IMPACT OF SEED PROTECTANTS ON SEED LONGEVITY AND STORAGE PESTS IN RICE (*Oryza sativa* L.)

By

HARSHAPRADA K

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DEPARTMENT OF SEED SCIENCE AND TECHNOLOGY

COLLEGE OF HORTICULTURE,

VELLANIKKARA, THRISSUR - 680 656

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THESIS

Submitted in partial fulfilment of the requirement for the degree of

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2020

DECLARATION

I, hereby declare that this thesis entitled **'Impact of seed protectants on seed longevity and storage pests in rice (*Oryza sativa* L.)'** is a bonafide record of the research work done by me during the course of research and the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

Vellanikkara

Date: 17/12/2020

Harshaprada K

(2018-11-149)

CERTIFICATE

Certified that this thesis entitled **'Impact of seed protectants on seed longevity and storage pests in rice (*Oryza sativa* L.)'** is a bonafide record of the research work done independently by Ms. Harshaprada K 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.

Vellanikkara

Date: 17/12/2020

Dr. Rose Mary Francies

(Major advisor)
Professor and Head
Department of Seed Science and Technology
College of Horticulture,
Vellanikkara

CERTIFICATE

We, the undersigned members of advisory committee of Ms. Harshaprada K. a candidate for the degree of Master of Science in Agriculture with major field in Seed Science and Technology, agree that the thesis entitled **'Impact of seed protectants on seed longevity and storage pests in rice (*Oryza sativa* L.)'** may be submitted by **Ms. Harshaprada K. (2018-11-149)** in partial fulfilment of the requirement for the degree.

Dr. Rose Mary Francies
(Major Advisor)
Professor and Head
Department of Seed Science and Technology
College of Horticulture, Vellanikkara

15/12/20
Dr. Dijee Bastian
Professor (Plant Breeding and Genetics)
Department of Seed Science and Technology
College of Horticulture, Vellanikkara

Dr. Berin Pathrose
Assistant Professor
Department of Agricultural Entomology
College of Horticulture, Vellanikkara

Dr. Anita Cherian K
Professor and Head
Department of Plant Pathology
College of Horticulture, Vellanikkara

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LIST OF ABBREVIATIONS

CRD- Completely Randomized Design

KAU -Kerala Agricultural University

MC- Moisture Content

MSCS- Minimum Seed Certification Standards

MSL- Mean Sea Level

RH- Relative Humidity

SC- Soluble Concentrate

^{o} C- Degree

Celsius mm-

millimetre

g- gram

kg- kilogram

cm- centimetre

mg- milligram

ml- milli litre

Introduction

1. INTRODUCTION

Cereal crops grown world-wide, are the members of the grass family called Poaceae and are grown for their edible grain. The term cereal is derived from the word ‘Ceres’, meaning ‘the Roman goddess of harvest’, as they serve as one of the major sources of energy for human existence, they are categorized as food staples. Cereals are a rich source of carbohydrates, proteins, vitamins and minerals. The most familiar cereal grains are rice, wheat, maize, bajra, sorghum, ragi, oats and other minor millets.

Among cereals, rice (*Oryza sativa* L.) is the major food crop. The rice grain is consumed by over two-third of the population of the world, particularly the Asians. During 2018-19, in India, rice was cropped in an area of 43.90 million hectares registering a production of 107.80 million tonnes and a productivity of 2455 kg/ha (GOI, 2019), while, in Kerala, the area under rice during the corresponding period was 0.20 million hectares with a production of 0.57 million tonnes and a productivity of 2850 kg/ha (GOK, 2019).

Seeds are to be safely stored after harvest till the next sowing or until further use, such that the viability and vigour is maintained intact or reasonably high. Seed being a living entity, degradation in both quality and quantity, is inevitable during storage (Kapoor *et al*., 2010). Seed deterioration is the loss of seed quality, viability and vigour due to the effect of adverse environmental factors with time. The qualitative and quantitative loss during storage are caused by many physiological factors like moisture content, atmospheric relative humidity, temperature, initial seed quality, physical and chemical composition of seed, gaseous exchange, storage structure, packaging materials, seed production location and techniques, *etc* and also by pests and pathogens, which regulates the viability and vigour of the seed (Doijode, 1990). The post-harvest losses are reported to be high in developing countries. In India, as much as 50-60 per cent of cereal grains is lost during storage owing to inefficient storage conditions alone. Insect pests have been causing huge losses not only in the fields but also in post-harvest commodities during storage. The damage caused by insects may amount to 5-10 per cent in the temperate and 20-30 per cent in the tropical zone (Nakakitha, 1998) and may reach as high as 50 - 60 per cent (Kumar and Kalitha, 2017). The use of scientific storage methods can reduce these losses to as low as one to two per cent.

Safe storage of rice seeds over a long period of time has always remained a great challenge. Insect infestation results in loss of weight, nutritional value, physical quality and seed viability. Among the insects attacking rice in storage, rice weevil *(Sitophilus oryzae)*, lesser grain borer (*Rhizopertha domnica),* angoumois grain moth *(Sitotroga cerealella)* are the major primary pests and red flour beetle (*Tribolium castaneum*) and rice moth *(Corcyra cephalonica)* are the major secondary pest causing huge losses.

The rate of seed deterioration due to insect pests during storage, can be lowered either by storing the seeds under controlled conditions or by imposing various seed treatment techniques. Seed treatment is the application of physical, chemical and biological agents to the seeds during storage to suppress, control or repel pathogens, insects and other pests that attack seeds and reduce vigour and viability of seeds during storage (Chormule *et al*., 2018). The use of controlled conditions for seed storage is expensive and is not affordable to all farmers, especially in developing countries. Success in safe storage of seeds has been largely achieved by seed treatