011) who reported that the pH of the beverage was found to decrease with increase in storage and could be correlated inversely with the acidity of product. Decrease in pH with storage time, affects the organoleptic qualities of fruits blends (Awsi and Dorcus, 2012). Shukla *et al*. (2013) developed a probiotic beverage based on whey and pineapple juice with *Lactobacillus acidophilus* with a pH of 4.82. In the present study blended aloe juice neutraceuticals pH ranged from 2.21 to 4.10.The study of Biswas *et al*. (2016) reported that acidity increased (0.179 - 0.192) and pH of the juice decreased progressively during the storage period of *Aloe vera* and pineapple juice blended beverage and also reported that, with the increase in concentration of *Aloe vera* from T_0 to T_3, the pH value seem to be increasing. This was due to high pH of *Aloe vera* juice. In the present study pH was highest for T_7 - Honey + Liquidized aloe juice (50 : 50). pH content in aloe herbal health drink were shown in figure 14.

Table 77. pH of aloe herbal health drink during storage

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 M AS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	2.21	2.22	2.21	2.20	2.18
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	2.51	2.53	2.51	2.50	2.47
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	2.77	2.79	2.78	2.76	2.74
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	3.99	3.99	3.98	3.97	3.95
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	3.62	3.63	3.62	3.61	3.59
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	3.69	3.70	3.69	3.67	3.66
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	4.10	4.10	4.09	4.08	4.08
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	3.98	3.99	3.98	3.97	3.95
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	3.82	3.84	3.82	3.81	3.79
SEm (±)	0.002	0.002	0.002	0.002	0.002
CD (0.05)	0.006	0.006	0.005	0.005	0.005

*Each value is the mean of six replication

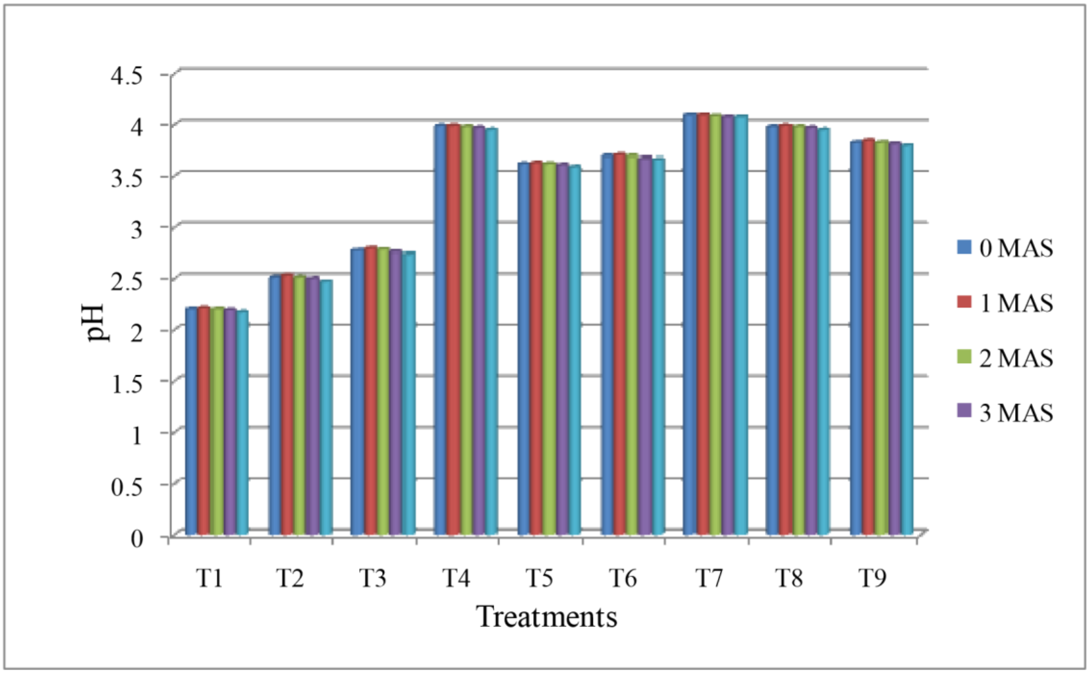


Figure 14. pH of aloe herbal health drink during storage

4.4.1.2.3 TSS

The TSS content of the treatments is presented in table 78.

Significantly higher TSS content during zero month after storage was noticed in T_7 - Honey + Liquidized aloe gel juice (50 : 50) recording a TSS of 51.67 ^{0}Brix followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with 32.50 ^{0}Brix. Lowest TSS value was noticed for T_3 - Lemon juice + Liquidized aloe gel juice (10: 90) with TSS content of about 1.83 ^{0}Brix. The next higher TSS was recorded from T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) and T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with a value of 4.67 ^{0}Brix. The TSS content ranged from 1.83 to 51.67 during zero month after storage.

The treatments showed significant difference in TSS during first month after storage. Treatment T_7 -Honey + Liquidized aloe gel juice (50:50) recorded a higher TSS content of 49.67 ^{0}Brix which was followed by treatment T_8 -Honey + Liquidized aloe gel juice (25 : 75) with a TSS of 29.83 ^{0}Brix. From the analysis the lowest value among the treatments was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with TSS content of about 1.67 ^{0}Brix. Treatment T_6 -Orange juice + Liquidized aloe gel juice (10 : 90) had a TSS content of 2.83 ^{0}Brix.

Statistical analysis of the data revealed that there was significant difference among the treatments during second month after storage. Highest TSS was noticed in Treatment T_7 - Honey + Liquidized aloe gel juice (50 : 50) with a TSS content of 47.83 ^{0}Brix which was followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with 27.83 ^{0}Brix TSS content. Among the treatments the least value was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with TSS content of about 1.50 ^{0}Brix which was followed by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) having TSS content of 2.67 ^{0}Brix.

A general decrease in TSS content was noticed in all the treatments during third month after storage. Among them highest pH was noticed for T_7 - Honey +

Liquidized aloe gel juice (50 : 50) which recorded a TSS content of 46.83 ^{0}Brix which was followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75), (25.83 ^{0}Brix). The lowest value among the treatments was observed in T_3 - Lemon juice + Liquidized aloe gel juice (10:90) with TSS content of about 0.67 ^{0}Brix while in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) TSS content recorded was 1.67 ^{0}Brix

The treatment T_7 - Honey + Liquidized aloe gel juice (50 : 50) recorded significantly superior TSS content (44.83 ^{0}Brix) during fourth month after storage which was followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with a TSS content of 23.83 ^{0}Brix. Least value among the treatments was observed in T_3 -Lemon juice + Liquidized aloe gel juice (10 : 90) with TSS content of about 0.17 ^{0}Brix. The treatment T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) recorded a TSS content of 0.83 ^{0}Brix. During the fourth month of storage the TSS content varied from 0.17 ^{0}Brix to 44.83 ^{0}Brix.

Wills *et al*. (1980) reported TSS content in apple juices decreased due to conversion of sugar into starch. In present study also TSS content decreased during storage. For preservation of good juice quality, retention or minimum increase in TSS content of juice during storage is desirable (Bhardwaj and Pandey, 2011)**,** but in present study the decrease in TSS content showed that during storage its quality was decreasing. Boghani *et al*. (2012) developed a blended aloe vera aonla based ready to-serve and the TSS ranged from 12 to 14.4 per cent on storage for six months. TSS content in the present study ranged from 1.83 ^{0}Brix to 51.67 ^{0}Brix among the neutraceuticals prepared, the highest TSS content was noticed in honey blended neutraceutical which might be due to the presence of honey. Ibironke *et al*. (2013) reported decrease in TSS content of tomato pulp during storage due to the breaking of solids. Reduction in total soluble solids was noticed due to the presence of microrganisms which causes deterioration of fruit blends because of sugar fermentation (Ana *et al*., 2013) and this supported the higher variation in TSS content of aloe juice blended with orange juice and lemon juice.

Constant pH, TSS, titrable acidity and viscosity of fruit juice beverages clearly indicated that there was no spoilage either due to microbial or enzymatic reaction (Renuka *et al*., 2009). In apple cultivars the increase in TSS was attributed to the breakdown of starch into sugars or the hydrolysis of cell wall polysaccharides (Jan and Rab, 2012). TSS content variation in aloe herbal health drink was presented in figure 15.

Table 78. TSS of aloe herbal health drink during storage, ^{0}Brix

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 M AS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	5.83	5.00	3.67	3.50	2.67
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	4.67	4.33	2.83	2.33	2.17
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	1.83	1.67	1.50	0.67	0.17
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	9.83	7.67	5.83	4.83	4.17
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	8.67	6.83	3.83	3.83	2.67
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	4.67	2.83	2.67	1.67	0.83
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	51.67	49.67	47.83	46.83	44.83
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	32.50	29.83	27.83	25.83	23.83
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	24.67	20.67	15.83	11.83	10.25
SEm (±)	0.199	0.174	0.184	0.179	0.167
CD (0.05)	0.568	0.497	0.527	0.512	0.478

*Each value is the mean of six replication

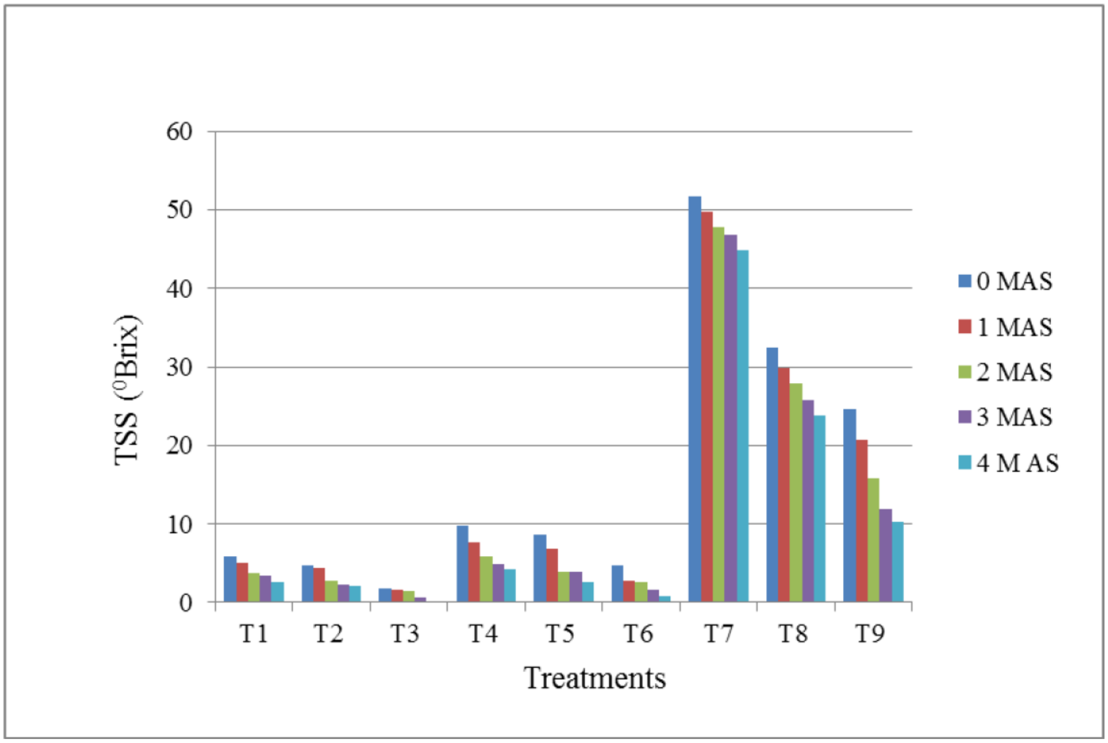


Figure 15. TSS of aloe herbal health drink during storage

4.4.1.2.4 Vitamin C

The vitamin C content of aloe herbal health drink produced by different combinations of lemon juice, orange juice and honey with stabilized Liquidized aloe gel juice is presented in table 79. The vitamin C content decreased from zero month after storage to fourth month after storage.

The vitamin C content was found to be significant among the treatment during zero month after storage. The highest vitamin C among the treatments was noticed inT_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) with a vitamin C content of 45.56 mg/100 ml which was followed by T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) having vitamin C content of about 42.32 mg/ 100 ml. Vitamin C content in T_9 - Honey + Liquidized aloe gel juice (10: 90) was 0.33 mg/100 ml which was found to be lowest and that of T_8 - Honey + Liquidized aloe gel juice (25 : 75) was 0.89 mg/ 100 ml.

On first month after storage the treatments showed significant difference in the case of vitamin C content among the treatments. The highest acidity content of 43.57 mg/100 ml was recorded in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) followed by T_4 - Orange juice + Liquidized aloe gel juice (50:50) with vitamin C content of 41.30 mg/100ml. Vitamin C content was lowest in T_9 - Honey + Liquidized aloe gel juice (10 : 90), (0.32 mg/100 ml) and that of T_8 - Honey + Liquidized aloe gel juice (25 : 75), was 0.87 mg/ 100 ml.

During second month after storage significantly highest value was noticed in T_1 -Lemon juice + Liquidized aloe gel juice (50 : 50), (42.33 mg/100 ml) which was followed by T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) having an vitamin C content of about 40.28 mg/100ml. Lowest vitamin C content among the health drinks was observed in T_9 - Honey + Liquidized aloe gel juice (10 : 90), with 0.31 mg/100 ml which was followed by T_8 - Honey + Liquidized aloe gel juice (25 : 75) with 0.85 mg/ 100 ml. The vitamin C content value ranged from 0.31 mg/100 ml to 42.33 mg/100 ml during second month after storage.

Significant variation was noticed among the treatments during third month after storage and it showed a decreasing trend. Among them highest content was noticed for T_1 - Lemon juice + Liquidized aloe gel juice (50: 50) which recorded a value of about 41.32 mg/100 ml which was followed by T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) having vitamin C content of about 40.01mg/100 ml. The least value was shown by T_9 - Honey (10 ml) + Liquidized aloe gel juice (10 : 90), with a vitamin C content of 0.30 mg/100ml. Treatment T_8 - Honey + Liquidized aloe gel juice (25 : 75) had a vitamin C content of 0.84 mg/100 ml.

There was significant difference in different treatments of aloe herbal health drinks in the case of vitamin C content during fourth month after preparation. The highest content was observed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) which recorded a value of about 40.56 mg/100 ml which was followed by T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) having an vitamin C content of about 39.21 mg/ 100 ml. Vitamin C content of 0.29 mg/100 ml which was the lowest in T_9 - Honey + Liquidized aloe gel juice (10 : 90). In T_8 - Honey + Liquidized aloe gel juice (25 : 75) the vitamin C content was 0.82 mg/ 100 ml. Vitamin C content of the neutraceuticals during fourth month after storage ranged from 0.29 mg/ 100ml to 40.56 mg/ 100 ml.

Vitamin C content of all roselle-fruit blends decreased during storage with the advancement of storage period, which was probably due to the sensitivity of vitamin C to oxygen, light and heat (Ziena, 2000). Majumdar *et al*. (2009) reported that after 6 months of storage 74 percent loss in vitamin C was observed in cucumber-litchi-lemon blended juice.

According to Inga *et al* (2007) the vitamin C is an important parameter for assessing the nutritional quality of fruits blends, as it degrades during storage. The trend of decrease in vitamin C in storage studies was reported in developed beverage and pickle (Mishra *et al*., 2010; Puranik *et al.*, 2011). According to Pena *et al.* (2011)

when vitamin C retention decreased to 50 percent of its initial amount, shelf life of the product ends. A loss in vitamin C was observed in guava blended with *Aloe vera* and roselle during 120 days of storage at ambient temperature (Kumar *et al*. 2012). Tiwari and Deen (2015) reported Vitamin C content continuously decreased from the first day (2.38 mg/100g) to the end of storage (1.93 mg/100g) throughout the storage period in blended ready to serve beverage from Bael and *Aloe vera* and reported that decrease in vitamin C content might be due to the oxidation of ascorbic acid into dehydro ascorbic acid. The above said studies supported the fact that the vitamin C content in the *Aloe vera* neutraceuticals continuously decreased, except the aloe juice blended beverage with honey which showed less variation during storage.Variation in vitamin C content during storage explained in figure 16.

Table 79. Vitamin C content of aloe herbal health drink during storage, (mg/100 ml)

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 M AS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	45.56	43.57	42.33	41.32	40.56
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	34.53	33.21	31.52	30.52	29.53
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	14.82	14.16	13.83	12.83	10.71
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	42.32	41.30	40.28	40.01	39.21
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	31.66	31.62	30.62	29.63	28.42
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	11.82	11.81	11.79	10.82	10.01
T_7- Honey + Liquidized aloe gel juice (50 : 50)	1.03	1.02	1.01	1.01	1.00
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	0.89	0.87	0.85	0.84	0.82
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	0.33	0.32	0.31	0.30	0.29
SEm (±)	0.003	0.002	0.003	0.002	0.002
CD (0.05)	0.010	0.006	0.010	0.005	0.007

*Each value is the mean of six replication

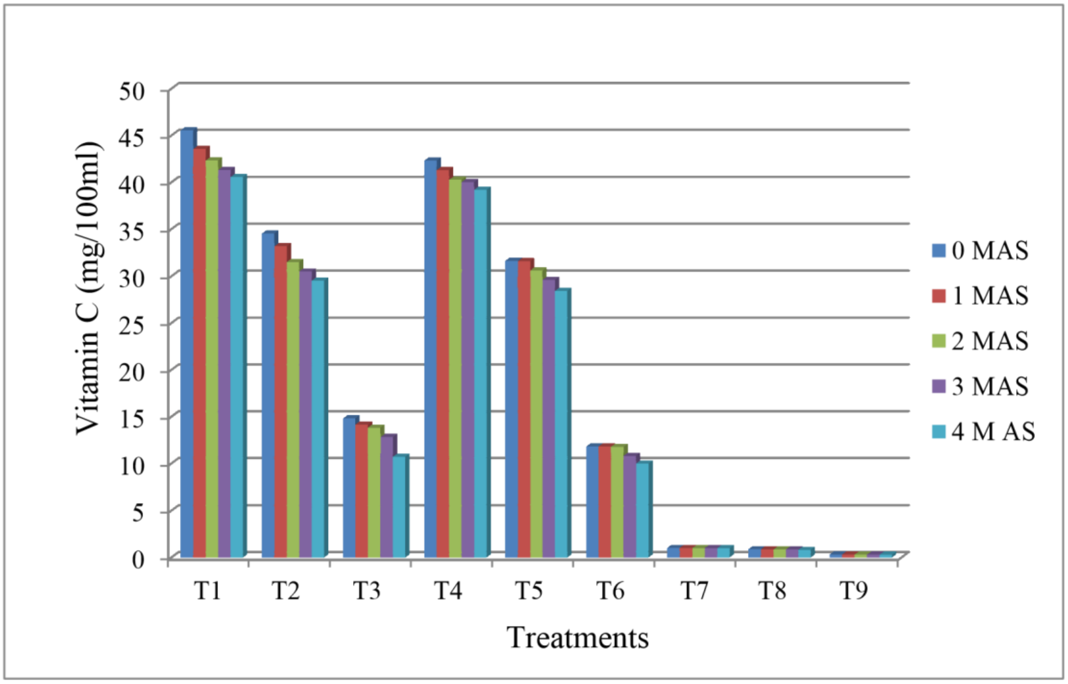


Figure 16. Vitamin C content of aloe herbal health drink during storage

4.4.1.2.5 Energy/calories

The energy content of the treatments is presented in table 80.

Significantly higher energy content during zero month after storage was noticed in T_7 – Honey + Liquidized aloe gel juice (50 : 50) with 11.17 kcal/100mg followed by T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with an energy content of 9.44 kcal/100 mg. Lowest energy value was noticed for T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with energy content of about 2.24 kcal/100 mg while T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) had an energy value of 2.30 kcal/100 mg. The energy content of neutraceuticals during zero month after storage ranged from 2.24 kcal/100 mg to 11.17 kcal/100 mg.

The treatments showed significant difference in energy during first month after storage. Treatment T_7 - Honey + Liquidized aloe gel juice (50 : 50) recorded a higher energy content of 11.06 kcal/100 mg which was followed by treatment T_4- Orange juice + Liquidized aloe gel juice (50 : 50) which showed 9.38 kcal/100 mg energy. Lowest value among the treatments was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with energy content of about 2.13 kcal/100 mg while that of T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) was 2.25 kcal/100 mg.

The data on energy revealed that there was significant difference among the treatments during second month after preparation. Highest value was noticed in Treatment T_7 – Honey + Liquidized aloe gel juice (50 : 50) recorded an energy content of about 10.99 kcal/100 mg which was followed by T_4 - Orange juice + Liquidized aloe gel juice (50 : 50), (9.38 kcal/100 mg). Among the treatments the least value was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with energy content of about 2.10 kcal/100 mg which was followed by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) having energy content of 2.25 kcal/100 mg.

A general decrease in energy content was noticed in all the treatments during third month after storage. Among them highest energy content was noticed for

T_7 - Honey + Liquidized aloe gel juice (50 : 50) which recorded an energy value of 10.81 kcal/100 mg which was followed by T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with 9.29 kcal/100 mg. The lowest value among the treatments was observed in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with energy content of about 2.01 kcal/100 mg which was followed by T_6 - Orange juice + Liquidized aloe gel juice (10:90) with 2.17 kcal/100 mg energy value.

The treatment, T_7 - Honey + Liquidized aloe gel juice (50 : 50) recorded significantly superior energy value (10.61 kcal/100 mg) during fourth month after preparation which was followed by T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with 9.21 kcal/100 mg. Least value among the treatments were observed in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with energy content of about 1.85 kcal/100 mg. During the final month after storage the energy content varied from 1.85 kcal/100 mg to 10.61 kcal/100 mg.

Energy content of the probiotic honey beverage was estimated as 288 kcal/100 g which was slightly higher than the non probiotic beverage of 256 kcal/100 g (Nath *et al*., 2015). The calculated energy value of *Aloe vera* - aonla blended functional squash using stevioside recorded 53.65 kcal/100 g compared to control (Sharma *et al*., 2018). During present study the energy content of health drink varied from 2.24 to 11.17 kcal/100mg. The energy for the aloe honey (50 : 50) health drink was more than the aloe orange and aloe lemon juice health drink of all the combinations which might be because of high energy of honey. Energy content in health drink were shown in figure 17.

Table 80. Energy/calories of aloe herbal health drink during storage, kcal/100 mg

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	7.60	7.47	7.39	7.26	7.10
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	3.99	3.90	3.77	3.65	3.45
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	2.24	2.13	2.10	2.01	1.85
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	9.44	9.38	9.38	9.29	9.21
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	4.46	4.41	4.41	4.33	4.25
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	2.30	2.25	2.25	2.17	2.08
T_7- Honey + Liquidized aloe gel juice (50 : 50)	11.17	11.06	10.99	10.81	10.61
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	5.04	4.94	4.85	4.77	4.61
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	3.36	3.22	3.14	2.97	2.81
SEm (±)	0.015	0.013	0.016	0.013	0.015
CD (0.05)	0.044	0.036	0.045	0.036	0.042

*Each value is the mean of six replication

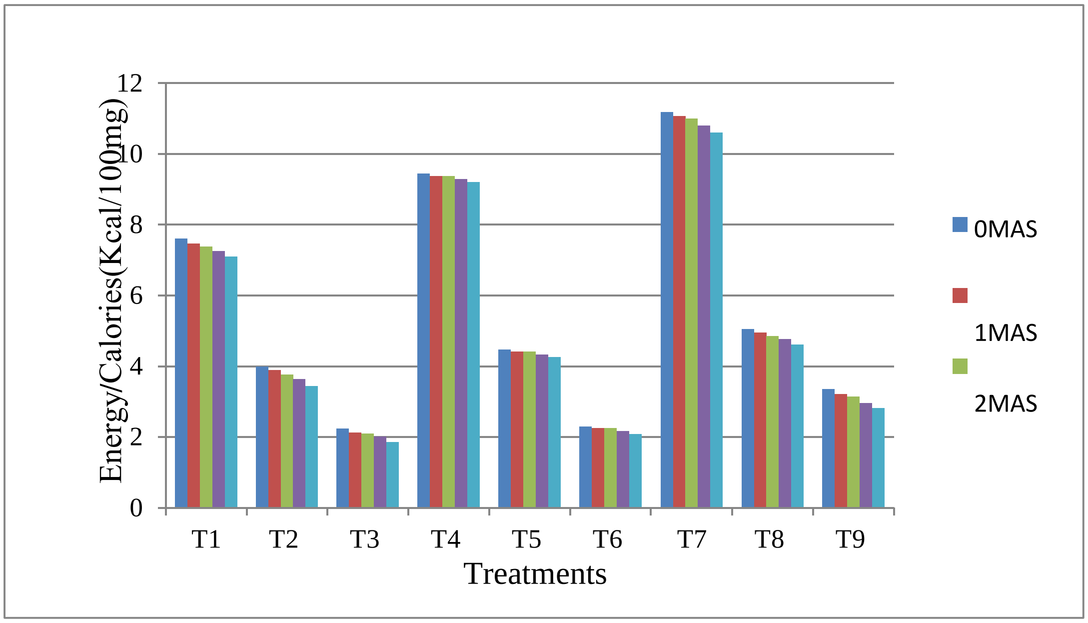


Figure 17. Energy/calorie content of aloe herbal health drink during storage

4.4.1.2.6 Iron

The iron content of aloe herbal health drink is presented in table 81.

The iron content was found to be significant among the treatment during zero month after preparation. The highest value among the treatments was noticed in T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with 0.08 per cent iron content which was followed by T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) and T_7 - Honey + Liquidized aloe gel juice (50 : 50) which was having an iron content of about 0.05 per cent. Lowest value was noticed in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90), T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and T_9 - Honey + Liquidized aloe gel juice (10 : 90) recording an iron content of about 0.02 per cent.

The iron content was significantly different under different treatments throughout first month after preparation. Treatment, T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) recorded highest iron content of 0.07 per cent which was followed by T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) and T_7 – Honey + Liquidized aloe gel juice (50 : 50) with iron content of 0.05 per cent. The least value among the treatments was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90), T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and T_9 - Honey + Liquidized aloe gel juice (10 : 90) with 0.02 per cent iron content.

Significant differences were observed among various treatments with respect to iron content during second month after storage. The results indicated that highest iron content was noticed in treatment ,T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with 0.06 per cent iron content followed by T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) and T_7 - Honey + Liquidized aloe gel juice (50 : 50) both having 0.04 per cent iron content. Among the treatments the least value was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) and T_9 - Honey + Liquidized aloe gel juice (10 : 90) with 0.01 per cent iron content. Treatments, T_5 - Orange juice +

Liquidized aloe gel juice (25 : 75) and T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) had shown 0.02 per cent iron content.

The iron content significantly differed among various treatments during third month after storage and highest value was noticed in T_4 - Orange juice + Liquidized aloe gel juice (50 : 50), with 0.04 per cent followed by T_7 - Honey + Liquidized aloe gel juice (50:50) which recorded an iron content of 0.03 per cent. The lowest value among the treatments was observed in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90), T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and T_9 - Honey + Liquidized aloe gel juice (10 : 90) with 0.01 per cent iron content.

During fourth month after storage, the iron content significantly varied among treatments. The highest iron content of 0.03 per cent was noticed in T_4 -Orange juice + Liquidized aloe gel juice (50 : 50), which was followed by T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50), T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) and T_7 -Honey + Liquidized aloe gel juice (50 : 50) all having an iron content of about 0.02 per cent. All the other five treatments showed an iron content of about 0.01per cent.

The calcium and iron contents of value added nutraceutical beverages of guava blended with *Aloe vera* and roselle indicated reduction in the minerals from their initial values (Kumar *et al*., 2012). This supported the present study which also showed decreasing trend in iron content of the health drink during storage. The iron content variation in aloe herbal health drink were explained in Figure 18.

Table 81. Iron content of aloe herbal health drink during storage, percentage

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 M AS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	0.05	0.05	0.04	0.02	0.02
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	0.04	0.04	0.03	0.02	0.01
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	0.02	0.02	0.01	0.01	0.01
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	0.08	0.07	0.06	0.04	0.03
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	0.03	0.03	0.02	0.02	0.02
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	0.02	0.02	0.02	0.01	0.01
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	0.05	0.05	0.04	0.03	0.02
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	0.04	0.04	0.03	0.02	0.01
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	0.02	0.02	0.01	0.01	0.01
SEm (±)	0.000	0.000	0.000	0.000	0.000
CD (0.05)	0.001	0.001	0.001	0.001	0.001

*Each value is the mean of six replication

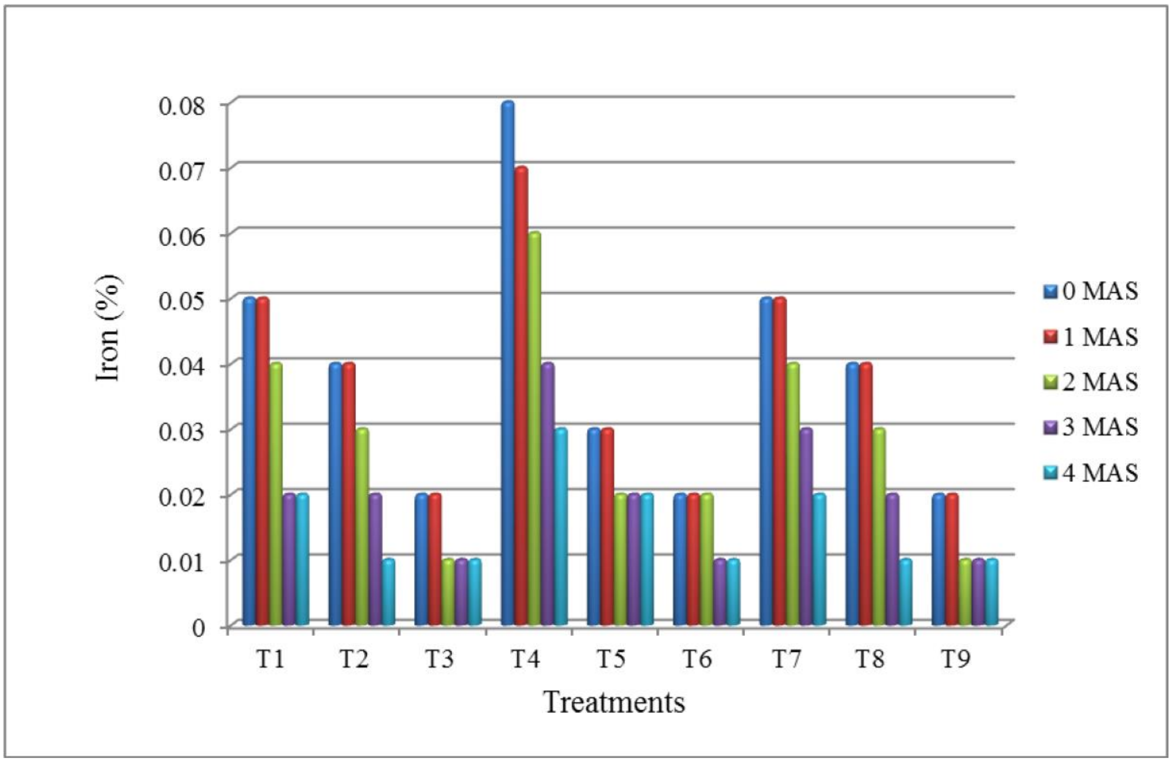


Figure18. Iron content of aloe herbal health drink during storage

4.4.1.2.7 Calcium

Significant variation in calcium content of aloe herbal health drink was noticed only in 0 MAS and 1 MAS and was non significant during the following period and is presented in table 82.

The calcium content was found to be significant among the treatment during zero month after storage. The highest calcium content of 0.008 per cent among the treatments was noticed in T_7 - Honey + Liquidized aloe gel juice (50 : 50) which was on par with T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) having a calcium content of about 0.007 per cent. Calcium content was found to be lowest in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90), T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and T_9 - Honey + Liquidized aloe gel juice (10 : 90) with a calcium content of 0.002 per cent which was on par with T_2- Lemon juice + Liquidized aloe gel juice (25:75), T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) and T_8 - Honey + Liquidized aloe gel juice (25 : 75) having calcium content of 0.003 per cent.

On first month after storage the treatments showed significant difference in the case of calcium content among the treatments. The highest calcium content of 0.008 per cent was recorded in T_7 – Honey + Liquidized aloe gel juice (50 : 50) which was followed by T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) and T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) having calcium content of about 0.006 per cent. Calcium content was lowest for T_6 - Orange juice + Liquidized aloe gel juice (10 : 90), (0.001per cent) which was on par with T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90), T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) and T_9 - Honey + Liquidized aloe gel juice (10 : 90) with 0.002 per cent. No variation in calcium content was noticed during 0 MAS and 1 MAS in treatment T_7 - Honey + Liquidized aloe gel juice (50 : 50).

From second month of storage onwards the calcium content were found to be non – significant for all the treatments.

Kenawi *et al.* (1994) reported a very slight change in the calcium content (4.5 and 4 per cent) after ten weeks of storage for both calcium unfortified and the fortified orange juices respectively. In the present study the calcium content differed among juices during the initial period and on 1 MAS not much difference was noticed in calcium content among the health drinks. Calcium content in aloe herbal health drink presented in Figure 19.

Table 82. Calcium content of aloe herbal health drink during storage, percentage

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	0.006	0.006	0.005	0.004	0.003
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	0.003	0.003	0.002	0.002	0.001
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	0.002	0.002	0.001	0.001	0.001
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	0.007	0.006	0.006	0.005	0.004
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	0.003	0.002	0.002	0.002	0.001
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	0.002	0.001	0.001	0.001	0.001
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	0.008	0.008	0.008	0.007	0.006
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	0.003	0.003	0.003	0.002	0.001
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	0.002	0.002	0.002	0.001	0.001
SEm (±)	0.000	0.000	0.000	0.000	0.000
CD (0.05)	0.001	0.001	NS	NS	NS

*Each value is the mean of six replication

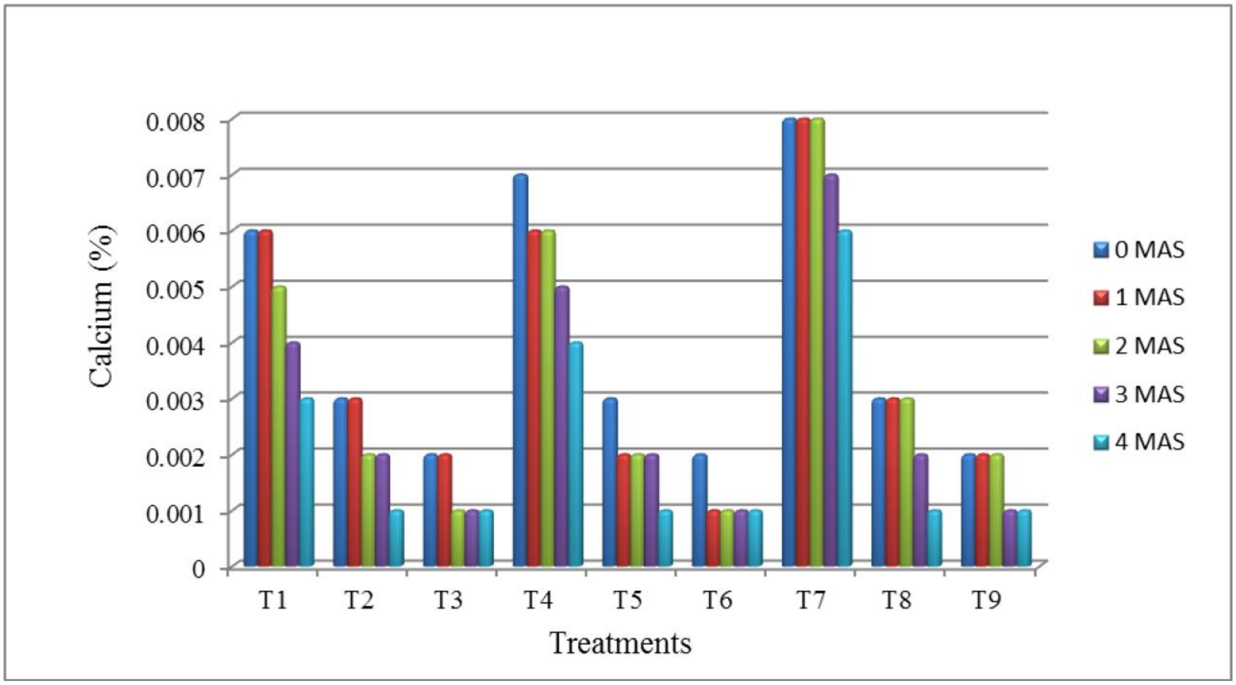


Figure 19. Calcium content of aloe herbal health drink during storage

4.4.1.2.8 Carbohydrate

The carbohydrate content of the aloe herbal health drink was analysed and presented in table 83.

Significantly higher carbohydrate content during zero month after storage were noticed in T_7 - Honey + Liquidized aloe gel juice (50 : 50) with 2.71 per cent followed by treatment T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with 2.32 per cent. Lowest carbohydrate value was noticed in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with carbohydrate content of about 0.48 per cent followed by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) having 0.55 per cent carbohydrate content. During zero month after storage the carbohydrate content ranged from 0.48 per cent to 2.71 per cent.

The treatments showed significant difference during first month after storage. treatment T_7 - Honey + Liquidized aloe gel juice (50 : 50) recorded a higher carbohydrate content of 2.70 per cent which was followed by treatment T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with 2.32 per cent. The lowest value among the treatments was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10:90) with carbohydrate content of about 0.47 per cent followed by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with carbohydrate content of 0.55 per cent.

There was significant variation among the treatments in carbohydrate content during second month after storage. The highest carbohydrate content was noticed in Treatment T_7 – Honey + Liquidized aloe gel juice (50 : 50) with 2.69 per cent followed by T_4 - Orange juice + liquidized aloe gel juice (50 : 50 ml) with 2.32 per cent. Among the treatments the least value of 0.47 per cent was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) followed by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) having carbohydrate content of 0.55 per cent. The carbohydrate content noticed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) was 1.75 per cent,T_2- Lemon juice + Liquidized aloe gel juice (25 : 75) was 0.87 per cent, T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) was 1.09 per cent, T_8 - Honey + Liquidized aloe gel juice (25 : 75) was1.18 per cent and T_9 - Honey + Liquidized aloe gel juice (10 : 90) was 0.77 per cent.

The significant changes in carbohydrate were noticed during third month after storage. Among them highest carbohydrate content was noticed for T_7 - Honey + Liquidized aloe gel juice (50 : 50) which recorded 2.65 per cent followed by T_4- Orange juice + Liquidized aloe gel juice (50 : 50) with 2.31 per cent. The lowest value among the treatments was observed in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with carbohydrate content of about 0.45 per cent which was followed by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) having carbohydrate content of about 0.54 per cent.

The treatment T_7 - Honey + Liquidized aloe gel juice (50 : 50) recorded significantly superior carbohydrate content (2.62 per cent) during fourth month after storage which was followed by T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with 2.29 per cent. Least value among the treatments was observed in T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) with carbohydrate content of about 0.43 per cent which was followed by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) having carbohydrate content of 0.52 per cent. During fourth month after storage the carbohydrate content varied from 0.43 per cent to 2.62 per cent among the treatments.

According to Lee and Nagy (1988) the fructose, glucose and sucrose got degraded in grapefruit juice during storage at 37 ^{0}C for 16 weeks. The polysaccharides found in aloe gel are not stable, especially under stress conditions such as heat, the presence of acid and enzymatic activities and it is the main factor contributing to health benefit (Hamman, 2008). The decrease in carbohydrate on storage might be due to degradation of polysaccharides to simple sugars due to enzymatic activity.Variation in carbohydrate content of aloe herbal health were presented in figure 20.

Table 83. Carbohydrate content of aloe herbal health drink during storage, percentage

Treatments	0 M AS	1 MAS	2 MAS	3 MAS	4 M AS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	1.78	1.76	1.75	1.73	1.71
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	0.90	0.89	0.87	0.85	0.82
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	0.48	0.47	0.47	0.45	0.43
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	2.32	2.32	2.32	2.31	2.29
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	1.09	1.09	1.09	1.08	1.06
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	0.55	0.55	0.55	0.54	0.52
T_7- Honey + Liquidized aloe gel juice (50 : 50)	2.71	2.70	2.69	2.65	2.62
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	1.20	1.19	1.18	1.16	1.14
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	0.80	0.78	0.77	0.73	0.70
SEm (±)	0.002	0.002	0.002	0.002	0.002
CD (0.05)	0.006	0.006	0.006	0.007	0.006

*Each value is the mean of six replication

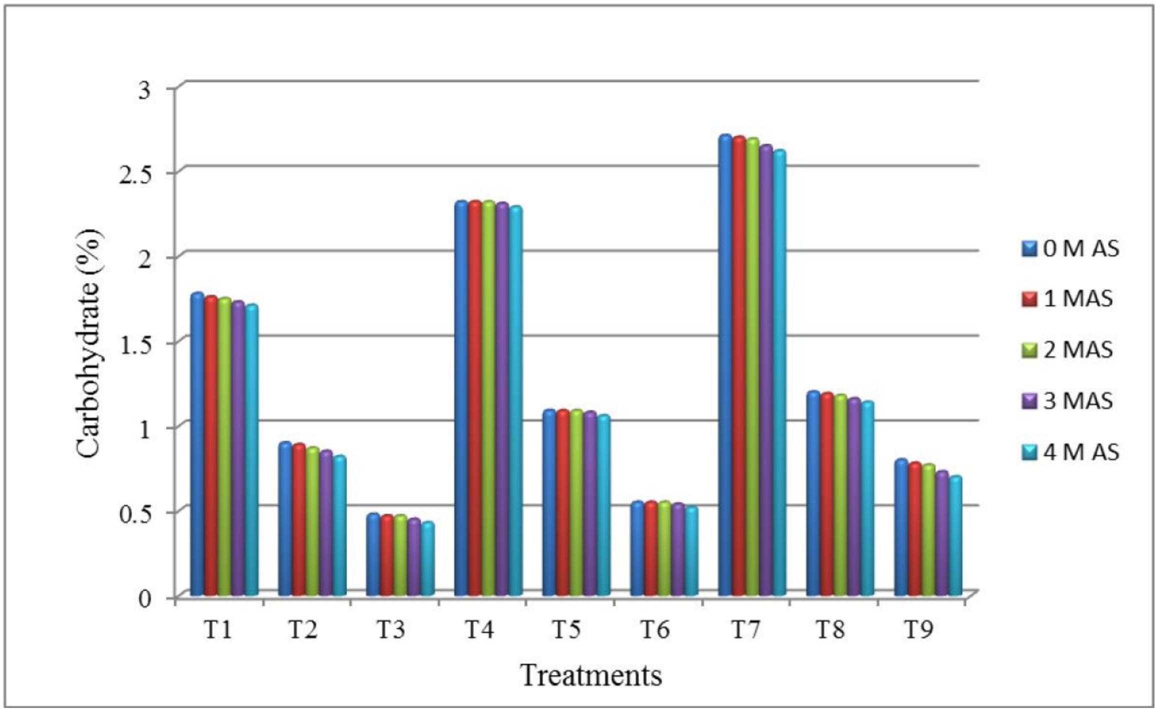


Figure 20. Carbohydrate content of aloe herbal health drink during storage

4.4.1.2.9. Fat

The data on the fat content of the neutraceuticals during zero month after storage upto fourth month after storage is presented in table 84.

There was significant difference in the fat content under different treatments during zero month after storage. The highest value were noticed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) and T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) having fat content of about 0.002 per cent. All the other treatments showed fat content of about 0.001per cent except T_8 - Honey + Liquidized aloe gel juice (25 : 75) and T_9 - Honey + Liquidized aloe gel juice (10 : 90).

No significant difference in fat content was noticed among the treatments during first month after storage.

The fat content varied significantly among treatments during second month after storage. In the evaluation, maximum fat content was recorded by T_1- Lemon juice + Liquidized aloe gel juice (50 : 50) and T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with 0.002 per cent. How ever no fat content was detected in T_8 - Honey + Liquidized aloe gel juice (25 : 75) and T_9 - Honey + Liquidized aloe gel juice (10 : 90). All the other treatments like T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75), T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90), T_5 - Orange juice + Liquidized aloe gel juice (25 : 75), T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and T_7 - Honey + Liquidized aloe gel juice (50 : 50) recorded a fat content of about 0.001 per cent.

No significant difference was noticed among the treatments during third month after storage and fourth month after storage.

According to Chandegara and Varshney (2013) fat content in aloe gel was reported as 0.09 g per 100 g. The present study reported fat content in aloe juice blended with honey, orange juice and lemon juice ranges from 0.000 per cent to 0.002 per cent and found to be non significant for first month after storage, third month after storage and fourth month after storage. Fat content in aloe herbal health drink were shown in figure 21.

Table 84. Fat content of aloe herbal health drink during storage, percentage

Treatments	0 M AS	1 MAS	2 MAS	3 MAS	4 M AS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	0.002	0.002	0.002	0.001	0.000
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	0.001	0.001	0.001	0.000	0.000
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	0.001	0.001	0.001	0.000	0.000
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	0.002	0.002	0.002	0.001	0.000
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	0.001	0.001	0.001	0.000	0.000
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	0.001	0.001	0.001	0.000	0.000
T_7- Honey + Liquidized aloe gel juice (50 : 50)	0.001	0.001	0.001	0.000	0.000
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	0.000	0.000	0.000	0.000	0.000
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	0.000	0.000	0.000	0.000	0.000
SEm (±)	0.000	0.000	0.000	0.000	0.000
CD (0.05)	0.001	NS	0.001	NS	NS

*Each value is the mean of six replication

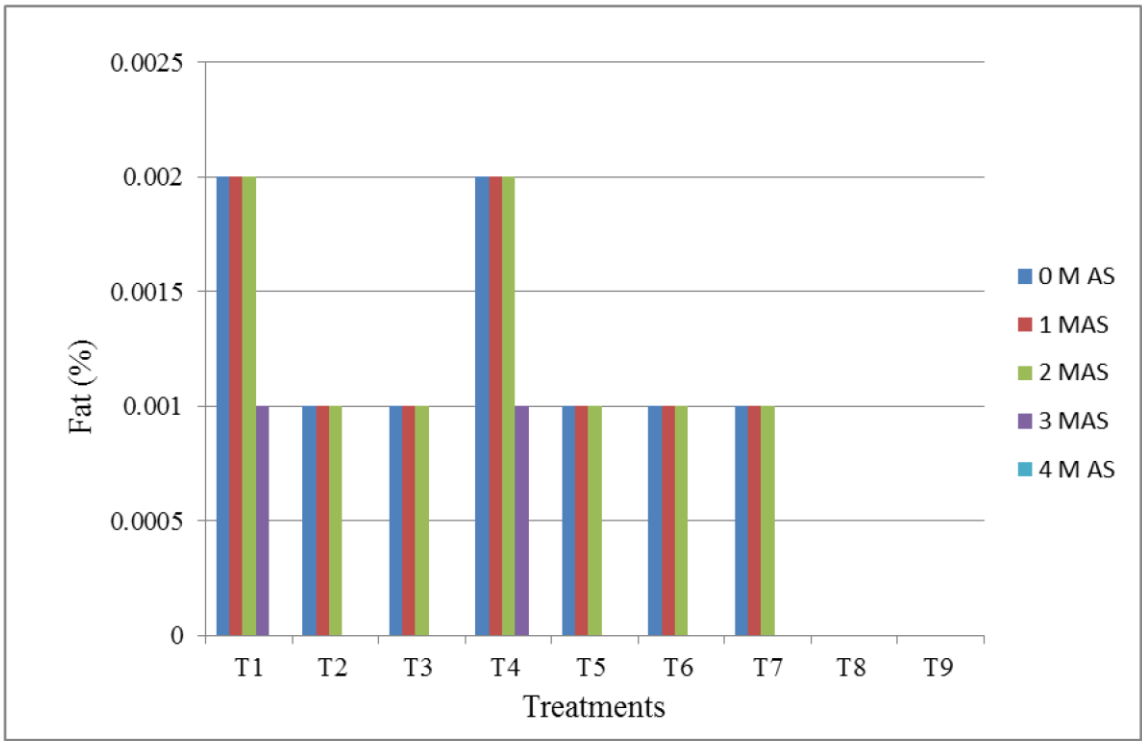


Figure 21. Fat content of aloe herbal health drink during storage

4.4.1.2.10 Protein

The protein content of aloe herbal health drink made of varying proportion of lemon juice, orange juice and honey with stabilized liquidized aloe gel juice is presented in table 85.

The protein content differed significantly among treatments during zero month after storage. Treatment T_1 - Lemon juice + Liquidized aloe gel juice (50:50) recorded maximum protein content of 0.11 per cent, followed by T_2 - Lemon juice + liquidized aloe gel juice (25 : 75) with 0.09 per cent protein content. Lowest value was noticed for T_8 - Honey + Liquidized aloe gel juice (25 : 75) and T_9 - Honey + Liquidized aloe gel juice (10 : 90) with a protein content of about 0.02 per cent which was followed by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and T_7 - Honey + Liquidized aloe gel juice (50 : 50) having 0.03 per cent protein content. The protein content of treatments T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) and T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) was 0.07 per cent and T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) was 0.05 per cent.

From the data it was revealed that the protein content varied significantly under different treatments throughout first month after storage and highest value was shown by the treatment, T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) with 0.11 per cent followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) with protein content of 0.09 per cent. The least value among the treatments was shown by T_8 - Honey + Liquidized aloe gel juice (25 : 75) and T_9 - Honey + Liquidized aloe gel juice (10 : 90) with 0.02 per cent protein content.

There was significant difference among various treatments with respect to protein content during second month after storage. The results indicated that highest value was noticed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) with 0.10 per cent which was followed by T_2 - Lemon juice + Liquidized aloe gel juice having 0.08 per cent protein content. Among the treatments the least value was shown by

T_6 - Orange juice + Liquidized aloe gel juice (10 : 90), T_8 - Honey + Liquidized aloe gel juice (25 : 75) and T_9 - Honey + Liquidized aloe gel juice (10 : 90) with 0.02 per cent protein content which were followed T_7 - Honey + Liquidized aloe gel juice (50 : 50) with 0.03 per cent protein content.

From the analysis it was observed that statistically highest value for protein content during third month after storage was noticed for T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) with 0.09 per cent which was followed by T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) which recorded protein content of 0.07per cent. The lowest value of 0.01 per cent protein content was observed inT_8 - Honey + Liquidized aloe gel juice (25 : 75) and T_9 (Honey + Liquidized aloe gel juice (10 : 90). In T_6 -Orange juice + Liquidized aloe gel juice (10 : 90) and T_7 - Honey + Liquidized aloe gel juice (50 : 50) the protein content noted was 0.02 per cent. The treatment T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) and T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) recorded 0.06 per cent and T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) recorded 0.04 per cent.

Fourth month after storage the treatments significantly varied with respect to protein content. The highest protein content was noticed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) (0.07 %) followed by T_2 - Lemon juice + Liquidized aloe gel juice with 0.05 per cent .The least value of 0.00 per cent protein content were observed in T_9 – Honey + Liquidized aloe gel juice (10 : 90) which was followed by T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) and T_8 - Honey + Liquidized aloe gel juice (25 : 75) with 0.01 per cent protein content. The protein content ranged from 0.00 per cent to 0.007per cent among various treatments.

Protein content of 0.11g per 100g of aloe gel was reported by Chandegara and Varshney (2013). In the study the protein content in neutraceuticals ranged from 0.02 to 0.11 percent during zero month after storage. The protein content of *Aloe vera* and pineapple juice blended beverage showed a slight increase with the increase in the quantity of aloe juice (Biswas *et al*., 2016). The effect of *Aloe vera* juice incorporation on protein content of the peda samples during storage at room temperature (37±1^{0}C) revealed that the protein was not affected by the incorporation of *Aloe vera* juice and storage period (Srikanth *et al*., 2017). In the present study the protein content between aloe health drinks differed and significantly higher protein content was noticed in aloe lemon juice in different proportions than other combinations. How ever on storage a slight decrease in protein content was noticed which might be due to slight degradation. Variation in protein content of aloe herbal health drink with clove oil presented in figure 22.

Table 85. Protien content of aloe herbal health drink during storage, percentage

Treatments	0 M AS	1 MAS	2 MAS	3 MAS	4 MAS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	0.11	0.11	0.10	0.09	0.07
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	0.09	0.09	0.08	0.07	0.05
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	0.07	0.07	0.06	0.06	0.04
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	0.07	0.07	0.06	0.06	0.04
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	0.05	0.05	0.04	0.04	0.02
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	0.03	0.03	0.02	0.02	0.01
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	0.03	0.03	0.03	0.02	0.02
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	0.02	0.02	0.02	0.01	0.01
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	0.02	0.02	0.02	0.01	0.00
SEm (±)	0.002	0.002	0.002	0.002	0.002
CD (0.05)	0.006	0.006	0.006	0.006	0.007

*Each value is the mean of six replication

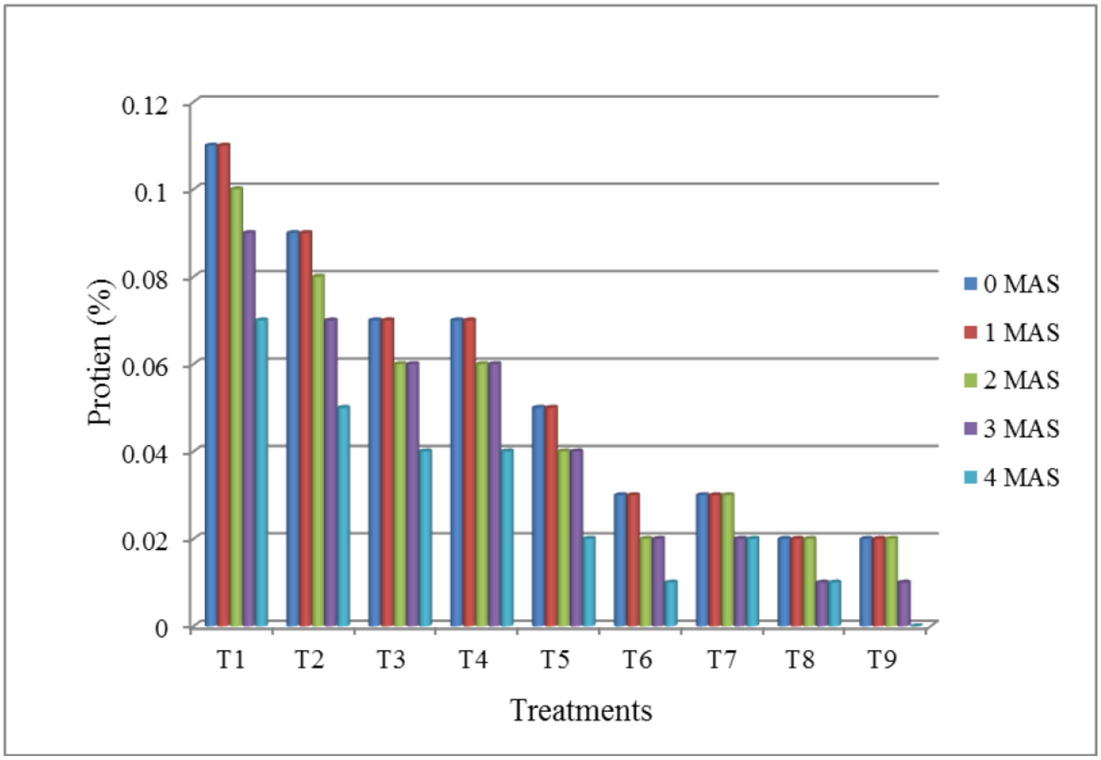


Figure 22. Protien content of aloe herbal health drink during storage

4.4.1.3 Shelf life

Shelf life of aloe - herbal health drink was determined by considering microbial population and sensory parameters like color, odour, taste, appearance and overall acceptability. Microbial population like mean fungal, bacterial and *E.coli* population is presented in tables 86 and 87, respectively. The fungal and bacterial population were absent up to second month after storage. No *E.coli* population was observed during the storage period of upto fourth month.

During third month after storage the fungal population was significantly different and highest value was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) and T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with a population of about 3.10 log cfu ml^{-1}. All treatments were on par with T_3 and T_6 except the treatment T_7, T_8 and T_9. T_7 - Honey + Liquidized aloe gel juice (50:50) recorded least fungal population of 0.50 log cfu ml^{-1}. T_8 - Honey + Liquidized aloe gel juice (25 : 75) had a fungal population of 1.50 log cfu ml^{-1} and T_9 - Honey + Liquidized aloe gel juice (10 : 90) had a fungal population of 2.00 log cfu ml^{-1}.

During fourth month after storage least value were shown by the treatment $T_{7.}$ The mean fungal population was significantly higher for T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) and T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) during fourth month after storage with a fungal population of 3.20 log cfu ml^{-1} and was on par with all the treatments except T_7 - Honey + Liquidized aloe gel juice (50 : 50) which recorded a population of 1.50 log cfu ml^{-1}.

There was no bacterial population during the first two months of storage in various treatments. The mean bacterial population significantly varied during third month after storage and fourth month after storage. During third month after storage the highest bacterial count was shown by T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90) and T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with 6.15 log cfu ml^{-1} which was on par with T_1 - Lemon juice + Liquidized aloe gel juice (50 :

50) and T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) both recording a population of 6.05 log cfu ml^{-1}. T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75) and T_5 - Orange juice + Liquidized aloe gel juice (25 : 75) recorded a bacterial population of 6.10 log cfu ml^{-1}. The treatment T_7 - Honey + Liquidized aloe gel juice (50 : 50) recorded no bacterial population at third month after storage. How ever in T_8 - Honey + Liquidized aloe gel juice (25 : 75) the bacterial population recorded was 1.00 log cfu ml^{-1}. The T_9 - Honey + Liquidized aloe gel juice (10 : 90) recorded bacterial population of 2.00 log cfu ml^{-1} and was on par with T_8. The highest bacterial population was observed during fourth month after storage in T_6 - Orange juice + Liquidized aloe gel juice (10 : 90) with 6.26 log cfu ml^{-1} and was on par with all the treatments except T_7 - Honey + Liquidized aloe gel juice (50 : 50) which recorded a bacterial population of 2.05 log cfu ml^{-1}.

Chauhan *et al*. (2012) reported slight increase in microbial count in functional herbal RTS beverage. The storage studies revealed that blended therapeutic RTS made from *Aloe vera* gel aonla fruits and ginger juices extracts could be successfully stored for the period of four months without significant change in chemical and sensory qualities (Kumar *et al*., 2013). Hirdyani (2015) and Hirdyani and Soni (2016) reported microbial load in RTS beverage prepared using traditional medicinal plants.The mixed jackfruit and aloe vera functional RTS beverage was stored up to five months without notable change in chemical and sensorial parameters at refrigerated temperature (Hossain *et al*., 2017). The study of *Aloe vera* fortified beverage with lemon oil revealed that the combined juice had better sensory and nutritional characters than when used individually and most recommended formulation was 90 percent lemon and 10 percent *Aloe vera* (Shailaja *et al*., 2018). The study on Orange - *Aloe vera* blended RTS revealed that the treatments were found to be suitable at 45 days of storage by the analysis of physicochemical, sensory attributes and microbiological analysis (Kausar *et al*., 2020). In the present experiment upto second month after storage no microbial population was noticed in all treatments. These studies supported the present study that the shelf life of aloe juice blended health drinks upto second month after storage without microbial load, but a slight increase in microbial count was noticed from third month after storage. However no *E. coli* population was noticed in the same till four month after storage. The products under present study were kept at room temperature. The products if stored under refrigeration might have increased shelf life. According to Hossain *et al*. (2017) jack fruit - *Aloe vera* blended ready to serve functional beverage was stored upto five months without notable change in chemical and seasonal parameters under refrigerated temperature. Variation in microbial count including fungal and bacterial population of aloe herbal health drink was presented in figure 23 and figure 24 respectively.

Table 86. Mean fungal population of aloe herbal health drink during storage, log cfu ml^{-1}

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	1.00 x 10^3 (3.00)	1.33 x 10^3 (3.10)
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	1.17 x 10^3 (3.05)	1.50 x 10^3 (3.15)
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	1.33 x 10^3 (3.10)	1.67 x 10^3 (3.20)
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	1.00 x 10^3 (3.00)	1.17 x 10^3 (3.05)
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	1.17 x 10^3 (3.05)	1.33 x 10^3 (3.10)
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	1.33 x 10^3 (3.10)	1.67 x 10^3 (3.20)
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.17 x 10^3 (0.50)	0.50 x 10^3 (1.50)
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.50 x 10^3 (1.50)	1.17 x 10^3 (3.05)
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.00 x 10^3 (0.00)	0.67 x 10^3 (2.00)	1.33 x 10^3 (3.10)
SEm (±)	0.00	0.00	0.00	0.35	0.23
CD (0.05)	NS	NS	NS	1.00	0.66

* Figures in parantheses indicate transformed values * Each value is the mean of six replication

Table 87. Mean bacterial population of aloe herbal health drink during storage, log cfu ml^{-1}

Treatments	0 MAS	1 MAS	2 MAS	3 MAS	4 MAS
T_1- Lemon juice + Liquidized aloe gel juice (50 : 50)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	1.17 x 10^6 (6.05)	1.33 x 10^6 (6.10)
T_2 - Lemon juice + Liquidized aloe gel juice (25 : 75)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	1.33 x 10^6 (6.10)	1.67 x 10^6 (6.18)
T_3 - Lemon juice + Liquidized aloe gel juice (10 : 90)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	1.50 x 10^6 (6.15)	1.83 x 10^6 (6.23)
T_4 - Orange juice + Liquidized aloe gel juice (50 : 50)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	1.17 x 10^6 (6.05)	1.17 x 10^6 (6.10)
T_5 - Orange juice + Liquidized aloe gel juice (25 : 75)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	1.33 x 10^6 (6.10)	1.83 x 10^6 (6.21)
T_6 - Orange juice + Liquidized aloe gel juice (10 : 90)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	1.50 x 10^6 (6.15)	2.00 x 10^6 (6.26)
T_{7-} Honey + Liquidized aloe gel juice (50 : 50)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.17 x 10^6 (0.00)	0.50 x 10^6 (2.05)
T_8 - Honey + Liquidized aloe gel juice (25 : 75)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.17 x 10^6 (1.00)	1.33 x 10^6 (6.10)
T_9 - Honey + Liquidized aloe gel juice (10 : 90)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.00 x 10^6 (0.00)	0.33 x 10^6 (2.00)	1.50 x 10^6 (6.15)
SEm (±)	0.000	0.000	0.000	0.54	0.43
CD (0.05)	NS	NS	NS	1.54	1.25

* Figures in parantheses indicate transformed values

* Each value is the mean of six replication

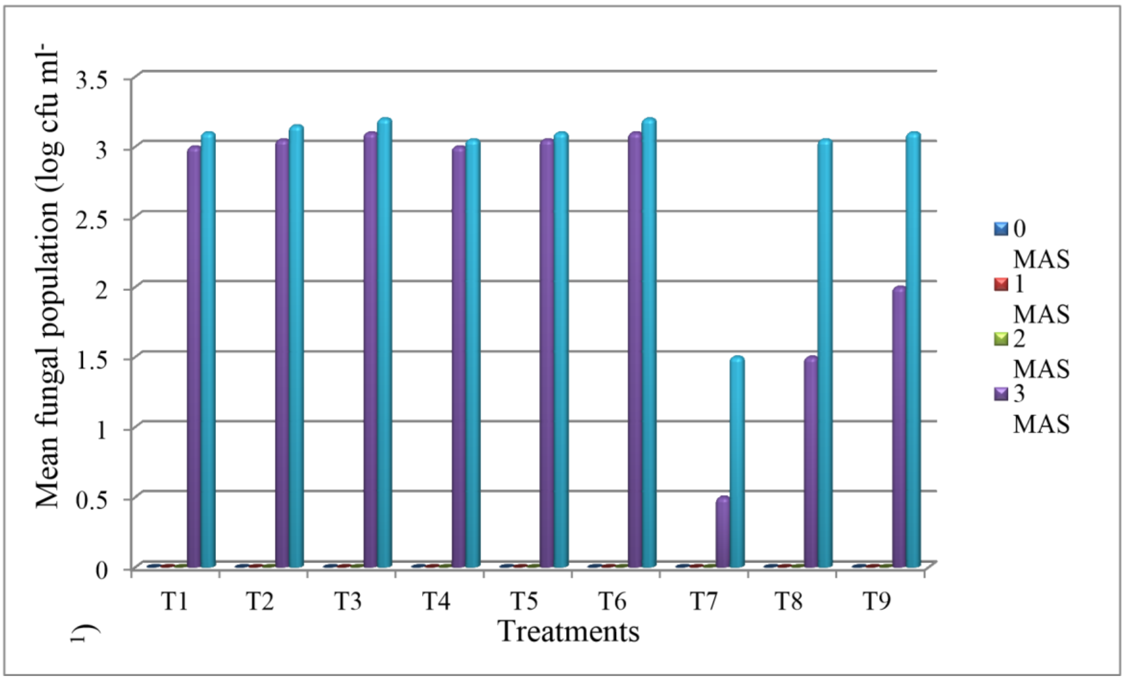


Figure 23. Mean fungal population of aloe herbal health drink during storage

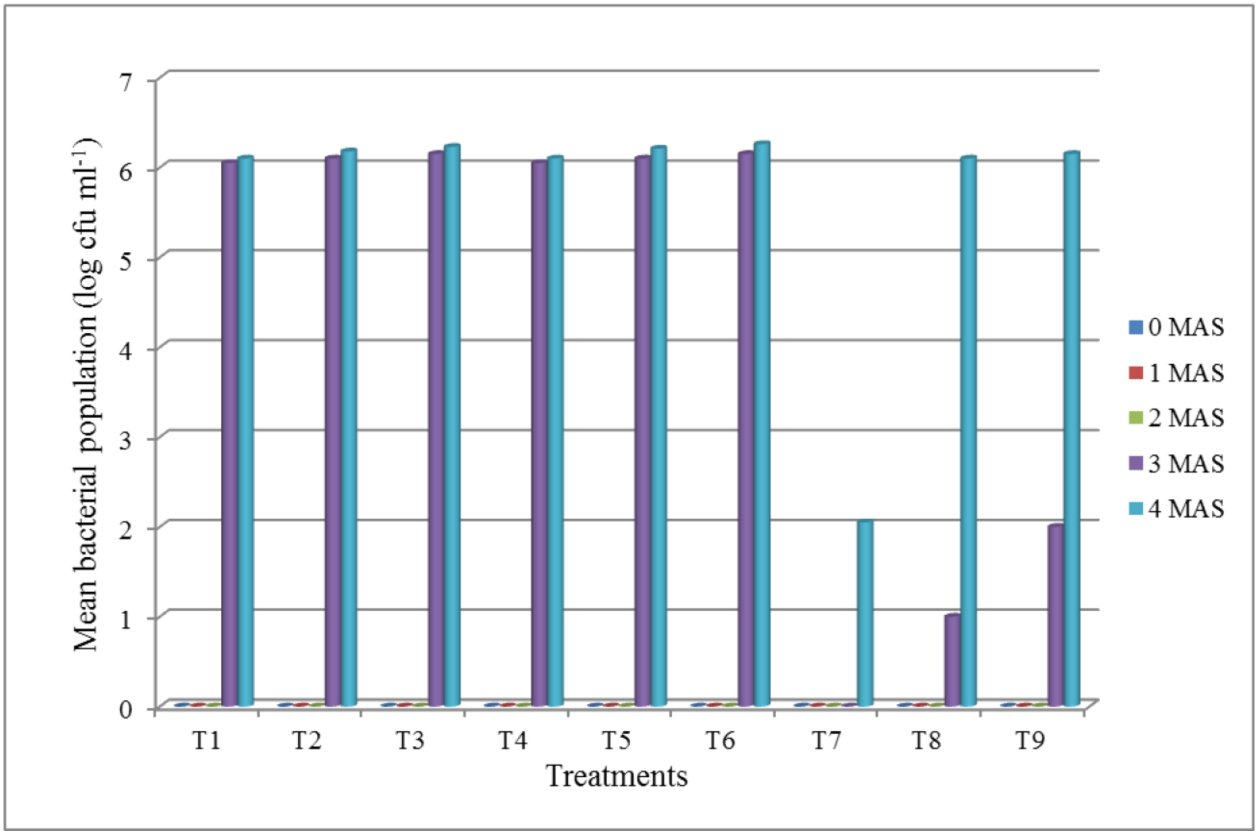


Figure 24. Mean bacterial population of aloe herbal health drink during storage

4.4.2 Aloe herbal powder

The preparation of aloe herbal powder by solar drying, air drying, oven drying or freeze drying resulted in a sticky product which could not be reconstituted with distilled water for quality comparison with fresh gel (Plate 14). Thus the herbal powder cannot be prepared in present study and it needs further investigation.

Aloe vera gel powder was produced through dehumidified air drying of Aloe vere gel at optimized conditions of temperature, relative humidity and air velocity of 64 °C, 18% and 0.8 $m.s^{-1}$, respectively. The powder was packed in laminated aluminum foil (AF), biaxially oriented polypropylene (BOPP) and polypropylene (PP). The storage stability of powder in terms of colour change revealed that AF was better than BOPP and PP (Ramachandra and Rao, 2013).

Plate 14A. Freeze dried aloe herbal powder

Plate 14B. Oven dried aloe herbal powder

Plate 14C. Sun dried aloe herbal powder

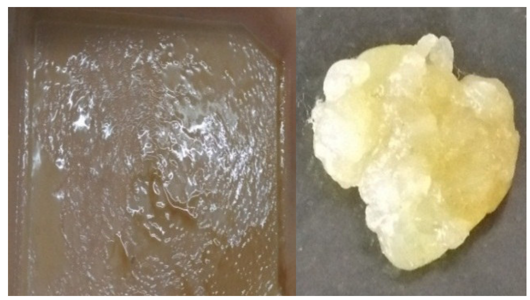


Plate 14D. Air dried aloe herbal powder

Plate 14. Aloe herbal powder under different drying methods

4.5 FUTURE LINE OF WORK

- Aloe gel stabilization with more natural herbal extracts under sterilized condition need to be standardised.
- Aloe gel preservation using various natural preservatives need to be explored and different packaging of the liquidized aloe gel juice need to be investigated.
- Natural product prepared using shade dried latex need to be standardized.
- Standardisation of appropriate low cost processing technique for the highly perishable commodity may encourage the innovative and progressive grower and entrepreneurs to take up aloe cultivation and manufacture nutraceuticals and cosmetic preparation thus making newer marketing products.

SUMMARY

5. SUMMARY

An experiment on "Protocol development for gel stabilization and nutraceuticals in *Aloe vera* (L.) Burm. f. was carried out at the Department of Plantation Crops and Spices, College of Agriculture, Vellayani during March 2016 to December 2019 with the objective of formulating a low cost stabilization technique for aloe gel using herbal extracts and aromatic oils and development of protocols for the preparation of dried latex and marketable nutraceuticals using aloe gel. The study was carried out as four experiments. The first experiment (Experiment I) studied on keeping quality and natural spoilage flora of fresh gel. The second experiment (Experiment II) consisted of standardization of curacao aloe (dried latex) preparation. The third experiment (Experiment III) was on gel standardization using herbal extracts/essential oils. The fourth experiment (Experiment IV) consisted of preparation of nutraceuticals from the stabilized liquidized aloe gel juice from experiment III. The salient findings obtained from four experiments are summarized below.

The first experiment on the keeping quality and natural spoilage flora of aloe gel were laid out in Completely Randomized Design. Keeping quality of liquidized aloe gel juice was studied by observing natural spoilage flora of fresh gel, color change, odour, pH, viscosity, refractive index and optical density for a week. The mean fungal population of the liquidized aloe gel juice samples did not show any significant variation during seven days of storage. The mean bacterial population on the first day of extraction was 6.5 log cfu ml^{-1} which increased to 8.5 log cfu ml^{-1} at the end of seventh day of extraction. *E. coli* was absent in the samples throughout the days of observation. The liquidized aloe gel juice showed off - white colour during first three days which got sedimented and became yellow in the subsequent days of storage. The odour of the liquidized aloe gel juice was mildly vegetative during the first three days and changed to foul smell from the fourth day. The pH of the liquidized aloe gel juice on the first day of extraction was 4.52 and then the pH increased significantly on the subsequent days reaching 4.85 on the seventh day of extraction (D_7). The viscosity of the liquidized aloe gel juice was the highest on the first day of extraction (2.59 cP) and then reduced significantly from the second day of extraction and was the minimum (1.36 cP) on the seventh day of extraction (D_7). No significant difference was noticed in the refractive index and optical density of liquidized aloe gel juice on storage for seven days. The liquidized aloe juice could not be stored for more than a day due to increased microbial population from the second day of storage and the subsequent change in sensory and physical characters. The liquidized gel pasteurized at 65 ^{0}C and 15 psi pressure for 13 minutes followed by flash cooling registered no microbial population even after seven days of storage.

The second experiment consisted of standardization of curacao aloe (dried latex) preparation. The latex collected from twenty five leaves constituting one replication were subjected to different methods of drying such as boiling followed by cooling (L_1), sun drying (L_2), shade drying (L_3) and oven drying (L_4) with five replications. The physical appearance and aloin content obtained by different drying methods were estimated and analysed statistically. The results showed that the colour as well as the appearance of dried latex ranked significantly high for shade drying (L_3). Aloin content of curacao aloe varied significantly between different methods of drying and the aloin content was highest for shade drying (L_3) (271.62 mg/ml) followed by oven drying (218.62 mg/ml), sun drying (159.96 mg/ml) and boiling followed by cooling (91.00 mg/ml).Thus among different methods of drying curacao aloe obtained by shade drying was the best.

The third experiment was on gel standardization using herbal extracts/essential oils. For the stabilization of liquidized aloe gel juice seven natural herbal extracts prepared in three forms of aqueous, tincture, decoction and five essential oils in different concentration were tried. The dosage for the final gel stabilization was fixed based on preliminary trials. The treatments taken for the preliminary trials of each of herbal extracts (aqueous, tincture and decoction) and essential oils were 1 ml, 1.25 ml, 1.50 ml, 1.75 ml and 2 ml respectively. Liquidized aloe gel juice with herbal extracts and essential oils were observed for sensory parameters, pH, refractive index, specific gravity, microbial populations and minimum inhibitory concentration. The final selection of the concentration of herbal extracts and essential oil were mainly based on microbial population and minimum inhibitory concentration.

The mean fungal, bacterial and *E. coli* population were absent in all the treatments for the first day of preparation of liquidized aloe gel juice added with 1 ml,1.25 ml,1.50 ml,1.75 ml and 2 ml of three forms of herbal extracts and essential oils. On 30th day of storage the *E. coli* population was absent in all forms of herbal extracts and essential oils.

The mean fungal population was non significant in *Gymnema sylvestre* aqueous extracts after 30th day of storage. On 30th day of storage mean bacterial population was significantly high and lowest bacterial population was observed in S_1 (6.40 log cfu ml^{-1}).The mean fungal population increased on storage and was insignificant among different concentration on the 30th day of storage in *Gymnema sylvestre* tincture extracts. The least mean bacterial population (6.06 log cfu ml^{-1}) was noticed from liquidized aloe gel juice added with 1ml of tincture extract of *Gymnema sylvestre* (S_6). The mean fungal and bacterial count was significantly different on 30th day of storage in *Gymnema sylvestre* decoction extracts. The mean fungal and bacterial population was found to be the lowest for S_{15}. In the case of minimum inhibitory concentration *Aspergillus* sp was found to have 100 per cent inhibition on the aqueous, tincture and decoction extracts of *Gymnema sylvestre* from the sample taken on first day of preparation and zero per cent inhibition on 30th day of storage. So among the aqueous and tincture extract 1 ml and in decoction 2 ml was taken for stabilization study.

On the 30th day of preparation the mean fungal and bacterial population increased and was significantly different among different concentrations of aqueous extracts of *Centella asiatica*. The mean fungal and bacterial population was the least for the liquidized aloe gel juice added with 1 ml of aqueous extract of *Centella asiatica* (S_{16}) on 30th day of storage. The mean fungal and bacterial population in the liquidized aloe gel juice was the least in $S_{25.}$ Among concentration of *Centella asiatica* decoction added to liquidized aloe gel juice, least fungal (3.49 log cfu ml^{-1}) and bacterial count (6.52 log cfu ml^{-1}) was observed in S_{26}. The minimum inhibitory concentration among all the treatments of aqueous extracts showed zero percentage inhibition on 30th day of observation. Application of 1 ml, 1.25 ml, 1.50 ml, 1.75 ml of tincture extracts showed zero per cent inhibition while 2ml of tincture extract had shown 100per cent inhibition during the 30th day of observation (S_{25}) and among the decoction all the treatments shown zero per cent inhibition on 30th day of storage. So among *Centella asiatica* extracts 1 ml of aqueous extracts, 2 ml of tincture extract and 1 ml of decoction extracts were taken for final stabilization study.

On the 30th day of preparation the mean fungal and bacterial population was significantly different among different concentrations of aqueous extracts of *Achyranthes aspera*. The mean fungal (3.00 log cfu ml^{-1}) and bacterial population (6.12 log cfu ml^{-1}) was the least for the liquidized aloe gel juice added with 2 ml of aqueous extract of *Achyranthes aspera* (S_{35}). The mean fungal population was the least for S_{39} (3.00 log cfu ml^{-1}) and S_{40} (3.00 log cfu ml^{-1}). Liquidized aloe gel juice added with 2 ml of *Achyranthes aspera* tincture extracts had the least bacterial population for S_{40} (6.00 log cfu ml^{-1}) which was on par with S_{39} (6.06 log cfu ml^{-1}) and S_{38} (6.12 log cfu ml^{-1}). *Achyranthes aspera* decoction at various concentration added to liquidized aloe gel juice produced significant variation in mean fungal and bacterial population on 30th day of preparation. The least bacterial (6.12 log cfu ml^{-1}) and fungal population was observed in S_{43} (3.12 log cfu ml^{-1}). In *Achyranthes aspera* aqueous extracts, the minimum inhibitory concentration showed zero per cent inhibition in treatments S_{31} and S_{32} on 30th day of preparation, 11.11 per cent inhibition by the treatment S_{33} and $S_{34,}$ while S_{35} has shown 22.22 per cent inhibition. The percentage inhibition of *Aspergillus* sp by liquidized aloe gel juice with different concentration of tincture extracts of *Achyranthes aspera* was zero on 30th day of storage. The minimum inhibitory concentration of liquidized aloe gel juice with 1.50 ml of *Achyranthes aspera* decoction extracts stored at 30 days showed 11.11 per cent inhibition of *Aspergillus* sp. Among the *Achyranthes aspera* extracts 2 ml aqueous extracts, 2 ml tincture extracts and 1.50 ml of decoction extracts were selected.

Liquidized aloe gel juice with *Tridax procumbens* aqueous extracts had least fungal population in S_{50} (3.66 log cfu ml^{-1}) with 2ml and the lowest bacterial population was shown by S_{49} (6.06 log cfu ml^{-1}) with 1.75 ml and S_{50} (6.06 x 10^6 log cfu ml^{-1}).The mean fungal population was non significant among different concentration of the tincture extracts of *Tridax procumbens* on the 30th day of storage. No bacterial population was noticed from liquidized aloe gel juice added with 2 ml of tincture extract of *Tridax procumbens* (S_{55}) even after 30 days of storage and was on par with S_{54} (1.20 log cfu ml^{-1}). The mean fungal population was found to be the lowest for S_{56} and S_{57} as 3.18 log cfu ml^{-1} which was on par with S_{58} (3.30 log cfu ml^{-1}) and the mean bacterial population was also the least for S_{56} (6.12 log cfu ml^{-1}) which was on par with S_{57} (6.28 log cfu ml^{-1}) in liquidized aloe gel juice with *Tridax procumbens* decoction. Among *Tridax procumbens* extracts, aqueous 2 ml, tincture 2 ml and decoction 1 ml was selected for final gel stabilization studies based on minimum inhibitory concentration studies which showed maximum inhibitory effects at these concentrations.

The mean fungal and bacterial population was the least for the liquidized aloe gel juice added with 1 ml of aqueous and decoction extract of *Terminalia chebula* (S_{61}). Liquidized aloe gel juice added with 2ml of *Terminalia chebula* tincture extracts had the least bacterial population. All the treatments had shown 100 per cent inhibition of *Aspergillus* sp on 1st day of preparation and 30th day of storage. In *Terminalia chebula* extracts aqueous 1 ml, tincture 1 ml and decoction 1 ml were selected for final gel stabilization studies.

The mean fungal and bacterial population on 30th day of storage was least in liquidized aloe gel juice added with 1 ml of aqueous and decoction and 2 ml of tincture extract of *Punica granatum.* The minimum inhibitory concentration studies of liquidized aloe gel juice added with aqueous, tincture and decoction extracts from the rind of *Punica granatum* on *Aspergillus* sp showed 100 per cent inhibition on the 1st day of preparation for all the treatments, but zero percentage inhibition was noticed on all treatments on 30th day of storage. So based on microbial count from *Punica granatum* extracts 1 ml aqueous extract, 2 ml tincture extracts and 1 ml decoction extract were selected for stabilization studies.

On the 30th day of storage of liquidized aloe gel juice with green tea aqueous extracts, the least mean fungal (3.06 log cfu ml^{-1}) and bacterial population(6.55 log cfu ml^{-1}) was observed with 2 ml of aqueous extract of green tea. Liquidized aloe gel juice added with 1 ml of green tea tincture extracts and 2 ml of decoction had the least bacterial population and fungal population. The minimum inhibitory concentration of liquidized aloe gel juice added with aqueous, tincture and decoction extracts of green tea showed zero percentage inhibition on 30th day of observation for all treatments except the treatment S_{95} which exhibited 66.67 per cent inhibition on *Aspergillus* sp. Among green tea extracts 2 ml aqueous extract, 1 ml tincture extract and 2 ml decoction extract were taken for further stabilization studies.

Microbial population of the liquidized aloe gel juice added with different concentration (1 ml, 1.25 ml, 1.50 ml,1.75 ml, and 2 ml)of essential oil of basil oil, lemon grass oil, cinnamon grass oil, clove oil and cardamom oil did not show any microbial count after 30 days of storage. In the case of minimum inhibitory concentration all the essential oils of concentration 1 ml, 1.25 ml, 1.50 ml, 1.75 ml and 2 ml at 30th day of storage showed 100 per cent inhibition on *Aspergillus* sp. Hence the minimum concentration of each of the essential oil was taken for the further study. The best concentrations from aqueous, tincture and decoction of herbal extracts and essential oil (1 ml) from basil oil, lemon grass oil, cinnamon bark oil, clove oil and cardamom oil and sodium benzoate (1 ml) as control were selected from the preliminary trials of herbal extracts and essential oils were subjected to final stabilization treatments and stored in room temperature for six months for recording observations on the storage life, the result of which are summarized below.

In the case of colour as revealed by the highest mean rank value among the treatments was noticed in1ml clove oil, G_{25} (592.90). The mean rank value of odour observed higher among essential oil treatments and this was significantly higher in liquidized aloe gel juice added with 1ml clove oil, G_{25} (593.70). Based on appearance, color and odour six month after storage highest mean rank value were noticed in liquidized aloe gel juice added with 1 ml clove oil.

Refractive index of liquidized aloe gel juice added with different concentration of aqueous, tincture and decoction of herbal extracts, essential oils (1 ml) of basil oil, lemon grass oil, cinnamon bark oil, clove oil and cardamom oil as well as sodium benzoate 1 ml did not show any significant variation from first day of storage upto six month after storage. In the present study specific gravity ranged from 1.000 to 1.014 upto six months of storage. Specific gravity increased during storage in all the treatments. After six months of storage least specific gravity was noticed in G_{25} (1.004), clove oil 1 ml which was on par with G_{24} (1.005), cinnamon bark oil 1 ml. pH of liquidized aloe gel juice added with herbal extracts and essential oil, maintained 3.50 at zero month increased during second month after storage and thereafter pH value decreased. On sixth month after storage, significantly high pH value among the treatments was shown by G_1 (5.30), *Gymnema sylvestre* aqueous 1 ml. The pH of the liquidized aloe gel juice added with essential oil varied from 2.82 in cardamom oil (G_{26}) to 3.49 in clove oil (G_{25}).The percentage of total solids was significantly higher in $G_{25,}$ clove oil 1ml (0.55 per cent) followed by 0.47 per cent in G_{23} (lemon grass oil 1 ml) at six months of storage. The monosaccharide content of the liquidized aloe gel juice added with 1 ml of clove oil showed a value of 0.04 per cent from zero MAS to 6 MAS, without showing any change while for the treatment with 1 ml cinnamon bark oil the monosaccharide content was the same from zero MAS to 5 MAS. At six month after storage high polysaccharide content of 0.25 per cent was observed from G_{22} (basil oil 1 ml), G_{23} (lemon grass oil 1 ml) and G_{24} (cinnamon bark oil 1 ml) which was followed by G_{25} (clove oil 1 ml) with a value of 0.24 per cent. The polysaccharide content of treatments with herbal extracts varied from 0.01 to 0.20 per cent while that of essential oils varied from 0.22 per cent to 0.25 per cent.

A general decrease in amino acid content of the liquidized aloe gel juice added with herbal extracts was noticed on storage. However the addition of clove oil, 1 ml the amino acid content did not change over six months of storage. Vitamin A and C content of liquidized aloe gel juice added with herbal extracts and essential oils decreased on storage. The amylase activity among treatments with essential oils ranged from 1338.80 in G_{24} (cinnamon bark oil1 ml) to 1491.20 in G_{25} (clove oil 1ml) and that for control G_{27} was 1135.40 unit per 100 ml at six month of storage. Lipase activity also reduced among the treatments on storage however the decrease was less for clove oil treated sample. In the case of protein content at sixth month of storage, protein content of 0.23 per cent was observed from G_{25} (clove oil 1 ml) and G_{26} (cardamom oil 1 ml). Higher ash content was noticed in G_{23} (lemon grass oil 1 ml) and G_{25} (clove oil 1 ml) with 0.89 per cent at zero MAS which reduced to 0.73 per cent in six month after storage G_{25} (clove oil 1ml). Significantly higher calorie content was noticed in G_{22} (basil oil 1ml) with 2.88 kcal/100 g was zero MAS while at six month of storage calorie content was highest in G_{23} (lemon grass oil 1ml) with 2.44 kcal/100 g. The fat content did not show any significant difference among the samples from 2 MAS to 6 MAS. The crude fibre content of all the treatments reduced on storage. Absence of fungal population was noticed in samples stabilized with clove oil, 1 ml from 0 MAS to 6 MAS. In the case of bacterial population during six month after storage least value was noticed in G_{25}, clove oil of 1 ml (2.40 log cfu ml^{-1}). Finally from gel stabilization treatments it was concluded that liquidized aloe gel juice stabilized with 1 ml of clove oil was best for the preparation of neutraceuticals.

The stabilized liquidized aloe gel juice with 1 ml clove oil was used for the preparation of nutraceuticals such as aloe herbal health drink and aloe herbal powder. Aloe herbal health drink was prepared by adding lemon juice, orange juice and honey to the stabilized liquidized aloe gel juice in the proportions 50 : 50, 25 : 75 and 10 : 90 followed by pasteurization and flash cooling and were stored in 30 ml glass bottles. Sensory/organoleptic parameters, chemical/nutrient composition and shelf life studies were carried out on these nutraceuticals prepared.

The highest mean rank for appearance and color among the treatments were scored by T_1 - Lemon juice + Liquidized aloe gel juice (50:50) at fourth month of storage which recorded high mean rank value of 158.30 for appearance and 151.20 for colour. The highest mean rank value for taste, odour and overall acceptability was recorded in T_7 -Honey + Liquidized aloe gel juice (50 : 50) from zero MAS to 4 MAS.

The highest acidity among the treatments was noticed inT_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) during zero MAS to 4 MAS and it increased on storage. The pH content was highest in T_7 - Honey + Liquidized aloe gel juice (50: 50) which changed from 4.10 to 4.08 from zero MAS to 4 MAS. Higher TSS content during zero and fourth month after storage was noticed in T_7- Honey + Liquidized aloe gel juice (50:50) recording 51.67 ^{0}Brix and 44.83 ^{0}Brix respectively. The vitamin C content decreased from zero month after storage to fourth month after storage and was highest in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50). Vitamin C content of T_1 ranged from 45.56 mg/100ml to 40.56 mg/100ml from zero MAS to 4 MAS. The energy content was highest in T_7 - Honey + Liquidized aloe gel juice (50 : 50) with 11.17 kcal/100mg in zero MAS which reduced to 10.61 kcal/100mg in 4 MAS. The highest iron content among the treatments was noticed in T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) with 0.08 per cent at zero MAS to 0.03 per cent at 4 MAS. The highest calcium content of 0.008 per cent among the treatments was noticed in T_7 - Honey + Liquidized aloe gel juice (50 : 50), but from second month of storage onwards the calcium content were found to be non significant for all the treatments. The carbohydrate content was the highest in T_7 - Honey + liquidized aloe gel juice (50 : 50) which recorded 2.62 per cent at 4 MAS. The highest fat content was noticed in T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) and T_4 - Orange juice + Liquidized aloe gel juice (50 : 50) having fat content of about 0.002 per cent and became non significant during 3 MAS and 4 MAS. T_1 - Lemon juice + Liquidized aloe gel juice (50 : 50) recorded maximum protein content of 0.11 per cent in zero MAS and 0.07 per cent in T_1 during 4 MAS. Shelf life of aloe – herbal health drink was determined by considering microbial population and sensory parameters like color, odour, taste, appearance andoverall acceptability. The fungal and bacterial population were absent up to second month after storage and *E. coli* population was absent during the storage period of upto fourth month.T_7 - Honey + Liquidized aloe gel juice (50:50) recorded least fungal population of 0.50 log cfu ml^{-1} during third month after storage and a population of 1.50 log cfu ml^{-1} in fourth month after storage. T_7 – Honey + Liquidized aloe gel juice (50:50) recorded no bacterial population at third month after storage and a least bacterial population of 2.05 log cfu ml^{-1} was observed during fourth month after storage. Hence it was concluded that aloe juice blended with honey (50 : 50) can be stored up to three months after storage with less microbial population.

The preparation of aloe herbal powder by solar drying, air drying, oven drying or freeze drying resulted in a sticky product which could not be reconstituted with distilled water for quality comparison with fresh gel. Thus the herbal powder cannot be prepared in present study and it needs further investigation.

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6. REFERENCES

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PROTOCOL DEVELOPMENT FOR GEL STABILIZATION AND NUTRACEUTICALS IN *Aloe vera* (L.) Burm. f.

by

MAHESWARI R. S. NAIR

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ABSTRACT

Submitted in partial fulfilment of the requirements for the degree of

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Faculty of Agriculture

Kerala Agricultural University

DEPARTMENT OF PLANTATION CROPS AND SPICES

COLLEGE OF AGRICULTURE

VELLAYANI, THIRUVANANTHAPURAM - 695 522

KERALA, INDIA

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ABSTRACT

The investigation entitled “Protocol development for gel stabilization and nutraceuticals in *Aloe vera* (L.) Burm. f.” was carried out in the Department of Plantation Crops and Spices, College of Agriculture, Vellayani during March 2016 to December 2019. The project envisaged formulation of a low cost stabilization technique for aloe gel using herbal extracts and aromatic oils and development of protocols for the preparation of dried latex and marketable nutraceuticals using aloe gel. The study was carried out as four experiments. The first experiment was to study the keeping quality and natural spoilage flora of fresh gel while the second experiment was for the standardization of curacao aloe (dried latex). The third experiment was on gel stabilization using herbal extracts and essential oils. Preparation of nutraceuticals from the stabilized liquidized aloe gel juice was the final experiment.

The keeping quality and natural spoilage flora of fresh gel were assessed by subjecting the liquidized aloe gel juice to storage in glass bottles under ambient condition for seven days. The liqudized aloe gel juice was off white in colour for first three days of extraction with mild vegetative odour and got sedimented with foul smell from fourth day onwards. The liquidized aloe juice could not be stored for more than a day due to increased microbial population from the second day of storage. Preliminary trails conducted by pasteurizing the liquidized aloe gel juice at 65 ^{0}C and 15 psi pressure for 13 minutes followed by flash cooling registered no microbial population even after seven days of storage.

The latex collected from aloe leaves was subjected to different methods of drying such as boiling followed by cooling, sun drying, shade drying and oven drying. Appearance, colour and aloin content (271.62 mg/ml) of dried latex was significantly higher for shade drying.

Liquidized aloe gel juice was pasteurized and added with varying concentrations of three forms (aqueous, tincture, decoction) of herbal extracts and

essential oils after adjusting the pH to 3.5 by adding 0.5 per cent of citric acid for gel stabilization. The treated samples were kept for a month and based on microbial population and minimum inhibitory concentration best treatment of each form was selected from preliminary trials for aloe gel stabilization. *Gymnema sylvestre* aqueous extract (1 ml), tincture (1 ml), decoction (2 ml), *Centella asiatica* aqueous extract (1 ml), tincture (2 ml), decoction (1 ml), *Achyranthes aspera* aqueous extract (2 ml), tincture (2 ml), decoction (1.50 ml), *Tridax procumbens* aqueous extract (2 ml), tincture (2 ml) , decoction (1 ml), *Terminalia chebula* aqueous extract (1 ml), tincture (1 ml), decoction (1 ml), *Punica granatum* aqueous extract (1 ml), tincture (2 ml), decoction (1 ml), green tea aqueous extract (2 ml), tincture (1 ml) and decotion (2 ml) and 1 ml each of sacred basil oil, lemon grass oil, cinnamon bark oil, clove oil and cardamom oil were selected and added to pH adjusted , pasteurized and liquidized aloe gel juice (25 ml) for gel stabilization. The gel stabilization was thus done using the selected twenty six treatments in a Completely Randomised Design replicated five times and compared with 0.08 per cent sodium benzoate as control and stored for six months. Appearance, colour and odour of all forms of the herbal extracts reduced on storage while those treatments with aromatic oils showed lesser percentage reduction in these parameters. Total solids, amylase and lipase activity decreased on storage. The amino acid content was the highest for liquidized aloe gel juice added with aqueous, tincture and decoction of *Achyranthes aspera* (0.08 ppm).Vitamin A and C were highest for treatment with green tea leaf aqueous extract which decreased subsequently on storage. An increase in microbial load was observed for all the treatments with herbal extracts from first month of storage. But addition of 1 ml clove oil resulted in stabilization of liquidized aloe gel juice which could be stored upto five months without microbial contamination or affecting the nutritive and sensory parameters.

Nutraceuticals were prepared using stabilized liquidized aloe gel juice containing clove oil blended with lemon juice, orange juice and honey in proportions of 50 : 50, 75 : 25 and 90 : 10 followed by pasteurization, flash cooling and stored for 6 months. Appearance, colour and vitamin C were significantly higher for Lemon juice (50 ml) + Liquidized aloe gel juice (50 ml) + 2 ml clove oil while odour, taste, overall acceptability, pH, TSS, carbohydrates and calories were significantly superior for Honey (50 ml) + Liquidized aloe gel juice (50 ml) + 2 ml clove oil. Growth of microbes could be detected from third month of storage for all the treatments. Aloe health drink with honey in the ratio 50 : 50 added with clove oil were selected as the accepted drink which could be preserved for two months without microbial contamination.

The preparation of aloe herbal powder by solar drying, air drying, oven drying or freeze drying resulted in a sticky product which could not be reconstituted with distilled water for quality comparison with fresh gel, thus warranting further investigation.

The present study revealed that liquidized aloe gel juice pasteurized and mixed with clove oil (4 per cent) is a low cost stabilization method which can be taken as a base material for the preparation of health drink. The nutraceutical with liquidized and stabilized aloe gel juice mixed with equal proportion of honey and preserved with clove oil is a palatable drink having higher calories which could be stored for two months. The dried aloe latex a byproduct produced by shade drying is superior with high aloin content and can also be used for the development of marketable product.

APPENDICES

APPENDIX I.

MEDIA COMPOSITION FOR MICROBIAL STUDY

I.A. Nutrient Agar Medium (pH – 7.0)

SL No.	Chemicals	Quantity Required
1	Peptone	5 g
2	Sodium	5 g
3	Beef extract	3 g
4	Agar	20 g
5	Distilled water	1000

I.B. EMB Agar Medium (pH – 7.0)

Sl. No.	Chemicals	Quantity Required
1	Peptic digest of animal tissue	10 g/litre
2	Dipotassium phosphate	2 g/litre
3	Lactose	5 g/litre
4	Sucrose	5 g/litre
5	Eosin – Y	0.40 g/litre
6	Methylene blue	0.065 g/litre
7	Agar	13.50 g/litre

NB: Suspend 35.96 grams of EMB agar medium in 1000 ml distilled water

I.C. Martin`s Rose Bengal Agar Medium (pH – 7.0)

Sl. No.	Chemicals	Quantity Required
1.	Glucose	10 g
2.	Peptone	5 g
3.	Potassium dihydrogen Phosphate	1 g
4.	Magnesium Sulphate heptahydrate	0.5 g
5.	Streptomycin	30 mg
6.	Agar	15 g
7.	Rose Bengal solution	1 ml of 3.5% solution
8.	Distilled water	1000 ml

APPENDIX II

SCORE CARD FOR ASSESSING THE PHYSICAL APPEARANCE OF CURACAO ALOE (DRIED LATEX)

COLLEGE OF AGRICULTURE, VELLAYANI

Dept. of Plantation Crops and Spices

Title: Protocol development for gel stabilization and nutraceuticals in *Aloe vera* (L.) Burm. f.

Sample: Curacao aloe (dried latex)

Instructions: You are given 4 samples of curacao aloe (dried latex). Evaluate them and give scores for each.

Criteria	Sample				Description
	232	327	252	222	
Colour					Whether different drying method affect the physical appearance of curacao aloe (dried latex)
Appearance					

<u>Score</u>

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

Date :

Name :

Signature :

APPENDIX III

SCORE CARD FOR ASSESSING THE SENSORY PARAMETERS OF TREATED LIQUIDIZED ALOE GEL JUICE WITH DIFFERENT CONCENTRATIONS OF HERBAL EXTRACTS/ESSENTIAL OILS

COLLEGE OF AGRICULTURE, VELLAYANI

Dept. of Plantation Crops and Spices

Title: Protocol development for gel stabilization and nutraceuticals in *Aloe vera* (L.) Burm. f.

Sample: Aloe juice

Instructions: You are given 10 samples of aloe juice. Evaluate them and give scores for each

Criteria	Sample										Description
	232	327	252	222	132	142	213	352	244	125	Whether the additives had affected the natural colour, odour/flavour appearance of aloe gel
Colour											
Odour/ Flavour											
Appearance											

<u>Score</u>

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

Date :

Name :

Signature :

APPENDIX IV
SCORE CARD FOR ASSESSING THE SENSORY PARAMETERS OF ALOE HERBAL HEALTH DRINK
COLLEGE OF AGRICULTURE, VELLAYANI
Dept. of Plantation Crops and Spices

Title: Protocol development for gel stabilization and nutraceuticals in *Aloe vera* Burm. f.

Sample: Aloe herbal health drinks

Instructions: You are given 9 samples of aloe herbal health drinks. Evaluate them and give scores for each

Criteria	Sample									Description
	232	327	252	222	132	142	213	352	244	Whether the additives had affected the natural colour, odour/flavour appearance, taste and overall acceptability of aloe herbal health drink
Colour										
Odour/ Flavour										
Appearance										
Taste										
Overall acceptability										

Score

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

Date :

Name :

Signature :

APPENDIX V

TREATMENT COMBINATION FOR PRELIMINARY STUDY

S_1	Liquidized aloe gel juice + *Gymnema sylvestre* aqueous extracts 1ml
S_2	Liquidized aloe gel juice + *Gymnema sylvestre* aqueous extracts 1.25ml
S_3	Liquidized aloe gel juice + *Gymnema sylvestre* aqueous extracts 1.50ml
S_4	Liquidized aloe gel juice + *Gymnema sylvestre* aqueous extracts 1.75ml
S_5	Liquidized aloe gel juice + *Gymnema sylvestre* aqueous extracts 2ml
S_6	Liquidized aloe gel juice + *Gymnema sylvestre* tincture extracts 1ml
S_7	Liquidized aloe gel juice + *Gymnema sylvestre* tincture extracts 1.25ml
S_8	Liquidized aloe gel juice + *Gymnema sylvestre* tincture extracts 1.50ml
S_9	Liquidized aloe gel juice + *Gymnema sylvestre* tincture extracts 1.75ml
S_{10}	Liquidized aloe gel juice + *Gymnema sylvestre* tincture extracts 2ml
S_{11}	Liquidized aloe gel juice + *Gymnema sylvestre* decoction extracts 1ml
S_{12}	Liquidized aloe gel juice + *Gymnema sylvestre* decoction extracts 1.25ml
S_{13}	Liquidized aloe gel juice + *Gymnema sylvestre* decoction extracts 1.50ml
S_{14}	Liquidized aloe gel juice + *Gymnema sylvestre* decoction extracts 1.75ml
S_{15}	Liquidized aloe gel juice + *Gymnema sylvestre* decoction extracts 2ml
S_{16}	Liquidized aloe gel juice + *Centella asiatica* aqueous extracts 1ml
S_{17}	Liquidized aloe gel juice + *Centella asiatica* aqueous extracts 1.25ml
S_{18}	Liquidized aloe gel juice + *Centella asiatica* aqueous extracts 1.50ml
S_{19}	Liquidized aloe gel juice + *Centella asiatica* aqueous extracts 1.75ml
S_{20}	Liquidized aloe gel juice + *Centella asiatica* aqueous extracts 2ml
S_{21}	Liquidized aloe gel juice + *Centella asiatica* tincture extracts 1ml
S_{22}	Liquidized aloe gel juice + *Centella asiatica* tincture extracts 1.25ml
S_{23}	Liquidized aloe gel juice + *Centella asiatica* tincture extracts 1.50ml
S_{24}	Liquidized aloe gel juice + *Centella asiatica* tincture extracts 1.75ml

S_{25}	Liquidized aloe gel juice + *Centella asiatica* tincture extracts 2ml
S_{26}	Liquidized aloe gel juice + *Centella asiatica* decoction extracts 1ml
S_{27}	Liquidized aloe gel juice + *Centella asiatica* decoction extracts 1.25ml
S_{28}	Liquidized aloe gel juice + *Centella asiatica* decoction extracts 1.50ml
S_{29}	Liquidized aloe gel juice + *Centella asiatica* decoction extracts 1.75ml
S_{30}	Liquidized aloe gel juice + *Centella asiatica* decoction extracts 2ml
S_{31}	Liquidized aloe gel juice + *Achyranthes aspera* aqueous extracts 1ml
S_{32}	Liquidized aloe gel juice + *Achyranthes aspera* aqueous extracts 1.25ml
S_{33}	Liquidized aloe gel juice + *Achyranthes aspera* aqueous extracts 1.50ml
S_{34}	Liquidized aloe gel juice + *Achyranthes aspera* aqueous extracts 1.75ml
S_{35}	Liquidized aloe gel juice + *Achyranthes aspera* aqueous extracts 2ml
S_{36}	Liquidized aloe gel juice + *Achyranthes aspera* tincture extracts 1ml
S_{37}	Liquidized aloe gel juice + *Achyranthes aspera* tincture extracts 1.25ml
S_{38}	Liquidized aloe gel juice + *Achyranthes aspera* tincture extracts 1.50ml
S_{39}	Liquidized aloe gel juice + *Achyranthes aspera* tincture extracts 1.75ml
S_{40}	Liquidized aloe gel juice + *Achyranthes aspera* tincture extracts 2ml
S_{41}	Liquidized aloe gel juice + *Achyranthes aspera* decoction extracts 1ml
S_{42}	Liquidized aloe gel juice + *Achyranthes aspera* decoction extracts 1.25ml
S_{43}	Liquidized aloe gel juice + *Achyranthes aspera* decoction extracts 1.50ml
S_{44}	Liquidized aloe gel juice + *Achyranthes aspera* decoction extracts 1.75ml
S_{45}	Liquidized aloe gel juice + *Achyranthes aspera* decoction extracts 2ml
S_{46}	Liquidized aloe gel juice + *Tridax procumbens* aqueous extracts 1ml
S_{47}	Liquidized aloe gel juice + *Tridax procumbens* aqueous extracts 1.25ml
S_{48}	Liquidized aloe gel juice + *Tridax procumbens* aqueous extracts 1.50ml
S_{49}	Liquidized aloe gel juice + *Tridax procumbens* aqueous extracts 1.75ml
S_{50}	Liquidized aloe gel juice + *Tridax procumbens* aqueous extracts 2ml
S_{51}	Liquidized aloe gel juice + *Tridax procumbens* tincture extracts 1ml

S_{52}	Liquidized aloe gel juice + *Tridax procumbens* tincture extracts 1.25ml
S_{53}	Liquidized aloe gel juice + *Tridax procumbens* tincture extracts 1.50ml
S_{54}	Liquidized aloe gel juice + *Tridax procumbens* tincture extracts 1.75ml
S_{55}	Liquidized aloe gel juice + *Tridax procumbens* tincture extracts 2ml
S_{56}	Liquidized aloe gel juice + *Tridax procumbens* decoction extracts 1ml
S_{57}	Liquidized aloe gel juice + *Tridax procumbens* decoction extracts 1.25ml
S_{58}	Liquidized aloe gel juice + *Tridax procumbens* decoction extracts 1.50ml
S_{59}	Liquidized aloe gel juice + *Tridax procumbens* decoction extracts 1.75ml
S_{60}	Liquidized aloe gel juice + *Tridax procumbens* decoction extracts 2ml
S_{61}	Liquidized aloe gel juice + *Terminalia chebula* aqueous extracts 1ml
S_{62}	Liquidized aloe gel juice + *Terminalia chebula* aqueous extracts 1.25ml
S_{63}	Liquidized aloe gel juice + *Terminalia chebula* aqueous extracts 1.50ml
S_{64}	Liquidized aloe gel juice + *Terminalia chebula* aqueous extracts 1.75ml
S_{65}	Liquidized aloe gel juice + *Terminalia chebula* aqueous extracts 2ml
S_{66}	Liquidized aloe gel juice + *Terminalia chebula* tincture extracts 1ml
S_{67}	Liquidized aloe gel juice + *Terminalia chebula* tincture extracts 1.25ml
S_{68}	Liquidized aloe gel juice + *Terminalia chebula* tincture extracts 1.50ml
S_{69}	Liquidized aloe gel juice + *Terminalia chebula* tincture extracts 1.75ml
S_{70}	Liquidized aloe gel juice + *Terminalia chebula* tincture extracts 2ml
S_{71}	Liquidized aloe gel juice + *Terminalia chebula* decoction extracts 1ml
S_{72}	Liquidized aloe gel juice + *Terminalia chebula* decoction extracts 1.25ml
S_{73}	Liquidized aloe gel juice + *Terminalia chebula* decoction extracts 1.50ml
S_{74}	Liquidized aloe gel juice + *Terminalia chebula* decoction extracts 1.75ml
S_{75}	Liquidized aloe gel juice + *Terminalia chebula* decoction extracts 2ml
S_{76}	Liquidized aloe gel juice + *Punica granatum* aqueous extracts 1ml
S_{77}	Liquidized aloe gel juice + *Punica granatum* aqueous extracts 1.25ml
S_{78}	Liquidized aloe gel juice + *Punica granatum* aqueous extracts 1.50ml

S_{79}	Liquidized aloe gel juice + *Punica granatum* aqueous extracts 1.75ml
S_{80}	Liquidized aloe gel juice + *Punica granatum* aqueous extracts 2ml
S_{81}	Liquidized aloe gel juice + *Punica granatum* tincture extracts 1ml
S_{82}	Liquidized aloe gel juice + *Punica granatum* tincture extracts 1.25ml
S_{83}	Liquidized aloe gel juice + *Punica granatum* tincture extracts 1.50ml
S_{84}	Liquidized aloe gel juice + *Punica granatum* tincture extracts 1.75ml
S_{85}	Liquidized aloe gel juice + *Punica granatum* tincture extracts 2ml
S_{86}	Liquidized aloe gel juice + *Punica granatum* decoction extracts 1ml
S_{87}	Liquidized aloe gel juice + *Punica granatum* decoction extracts 1.25ml
S_{88}	Liquidized aloe gel juice + *Punica granatum* decoction extracts 1.50ml
S_{89}	Liquidized aloe gel juice + *Punica granatum* decoction extracts 1.75ml
S_{90}	Liquidized aloe gel juice + *Punica granatum* decoction extracts 2ml
S_{91}	Liquidized aloe gel juice + green tea aqueous extracts 1ml
S_{92}	Liquidized aloe gel juice + green tea aqueous extracts 1.25ml
S_{93}	Liquidized aloe gel juice + green tea aqueous extracts 1.50ml
S_{94}	Liquidized aloe gel juice + green tea aqueous extracts 1.75ml
S_{95}	Liquidized aloe gel juice + green tea aqueous extracts 2ml
S_{96}	Liquidized aloe gel juice + green tea tincture extracts 1ml
S_{97}	Liquidized aloe gel juice + green tea tincture extracts 1.25ml
S_{98}	Liquidized aloe gel juice + green tea tincture extracts 1.50ml
S_{99}	Liquidized aloe gel juice + green tea tincture extracts 1.75ml
S_{100}	Liquidized aloe gel juice + green tea tincture extracts 2ml
S_{101}	Liquidized aloe gel juice + green tea decoction extracts 1ml
S_{102}	Liquidized aloe gel juice + green tea decoction extracts 1.25ml
S_{103}	Liquidized aloe gel juice + green tea decoction extracts 1.50ml
S_{104}	Liquidized aloe gel juice + green tea decoction extracts 1.75ml
S_{105}	Liquidized aloe gel juice + green tea decoction extracts 2ml

S_{106}	Liquidized aloe gel juice + sacred basil oil 1ml
S_{107}	Liquidized aloe gel juice + sacred basil oil 1.25ml
S_{108}	Liquidized aloe gel juice + sacred basil oil 1.50ml
S_{109}	Liquidized aloe gel juice + sacred basil oil 1.75ml
S_{110}	Liquidized aloe gel juice + sacred basil oil 2ml
S_{111}	Liquidized aloe gel juice + lemon grass oil 1ml
S_{112}	Liquidized aloe gel juice + lemon grass oil 1.25ml
S_{113}	Liquidized aloe gel juice + lemon grass oil 1.50ml
S_{114}	Liquidized aloe gel juice + lemon grass oil 1.75ml
S_{115}	Liquidized aloe gel juice + lemon grass oil 2ml
S_{116}	Liquidized aloe gel juice + cinnamon bark oil 1ml
S_{117}	Liquidized aloe gel juice + cinnamon bark oil 1.25ml
S_{118}	Liquidized aloe gel juice + cinnamon bark oil 1.50ml
S_{119}	Liquidized aloe gel juice + cinnamon bark oil 1.75ml
S_{120}	Liquidized aloe gel juice + cinnamon bark oil 2ml
S_{121}	Liquidized aloe gel juice + clove oil 1ml
S_{122}	Liquidized aloe gel juice + clove oil 1.25ml
S_{123}	Liquidized aloe gel juice + clove oil 1.50ml
S_{124}	Liquidized aloe gel juice + clove oil 1.75ml
S_{125}	Liquidized aloe gel juice + clove oil 2ml
S_{126}	Liquidized aloe gel juice + cardamom oil 1ml
S_{127}	Liquidized aloe gel juice + cardamom oil 1.25ml
S_{128}	Liquidized aloe gel juice + cardamom oil 1.50ml
S_{129}	Liquidized aloe gel juice + cardamom oil 1.75ml
S_{130}	Liquidized aloe gel juice + cardamom oil 2ml

APPENDIX VI

SENSORY PARAMETERS, pH, REFRACTIVE INDEX AND SPECIFIC GRAVITY OF LIQUIDIZED ALOE GEL JUICE IN PRELIMINARY STUDIES

VI (1). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Gymnema sylvestre* aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_1	78.66	67.36	75.50	68.06	88.18	76.52
S_2	69.76	66.12	65.96	60.26	73.14	72.26
S_3	66.20	63.16	65.48	58.57	65.50	59.90
S_4	58.96	59.42	62.98	68.86	45.72	50.90
S_5	41.42	58.94	45.08	61.94	42.46	56.30
KW value	17.11	1.25	11.28	1.80	30.31	10.04
χ^2	9.49					
C D value	2.14					

VI (2). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Gymnema sylvestre* aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	RI	SG	pH	RI	SG	pH
S_1	1.333	1.000	3.51	1.333	1.001	6.75
S_2	1.333	1.000	3.50	1.334	1.003	6.78
S_3	1.333	1.000	3.49	1.335	1.005	6.86
S_4	1.333	1.000	3.50	1.335	1.005	7.04
S_5	1.333	1.000	3.50	1.335	1.005	7.21
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	NS	NS	NS	0.04

VI (3). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Gymnema sylvestre* tincture extracts

Tincture	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_6	66.64	89.94	81.26	76.02	72.14	73.20
S_7	65.34	61.04	66.10	71.86	66.62	65.00
S_8	63.04	60.16	61.44	71.48	64.78	63.36
S_9	61.36	58.68	53.10	56.16	58.06	59.22
S_{10}	58.62	50.18	53.10	39.48	53.40	54.22
KW value	0.89	14.63	11.85	21.38	4.66	4.21
χ^2	9.49					
C D value	2.14					

VI (4). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Gymnema sylvestre* tincture extracts

Tincture	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_6	1.333	1.000	3.50	1.333	1.001	4.01
S_7	1.333	1.000	3.50	1.333	1.001	4.05
S_8	1.333	1.000	3.50	1.333	1.001	4.10
S_9	1.333	1.000	3.50	1.333	1.001	4.18
S_{10}	1.333	1.000	3.50	1.333	1.001	4.27
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	NS	NS	NS	0.02

VI (5). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Gymnema sylvestre* decoction extracts

Decoction	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{11}	75.52	71.82	79.44	52.1 0	65.72	50.62
S_{12}	72.34	70.58	74.82	68.60	63.62	70.70
S_{13}	60.26	55.98	58.96	7 7.90	81.38	70.68
S_{14}	53.18	68.32	58.96	5 9.80	63.32	70.70
S_{15}	53.70	48.30	42.82	56.60	40.96	52.30
KW value	9.73	9.04	19.41	9.12	18.43	9.46
χ^2	9.49					
C D value	2.14					

VI (6). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Gymnema sylvestre* decoction extracts

Decoction	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{11}	1.333	1.000	3.50	1.335	1.005	6.62
S_{12}	1.333	1.000	3.50	1.334	1.003	6.43
S_{13}	1.333	1.000	3.50	1.333	1.001	6.35
S_{14}	1.333	1.000	3.50	1.335	1.005	6.70
S_{15}	1.333	1.000	3.50	1.335	1.005	6.73
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.01
CD (0.05)	NS	NS	NS	NS	NS	0.03

VI (7). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Centella asiatica* aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{16}	80.04	77.46	76.50	82.18	84.06	88.92
S_{17}	73.60	66.00	67.02	64.04	66.04	78.34
S_{18}	57.40	64.12	63.04	60.62	59.12	56.16
S_{19}	54.06	55.44	56.72	58.66	55.72	49.26
S_{20}	49.90	51.98	51.72	49.50	51.74	42.32
KW value	14.68	8.38	8.33	12.10	13.84	33.83
χ^2	9.49					
CD value	2.14					

VI (8). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Centella asiatica* aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{16}	1.333	1.000	3.30	1.333	1.001	2.42
S_{17}	1.333	1.000	3.49	1.333	1.001	2.93
S_{18}	1.333	1.000	3.50	1.333	1.001	4.50
S_{19}	1.333	1.000	3.48	1.334	1.001	4.77
S_{20}	1.333	1.000	3.49	1.334	1.002	4.89
SEm(±)	0.00	0.00	0.05	0.00	0.00	0.05
CD (0.05)	NS	NS	NS	NS	NS	0.15

VI (9). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Centella asiatica* tincture extracts

Tincture	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{21}	82.16	68.22	80.06	54.16	55.28	57.58
S_{22}	72.18	67.32	80.64	60.50	58.32	59.30
S_{23}	63.80	60.28	79.04	63.02	61.80	64.48
S_{24}	49.90	59.94	42.06	65.06	68.78	65.86
S_{25}	46.96	59.24	33.20	72.26	70.82	67.78
KW value	18.58	1.63	47.16	4.08	3.70	1.66
χ^2	9.49					
CD value	2.14					

VI (10). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Centella asiatica* tincture extracts

Tincture	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{21}	1.333	1.000	3.45	1.334	1.002	2.98
S_{22}	1.333	1.001	3.50	1.333	1.001	3.23
S_{23}	1.333	1.001	3.50	1.333	1.001	3.40
S_{24}	1.333	1.001	3.50	1.333	1.000	3.51
S_{25}	1.333	1.001	3.50	1.333	1.000	3.49
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.05
CD (0.05)	NS	NS	NS	NS	NS	0.15

VI (11). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Centella asiatica* decoction extracts

Decoction	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{26}	90.36	75.40	85.14	83.56	88.00	69.32
S_{27}	68.82	69.66	79.38	71.54	71.48	66.32
S_{28}	59.06	66.28	63.34	59.68	67.88	61.22
S_{29}	52.26	52.22	46.08	58.26	54.74	60.98
S_{30}	44.50	51.44	41.06	41.96	32.90	57.16
KW value	26.39	9.71	32.16	20.26	35.21	2.07
χ^2	9.49					
CD value	2.14					

VI (12). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of Centella *asiatica* decoction extracts

Decoction	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{26}	1.333	1.000	3.49	1.333	1.000	3.49
S_{27}	1.333	1.000	3.50	1.333	1.000	3.49
S_{28}	1.333	1.000	3.49	1.333	1.000	3.49
S_{29}	1.333	1.000	3.49	1.333	1.000	3.50
S_{30}	1.333	1.000	3.50	1.333	1.000	3.52
SEm(±)	0.000	0.000	0.00	0.000	0.000	0.00
CD (0.05)	NS	NS	NS	NS	0.01	0.01

VI (13). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Achyranthes aspera* aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{31}	71.10	74.42	67.22	66.40	66.14	73.38
S_{32}	66.86	72.06	67.22	71.12	78.62	75.34
S_{33}	63.12	66.30	63.32	65.98	60.86	66.90
S_{34}	58.28	62.64	62.48	56.46	58.84	52.86
S_{35}	55.64	39.58	54.76	55.04	50.54	46.52
KW value	3.66	17.17	2.37	4.17	9.27	14.28
χ^2	9.49					
CD value	2.14					

VI (14). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Achyranthes aspera* aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{31}	1.333	1.000	3.49	1.333	1.001	3.92
S_{32}	1.333	1.000	3.48	1.333	1.001	3.90
S_{33}	1.333	1.000	3.47	1.333	1.001	3.92
S_{34}	1.333	1.000	3.49	1.333	1.001	3.94
S_{35}	1.333	1.000	3.49	1.333	1.001	3.97
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	NS	NS	NS	0.01

VI (15). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Achyranthes aspera* tincture extracts

Tincture	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{36}	77.22	77.10	70.18	56.54	50.50	42.30
S_{37}	68.28	66.30	68.22	60.86	52.74	63.66
S_{38}	60.16	64.42	64.08	61.70	68.44	66.36
S_{39}	55.34	54.24	60.66	64.50	69.64	68.22
S_{40}	54.00	52.94	51.86	71.40	73.68	74.46
KW value	8.73	8.66	4.81	2.59	9.56	14.61
χ^2	9.49					
CD value	2.14					

VI (16). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Achyranthes aspera* tincture extracts

Tincture	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{36}	1.333	1.000	3.48	1.334	1.001	3.23
S_{37}	1.333	1.000	3.49	1.333	1.001	3.25
S_{38}	1.333	1.000	3.50	1.333	1.001	3.33
S_{39}	1.333	1.001	3.49	1.333	1.000	3.48
S_{40}	1.333	1.001	3.49	1.333	1.000	3.52
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.02
CD (0.05)	NS	NS	NS	NS	NS	0.06

VI (17). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Achyranthes aspera* decoction extracts

Decoction	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{41}	70.98	72.14	66.90	83.04	74.50	75.48
S_{42}	64.26	70.56	68.16	73.04	70.34	63.24
S_{43}	61.50	61.52	63.44	71.52	57.92	60.42
S_{44}	61.86	58.40	59.98	47.94	57.92	58.20
S_{45}	56.40	52.38	56.52	39.46	54.26	57.66
KW value	2.57	5.98	2.31	28.67	7.04	5.18
χ^2	9.49					
CD value	2.14					

VI (18). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Achyranthes aspera* decoction extracts

Decoction	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{41}	1.333	1.000	3.49	1.334	1.002	4.54
S_{42}	1.333	1.000	3.49	1.334	1.002	4.67
S_{43}	1.333	1.000	3.50	1.334	1.002	5.24
S_{44}	1.333	1.000	3.49	1.334	1.002	5.88
S_{45}	1.333	1.000	3.50	1.334	1.002	5.95
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.08
CD (0.05)	NS	NS	NS	NS	NS	0.25

VI (19). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Tridax procumbens* aqueous extracts

Aqueous	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{46}	75.00	74.46	76.66	72.38	72.80	101.22
S_{47}	71.28	66.54	74.66	70.92	66.22	76.08
S_{48}	60.54	62.06	67.82	69.70	64.54	60.46
S_{49}	55.38	60.56	52.48	56.64	63.14	46.54
S_{50}	52.80	51.38	43.38	45.36	48.30	30.70
KW value	8.18	6.06	18.52	11.69	7.07	61.31
χ^2	9.49					
CD value	2.14					

VI (20). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Tridax procumbens* aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{46}	1.333	1.000	3.49	1.334	1.003	4.36
S_{47}	1.333	1.000	3.50	1.334	1.003	4.85
S_{48}	1.333	1.000	3.50	1.334	1.003	4.79
S_{49}	1.333	1.000	3.50	1.334	1.004	2.77
S_{50}	1.333	1.000	3.50	1.334	1.004	2.61
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.18
CD (0.05)	NS	NS	0.01	NS	NS	0.52

VI (21). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Tridax procumbens* tincture extracts

Tincture	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{51}	71.30	72.12	73.98	28.46	48.70	48.18
S_{52}	63.87	71.00	64.56	54.90	56.30	56.34
S_{53}	62.86	61.68	59.84	66.28	58.80	61.68
S_{54}	58.64	57.82	58.44	82.48	74.50	66.20
S_{55}	55.88	52.38	58.18	82.88	76.70	82.60
KW value	2.98	6.38	4.37	43.25	12.33	13.71
χ^2	9.49					
CD value	2.14					

VI (22). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Tridax procumbens* tincture extracts

Tincture	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{51}	1.333	1.000	3.51	1.334	1.003	4.73
S_{52}	1.333	1.000	3.51	1.334	1.003	4.69
S_{53}	1.333	1.000	3.50	1.334	1.002	4.63
S_{54}	1.333	1.001	3.49	1.334	1.001	4.56
S_{55}	1.333	1.001	3.50	1.333	1.001	4.00
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.02
CD (0.05)	NS	NS	NS	NS	NS	0.04

VI (23). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Tridax procumbens* decoction extracts

Decoction	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{56}	80.90	69.28	70.34	81.40	76.46	92.30
S_{57}	79.48	65.22	68.20	63.00	63.76	70.80
S_{58}	54.90	65.14	64.48	60.32	60.14	60.58
S_{59}	50.96	61.00	59.84	57.06	60.08	48.52
S_{60}	48.76	54.36	52.14	53.22	54.56	42.80
KW value	21.25	2.87	4.90	10.07	5.92	31.79
χ^2	9.49					
CD (0.05)	2.14					

VI (24). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Tridax procumbens* decoction extracts

Decoction	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{56}	1.333	1.000	3.50	1.334	1.004	5.74
S_{57}	1.333	1.000	3.50	1.334	1.004	5.84
S_{58}	1.333	1.000	3.49	1.335	1.004	6.34
S_{59}	1.333	1.000	3.50	1.335	1.004	6.47
S_{60}	1.333	1.000	3.51	1.335	1.004	6.50
SEm(±)	0.00	0.00	0.01	0.00	0.00	0.04
CD (0.05)	NS	NS	0.01	NS	NS	0.13

VI (25). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Terminalia chebula* aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{61}	85.20	68.76	97.12	81.68	81.34	75.10
S_{62}	81.86	63.66	80.94	81.20	76.66	66.92
S_{63}	65.86	61.12	50.78	57.82	67.62	66.36
S_{64}	51.38	54.68	48.28	48.74	47.96	57.74
S_{65}	30.70	66.78	37.88	45.56	41.42	48.88
KW value	43.11	2.75	53.49	24.97	25.57	8.67
χ^2	9.49					
CD value	2.14					

VI (26). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Terminalia chebula* aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{61}	1.333	1.000	3.49	1.334	1.004	4.61
S_{62}	1.333	1.000	3.49	1.334	1.004	4.61
S_{63}	1.333	1.000	3.50	1.334	1.004	4.62
S_{64}	1.333	1.000	3.50	1.334	1.004	4.62
S_{65}	1.333	1.000	3.50	1.335	1.004	4.63
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	0.01	NS	NS	NS

VI (27). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Terminalia chebula* tincture extracts

Tincture	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{66}	102.86	77.50	78.32	78.44	74.84	68.00
S_{67}	73.82	69.28	65.48	60.64	68.72	65.28
S_{68}	60.54	61.21	65.02	60.28	62.22	62.76
S_{69}	43.94	58.32	54.78	59.74	57.60	60.48
S_{70}	33.84	51.28	51.40	55.90	51.62	58.48
KW value	60.19	8.53	10.06	6.93	6.96	1.29
χ^2	9.49					
CD value	2.14					

VI (28). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Terminalia chebula* tincture extracts

Tincture	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{66}	1.333	1.000	3.50	1.333	1.002	3.84
S_{67}	1.333	1.000	3.50	1.334	1.002	3.85
S_{68}	1.333	1.001	3.50	1.334	1.002	3.86
S_{69}	1.333	1.001	3.50	1.334	1.002	3.86
S_{70}	1.333	1.001	3.50	1.334	1.002	3.88
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	NS	NS	NS	0.01

VI (29). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Terminalia chebula* decoction extracts

Decoction	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{71}	83.40	73.96	83.72	94.08	70.80	98.58
S_{72}	80.40	67.90	77.84	82.70	64.10	92.52
S_{73}	71.70	66.38	50.68	65.90	63.10	61.30
S_{74}	42.76	55.10	56.44	36.46	59.00	33.58
S_{75}	38.44	51.66	46.32	35.86	58.00	29.02
KW value	37.29	7.72	24.34	56.37	2.24	83.27
χ^2	9.49					
CD value	2.14					

VI (30). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Terminalia chebula* decoction extracts

Decoction	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{71}	1.333	1.000	3.50	1.334	1.004	3.67
S_{72}	1.333	1.000	3.50	1.335	1.004	3.69
S_{73}	1.333	1.000	3.49	1.335	1.004	3.71
S_{74}	1.333	1.000	3.51	1.335	1.004	3.89
S_{75}	1.333	1.000	3.50	1.335	1.005	4.12
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.03
CD (0.05)	NS	NS	NS	NS	NS	0.08

VI (31). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Punica granatum* (rind) aqueous extracts

Aqueous extract	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{76}	82.50	65.56	74.16	73.10	73.18	87.60
S_{77}	72.00	63.08	71.96	73.10	69.10	77.48
S_{78}	68.00	62.56	69.20	62.10	63.94	68.68
S_{79}	58.00	62.32	51.48	57.50	55.16	48.84
S_{80}	34.50	61.48	48.20	49.20	53.62	32.40
KW value	29.73	0.21	13.96	9.05	6.53	42.47
χ^2	9.49					
CD value	2.14					

VI (32). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Punica granatum* (rind) aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{76}	1.333	1.000	3.49	1.334	1.002	3.71
S_{77}	1.333	1.000	3.51	1.334	1.003	3.76
S_{78}	1.333	1.000	3.50	1.334	1.004	3.78
S_{79}	1.333	1.000	3.51	1.335	1.005	3.83
S_{80}	1.333	1.000	3.50	1.335	1.006	3.89
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	0.01	NS	NS	0.01

VI (33). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Punica granatum* (rind) tincture extracts

Tincture	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{81}	67.28	68.90	75.14	58.94	56.78	42.26
S_{82}	64.48	65.60	68.98	60.96	59.04	52.04
S_{83}	64.78	64.04	68.42	60.96	60.74	68.94
S_{84}	61.34	60.32	57.24	67.02	67.84	72.24
S_{85}	51.12	56.14	45.22	67.12	70.60	79.52
KW value	1.40	2.01	12.28	1.30	2.91	20.99
χ^2	9.49					
CD value	2.14					

VI (34). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Punica granatum* (rind) tincture extracts

Tincture	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{81}	1.333	1.000	3.50	1.339	1.015	3.95
S_{82}	1.333	1.000	3.50	1.338	1.015	3.89
S_{83}	1.333	1.000	3.50	1.338	1.014	3.83
S_{84}	1.333	1.001	3.50	1.338	1.014	3.70
S_{85}	1.333	1.001	3.50	1.338	1.014	3.45
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.03
CD (0.05)	NS	NS	NS	NS	NS	0.09

VI (35). Sensory parameters of liquidized aloe gel juice added with different concentrations of *Punica granatum* (rind) decoction extracts

Decoction	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{86}	91.54	75.34	97.24	76.78	73.26	81.16
S_{87}	67.68	67.58	61.84	75.64	69.66	75.20
S_{88}	66.70	61.94	61.16	62.12	64.92	71.92
S_{89}	40.26	57.22	49.24	59.82	57.26	54.76
S_{90}	48.82	52.92	45.52	40.64	49.90	31.96
KW value	33.40	6.95	35.64	18.31	7.48	32.94
χ^2	9.49					
CD value	2.14					

VI (36). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of *Punica granatum* (rind) decoction extracts

Decoction	1st day			1st month		
Observations	RI	Specific gravity	pH	RI	Specific gravity	Ph
S_{86}	1.333	1.000	3.50	1.334	1.003	2.47
S_{87}	1.333	1.000	3.50	1.334	1.003	2.45
S_{88}	1.333	1.000	3.50	1.334	1.003	2.42
S_{89}	1.333	1.000	3.51	1.334	1.003	2.36
S_{90}	1.333	1.000	3.50	1.334	1.004	2.31
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.01
CD (0.05)	NS	NS	0.01	NS	NS	0.02

VI (37). Sensory parameters of liquidized aloe gel juice added with different concentrations of green tea aqueous extracts

Aqueous extracts	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{91}	99.66	70.20	94.76	44.28	41.44	38.00
S_{92}	85.70	67.70	62.08	49.90	48.56	60.60
S_{93}	52.76	66.76	59.84	52.28	67.54	67.44
S_{94}	40.54	59.88	59.84	76.88	77.30	73.52
S_{95}	36.34	50.46	38.48	90.27	80.16	75.44
KW value	65.98	5.37	40.34	35.57	25.81	19.83
χ^2	9.49					
CD value	2.14					

VI (38). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of green tea aqueous extracts

Aqueous extract	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{91}	1.333	1.000	3.49	1.334	1.005	4.84
S_{92}	1.333	1.000	3.50	1.334	1.005	4.52
S_{93}	1.333	1.000	3.50	1.334	1.003	4.37
S_{94}	1.333	1.000	3.50	1.334	1.002	4.35
S_{95}	1.333	1.000	3.50	1.334	0.996	4.24
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	NS	NS	0.01	0.01

VI (39). Sensory parameters of liquidized aloe gel juice added with different concentrations of green tea tincture extracts

Tincture	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{96}	93.32	78.68	88.44	88.36	82.76	74.10
S_{97}	79.88	64.52	67.32	80.66	64.92	69.98
S_{98}	51.88	62.66	62.52	70.14	57.64	66.44
S_{99}	47.88	56.92	60.60	51.08	56.32	59.16
S_{100}	42.04	52.22	36.12	24.76	53.36	45.32
KW value	43.69	8.68	30.19	53.88	11.41	10.83
χ^2	9.49					
CD value	2.14					

VI (40). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of green tea tincture extracts

Tincture	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{96}	1.333	1.000	3.50	1.334	1.000	3.52
S_{97}	1.333	1.000	3.50	1.334	1.000	3.53
S_{98}	1.333	1.000	3.50	1.334	1.000	3.53
S_{99}	1.333	1.000	3.50	1.334	1.000	3.57
S_{100}	1.333	1.000	3.50	1.334	1.000	3.58
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	NS	NS	NS	0.01

VI (41). Sensory parameters of liquidized aloe gel juice added with different concentrations of green tea decoction extracts

Decoction	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{101}	100.28	70.88	80.46	75.86	84.48	98.24
S_{102}	88.58	68.42	75.96	75.32	76.36	86.90
S_{103}	52.20	65.18	62.32	58.18	59.32	45.50
S_{104}	45.16	55.30	52.96	53.54	55.20	48.54
S_{105}	28.78	55.22	43.30	52.10	39.64	35.82
KW value	72.96	4.54	20.78	12.34	26.07	64.43
χ^2	9.49					
CD value	2.14					

VI (42). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of green tea decoction extracts

Decoction	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{101}	1.333	1.000	3.50	1.334	0.999	3.55
S_{102}	1.333	1.000	3.50	1.335	1.003	3.96
S_{103}	1.333	1.000	3.50	1.335	1.003	4.17
S_{104}	1.333	1.000	3.50	1.335	1.004	5.31
S_{105}	1.333	1.000	3.51	1.335	1.004	5.50
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.01
CD (0.05)	NS	NS	NS	NS	NS	0.03

VI (43). Sensory parameters of liquidized aloe gel juice added with different concentrations of sacred basil oil

Sacred basil oil	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{106}	100.44	93.24	75.40	85.64	81.80	67.02
S_{107}	65.42	77.34	62.70	67.46	76.84	65.68
S_{108}	51.26	73.90	64.22	64.62	61.76	65.54
S_{109}	49.86	65.42	57.10	52.00	47.38	59.26
S_{110}	48.02	55.10	55.58	45.28	47.22	57.50
KW value	42.50	17.19	5.11	22.59	22.31	1.73
χ^2	9.49					
CD value	2.14					

VI (44). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of sacred basil oil

Sacred basil oil	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{106}	1.333	1.000	3.50	1.333	1.001	3.52
S_{107}	1.333	1.000	3.50	1.333	1.001	3.54
S_{108}	1.333	1.000	3.49	1.333	1.001	3.52
S_{109}	1.333	1.000	3.50	1.333	1.001	3.53
S_{110}	1.333	1.000	3.50	1.333	1.001	3.54
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	NS	NS	NS	0.01

VI (45). Sensory parameters of liquidized aloe gel juice added with different concentrations of lemon grass oil

Lemon grass oil	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{111}	87.74	90.60	72.86	70.26	71.22	72.84
S_{112}	74.12	75.30	67.22	68.88	66.02	64.66
S_{113}	58.31	57.62	65.78	64.80	64.96	62.44
S_{114}	52.34	53.52	55.82	61.32	63.68	59.40
S_{115}	39.82	37.96	53.32	49.74	49.12	55.66
KW value	29.74	33.86	6.02	6.00	5.73	3.89
χ^2	9.49					
CD value	2.14					

VI (46). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of lemongrass oil

Lemon grass oil	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{111}	1.334	1.000	3.51	1.334	1.000	3.51
S_{112}	1.334	1.000	3.50	1.334	1.000	3.52
S_{113}	1.334	1.000	3.50	1.334	1.000	3.52
S_{114}	1.334	1.000	3.50	1.335	1.000	3.53
S_{115}	1.334	1.000	3.50	1.335	1.001	3.53
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.01
CD (0.05)	NS	NS	NS	NS	NS	NS

VI (47). Sensory parameters of liquidized aloe gel juice added with different concentrations of cinnamon bark oil

Cinnamon bark oil	1st day			1st month		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{116}	80.04	80.96	95.00	73.68	76.42	70.50
S_{117}	65.18	74.66	66.80	67.56	76.14	64.44
S_{118}	59.66	54.60	55.24	66.12	57.84	60.94
S_{119}	59.26	53.88	51.32	63.84	57.02	59.56
S_{120}	50.86	50.90	46.64	43.80	47.58	59.56
KW value	10.47	17.82	31.39	11.41	15.21	1.92
χ^2	9.49					
CD value	2.14					

VI (48). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of cinnamon bark oil

Cinnamon bark oil	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{116}	1.333	1.000	3.49	1.333	1.000	3.53
S_{117}	1.333	1.000	3.50	1.333	1.000	3.58
S_{118}	1.333	1.000	3.50	1.333	1.000	3.63
S_{119}	1.333	1.001	3.50	1.334	1.001	3.64
S_{120}	1.334	1.001	3.50	1.334	1.001	3.68
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.01
CD (0.05)	NS	NS	NS	NS	NS	0.02

VI (49). Sensory parameters of liquidized aloe gel juice added with different concentrations of clove oil

Clove oil	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{121}	87.94	70.98	84.10	78.20	74.44	80.04
S_{122}	77.92	68.30	73.26	78.20	64.80	73.86
S_{123}	45.10	63.66	58.22	59.96	62.72	66.46
S_{124}	46.30	60.36	55.32	55.00	60.48	51.12
S_{125}	57.74	51.70	44.10	43.64	52.56	43.52
KW value	31.25	5.40	21.83	20.37	5.86	20.94
χ^2	9.49					
CD value	2.14					

VI (50). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of clove oil

Clove oil	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	pH
S_{121}	1.333	1.001	3.50	1.333	1.000	3.50
S_{122}	1.333	1.001	3.50	1.333	1.000	3.51
S_{123}	1.333	1.001	3.50	1.333	1.000	3.52
S_{124}	1.334	1.001	3.50	1.334	1.001	3.53
S_{125}	1.334	1.001	3.50	1.334	1.001	3.53
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	NS	NS	NS	0.01

VI (51). Sensory parameters of liquidized aloe gel juice added with different concentrations of cardamom oil

Cardamom oil	1st day			30th day		
Observations	Appearance	Odour	Colour	Appearance	Odour	Colour
S_{126}	74.36	82.96	70.58	85.60	74.96	85.36
S_{127}	67.96	78.10	66.26	79.66	73.44	75.24
S_{128}	65.60	60.60	63.50	70.36	64.86	54.42
S_{129}	58.68	49.84	62.72	46.50	54.88	52.76
S_{130}	48.40	43.50	51.94	32.88	46.86	47.22
KW value	9.68	27.07	4.34	44.16	13.29	22.88
χ^2	9.49					
CD value	2.14					

VI (52). Refractive index, specific gravity and pH of liquidized aloe gel juice with different concentrations of cardamom oil

Cardamom oil	1st day			30th day		
Observations	RI	Specific gravity	pH	RI	Specific gravity	Ph
S_{126}	1.333	1.000	3.49	1.334	1.000	3.51
S_{127}	1.333	1.000	3.50	1.334	1.000	3.52
S_{128}	1.334	1.001	3.50	1.334	1.000	3.52
S_{129}	1.334	1.001	3.50	1.334	1.000	3.52
S_{130}	1.334	1.001	3.50	1.334	1.000	3.53
SEm(±)	0.00	0.00	0.00	0.00	0.00	0.00
CD (0.05)	NS	NS	NS	NS	NS	NS

സംഗ്രഹം

വെള്ളായണി കാർഷിക കോളേജിലെ തോട്ടവിള സുഗന്ധവ്യഞ്ജന വിഭാഗത്തിൽ 2016 മാർച്ച് മുതൽ 2019 ഡിസംബർ വരെ "കറ്റാർ വാഴയിലെ (*അലോ വേര* ബർമി. എഫ്.) ന്യൂട്രാസ്യൂട്ടിക്കൽസും ജെൽ സ്ഥിരതയ്ക്കുള്ള പ്രോട്ടോക്കോൾ വികസനവും" എന്ന വിഷയത്തിൽ ഗവേഷണം നടത്തുകയുണ്ടായി. കറ്റാർവാഴയ്ക്ക് കുറഞ്ഞ ചെലവിൽ ഔഷധിയസത്തും തൈലവും ഉപയോഗിച്ച്സ്ഥിരത കൈവരിക്കുന്നതിനുള്ള സാങ്കേതിക വിദ്യ രൂപീകരിക്കുന്നതിനും കറ ഉണക്കി തയ്യാറാക്കുന്നതിനുള്ള പ്രോട്ടോകോൾ വികസിപ്പിക്കുന്നതിനും ന്യൂട്രാസ്യൂട്ടിക്കൽസ് വിപണനം ചെയ്യുന്നതിനുമുള്ള പദ്ധതിയാണ് വിഭാവനം ചെയ്തത്.നാല് പരീക്ഷണങ്ങളിൽ ആയിട്ടാണ് ഗവേഷണം നടത്തിയത്. ഒന്നാമത്തെ പരീക്ഷണം ജെല്ലിന്റെ ഗുണനിലവാരവും സ്വാഭാവികമായി ഉണ്ടാകാവുന്ന കേടുപാടുകളും വിലയിരുത്തുക എന്നതായിരുന്നു. അതേസമയം കറ്റാർവാഴയുടെ (കുർകാവോ അലോ) ഉണങ്ങിയ കറ സ്ഥിരപ്പെടുത്തുക എന്നതായിരുന്നു രണ്ടാമത്തെ പരീക്ഷണം. മൂന്നാമത്തെ പരീക്ഷണം ഔഷധിയ സത്തും തൈലങ്ങളും ഉപയോഗിച്ച് ജെൽ സ്ഥിരപ്പെടുത്തുക എന്നതായിരുന്നു. കറ്റാർവാഴയുടെ സത്തിൽ നിന്നും ന്യൂട്രാസ്യൂട്ടിക്കൽസ് തയ്യാറാക്കുക എന്നതായിരുന്നു അവസാന പരീക്ഷണം.

`ശുദ്ധമായ ജെല്ലിന്റെ ഗുണനിലവാരവും സ്വാഭാവികമായി ഉണ്ടാകാവുന്ന കേടുപാടുകളും വിലയിരുത്തുന്നതിന് വേണ്ടി ദ്രവീകൃതമായ കറ്റാർവാഴയുടെ ജെല്ലിന്റെ സത്ത് ഏഴു ദിവസത്തേക്ക് ഗ്ലാസ് കുപ്പികളിൽ സൂക്ഷിച്ചു വയ്ക്കുകയുണ്ടായി. ആദ്യത്തെ മൂന്ന് ദിവസം വെളുത്ത നിറത്തിൽ കാണപ്പെട്ട കറ്റാർവാഴയുടെ ജെല്ലിന്റെ സത്ത് നാലാം ദിവസം മുതൽ ദുർഗന്ധത്തോടു കൂടി കുപ്പിയിൽ അടിഞ്ഞുകൂടുന്നത് ആയി കാണപ്പെട്ടു. രണ്ടാം ദിവസം മുതൽ സൂക്ഷ്മജീവികളുടെ എണ്ണം വർദ്ധിക്കുന്നതിനാൽ കറ്റാർവാഴയുടെ ജെല്ലിന്റെ സത്ത് ഒരു ദിവസത്തിൽ കൂടുതൽ സൂക്ഷിക്കാൻ ആവുന്നതല്ല. ദ്രവീകൃതമായ കറ്റാർവാഴയുടെ ജെല്ലിന്റെ സത്ത് 65 ഡിഗ്രി സെൽഷ്യസിൽ 15 പിഎസ്ഐ മർദ്ദത്തിൽ 13 മിനിറ്റ്പാസ്ചറൈസ് ചെയ്തു ഫ്ലാഷ് കൂളിംഗ് നടത്തിയതിൻറെ ഫലമായി ഏഴാം ദിവസത്തിനു ശേഷവും സൂക്ഷ്മാണുക്കളുടെ സാന്നിധ്യം കണ്ടെത്താനായില്ല.

കറ്റാർവാഴയുടെ ഇലകളിൽ നിന്ന് ശേഖരിച്ച കറ തിളപ്പിക്കൽ, തണുപ്പിക്കൽ എന്നീ പ്രക്രിയകൾക്കൊപ്പം സൂര്യപ്രകാശം, തണൽ, ഓവൻ ഉപയോഗിച്ചുള്ള ഉണക്കൽ എന്നീ വ്യത്യസ്ത ഉണക്കൽ രീതികൾക്ക് വിധേയമാക്കി. ഉണങ്ങിയ കറയുടെ രൂപവും നിറവും അലോയിൻ അളവും (271.62 മി.ഗ്രാം/മില്ലി) തണലിൽ ഉണക്കിയവയ്ക്ക് ഉയർന്നതായി കാണപ്പെട്ടു.

പാസ്ചറൈസ് ചെയ്ത കറ്റാർവാഴയുടെ ജെല്ലിന്റെ സത്ത് 0.5 ശതമാനം സിട്രിക് ആസിഡ് ചേർത്ത് പി.എച്ച് 3.5 ആയി ക്രമീകരിച്ചതിനുശേഷം വ്യത്യസ്ത ഗാഢതയിൽ ഔഷധിയ സത്തും തൈലവും മൂന്ന് രൂപങ്ങളിൽ (ജലീയ സത്ത്, ടിങ്ങ്ചർ, കഷായം) ചേർത്തു. ഈ സാമ്പിളുകൾ ഒരു മാസത്തേക്ക് സൂക്ഷിച്ചു വെക്കുകയും

സൂക്ഷ്മാണുക്കളുടെ എണ്ണത്തെയും കുറഞ്ഞ ഇൻഹിബിറ്ററി കോൺസെൻട്രേഷനെയും അടിസ്ഥാനമാക്കി മികച്ച ട്രീറ്റ്മെൻറ് ജെൽ സ്ഥിരതയ്ക്കായി നടത്തിയ പ്രാഥമിക പരീക്ഷണങ്ങളിൽ നിന്ന് തെരഞ്ഞെടുക്കുകയും ചെയ്തു. *ജിംനെമ സിൽവെസ്ട്രെ* യുടെ ജലീയ സത്ത് (1 മില്ലി), ടിങ്ങ്ചർ (1 മില്ലി), കഷായം (2 മില്ലി), *സെന്റെല്ല ഏഷ്യാറ്റിക്ക* യുടെ ജലീയ സത്ത് (1 മില്ലി), ടിങ്ങ്ചർ (2 മില്ലി), കഷായം (1 മില്ലി), *അചൈറന്തസ് അസ്പെറ* യുടെ ജലീയ സത്ത് (2 മില്ലി), ടിങ്ങ്ചർ (2 മില്ലി), കഷായം (1.5 മില്ലി), *ട്രിഡാക്സ്* ന്റെ ജലീയ സത്ത് (2 മില്ലി), ടിങ്ങ്ചർ (2 മില്ലി), കഷായം (1 മില്ലി), *ടെർമിനാലിയ ചെബുല* യുടെ ജലീയ സത്ത് (1 മില്ലി), ടിങ്ങ്ചർ (1 മില്ലി), കഷായം (1 മില്ലി), *പ്യൂണിക്ക ഗ്രാനാറ്റ* തിന്റെ ജലീയ സത്ത് (1 മില്ലി), ടിങ്ങ്ചർ (2 മില്ലി), കഷായം (1 മില്ലി), തേയിലയുടെ പച്ചയിലയുടെ ജലീയ സത്ത് (2 മില്ലി), ടിങ്ങ്ചർ (1 മില്ലി), കഷായം (2 മില്ലി) എന്നിവയോടൊപ്പം ഒരു 1 മില്ലി വീതം തുളസി തൈലം, പുൽത്തൈലം, കറുവാപ്പട്ടയുടെ പുറംതൊലിയിൽ നിന്നുള്ള തൈലം, ഗ്രാമ്പൂ തൈലം, ഏലക്ക തൈലം എന്നിവകൂടി തിരഞ്ഞെടുത്ത് ജെൽ സ്ഥിരതയ്ക്കായി, പി.എച്ച് ക്രമീകരിച്ച്, പാസ്ചറൈസ് ചെയ്ത് ദ്രവീകൃതമാക്കിയ കറ്റാർവാഴയുടെ സത്തിലേക്ക് (25 മില്ലി) ചേർത്തു കൊടുത്തു. തിരഞ്ഞെടുത്ത ഇരുപത്തിയാറ് ട്രീറ്റ്മെൻറ്സ് കംപ്ലീറ്റ്ലി റൻഡമൈസ്ഡ് ഡിസൈനിൽ 5 പ്രാവശ്യം റപ്ലിക്കേറ്റ് ചെയ്യുകയും അവ 0.08 ശതമാനം സോഡിയം ബെൻസോയേറ്റുമായി താരതമ്യപ്പെടുത്തുകയും ആറുമാസത്തേക്ക് സൂക്ഷിച്ചു വെക്കുകയും ചെയ്തു.

ഔഷധിയ സത്ത് അടങ്ങിയ ട്രീറ്റ്മെൻറ്കൾക്ക് തൈലം കൊണ്ടുള്ള ട്രീറ്റ്മെൻറ്നേക്കാൾ രൂപവും നിറവും ഗന്ധവും കുറയുന്നതായി കാണപെട്ടു. ടോട്ടൽ സോളിഡ്ന്റെ അളവും അമിലേസ്, ലിപേസ് എന്നിവയുടെ പ്രവർത്തനവും കുറയുന്നതായി കണ്ടു. കറ്റാർ വാഴയുടെ ജെല്ലിന്റെ സത്ത് അക്കൈറന്തസ് അസ്പെറയുടെ (0.08 പിപിഎം) ജലീയ സത്ത്, മൂല കഷായം, കഷായം എന്നിവയുമായി ചേർത്തപ്പോൾ അമിനോആസിഡിന്റെ അളവ് കൂടുതലായിരുന്നു. തേയിലയുടെ പച്ചയിലയുടെ ജലീയ സത്ത് അടങ്ങിയ ട്രീറ്റ്മെൻറ്ൽ വൈറ്റമിൻ എയും സിയും കൂടുന്നതായും പഴക്കം ഏറും തോറും വൈറ്റമിനുകൾ കുറയുന്നതായും കണ്ടെത്തി. ഹെർബൽ എക്സ്ട്രാക്റ്റ് അടങ്ങിയ ട്രീറ്റ്മെൻറ്കളിൽ സൂക്ഷ്മജീവികളുടെ എണ്ണം ഒന്നാം മാസം മുതൽ വർദ്ധിക്കുന്നതായി കണ്ടെത്തി. 1 മില്ലി ഗ്രാമ്പൂ തൈലം കറ്റാർവാഴയുടെ സത്തുമായി കൂട്ടി കലർത്തുന്നത് വഴി 5 മാസം വരെ സൂക്ഷ്മാണുക്കളുടെ മലിനീകരണം ഇല്ലാതെ സൂക്ഷിക്കാവുന്നതാണ്.

ഗ്രാമ്പൂ തൈലം അടങ്ങിയ കറ്റാർ വാഴയുടെ ജെല്ലിന്റെ സത്ത് നാരങ്ങ നീര്, ഓറഞ്ച് ജ്യൂസ്, തേൻ എന്നിവ 50:50, 75:25, 90:10 അനുപാതത്തിൽ ചേർത്ത മിശ്രിതം ഉപയോഗിച്ചാണ് ന്യൂട്രാസ്യൂട്ടിക്കൽസ് തയ്യാറാക്കിയത്. തുടർന്ന് പാസ്ചറൈസേഷൻ, ഫ്ലാഷ് കൂളിംഗ് എന്നിവ വഴി 6 മാസം വരെ സൂക്ഷിക്കാവുന്നതാണ്. രൂപവും നിറവും വിറ്റാമിൻ സിയും നാരങ്ങ നീര് (50 മില്ലി) + കറ്റാർ വാഴയുടെ ജെല്ലിന്റെ സത്ത് (50 മില്ലി) + 2 മില്ലി ഗ്രാമ്പൂ തൈലം എന്നിവയ്ക്ക് വളരെ കൂടുതലായിരുന്നു. ഗന്ധം, രുചി, മൊത്തത്തിലുള്ള സ്വീകാര്യത, പിഎച്ച്, ടിഎസ്എസ്, കാർബോഹൈഡ്രേറ്റ്, കലോറി

എന്നിവ തേൻ (50 മില്ലി) +കറ്റാർ വാഴയുടെ ജെല്ലിന്റെ സത്ത് (50 മില്ലി) + 2 മില്ലി ഗ്രാമ്പൂ എണ്ണ എന്ന മിശ്രിതത്തിന് ഉയർന്നതായി കാണപ്പെട്ടു. മൂന്നാം മാസം മുതൽ സൂക്ഷ്മാണുക്കളുടെ വളർച്ച കണ്ടെത്താനാകും. കറ്റാർ വാഴയിൽ നിന്നുള്ള പാനീയം തേനുമായി 50: 50 എന്ന അനുപാതത്തിൽ തയ്യാറാക്കി ഗ്രാമ്പൂ തൈലം ചേർത്ത് സൂക്ഷ്മാണുക്കൾ ഇല്ലാതെ രണ്ട് മാസം വരെ കേടുകൂടാതെ സൂക്ഷിക്കാവുന്നതാണ്.

സൂര്യപ്രകാശം, വായു, ഓവൻ, ശീതീകരണം എന്നിവ വഴി ഉണക്കിയെടുത്ത കറ്റാർവാഴയുടെ പൊടി വഴുവഴുപ്പുള്ളതായി കാണപ്പെട്ടതിനാൽ സ്വേദിതജലം വഴി പുനക്രമീകരിക്കാൻ സാധിക്കാത്തതോടൊപ്പം ശുദ്ധമായ ജെല്ലുമായി താരതമ്യപ്പെടുത്താൻ കഴിയാതെ വരികയും ചെയ്തു.

ശുദ്ധീകരിച്ച കറ്റാർ വാഴയുടെ ജെല്ലിന്റെ സത്ത് ഗ്രാമ്പൂ തൈലവുമായി (4 ശതമാനം)ചേർന്ന മിശ്രിതമാണ്കുറഞ്ഞ ചെലവിലുള്ള സ്ഥിരപ്പെടുത്തൽ പ്രക്രിയക്ക് അനുയോജ്യം എന്നും കൂടാതെ ഇവ ആരോഗ്യ പാനീയം തയ്യാറാക്കുന്നതിനുള്ള അടിസ്ഥാന ഘടകം ആണെന്നുമാണ് ഇപ്പോൾ നടത്തിയ പഠനം തെളിയിക്കുന്നത്. ദ്രവീകൃതമായ കറ്റാർ വാഴയുടെ ജെല്ലിന്റെ സത്ത് തേനിനൊപ്പം തുല്യ അനുപാതത്തിൽ കലർത്തി ഗ്രാമ്പൂ എണ്ണ ചേർത്ത്സംരക്ഷിക്കുന്നു. ഉയർന്ന കലോറി അടങ്ങിയിട്ടുള്ള രുചികരമായ ഈ പാനീയം രണ്ടുമാസം വരെ കേടുകൂടാതെ സൂക്ഷിക്കാവുന്നതാണ്. തണലിൽ ഉണക്കിയ കറ്റാർവാഴയുടെ കറ ഉയർന്ന അലോയിൻ അടങ്ങിയിരിക്കുന്നതിനാൽ വിപണനം ചെയ്യാവുന്ന ഒരു മികച്ച ഉപോത്പന്നമാണ്.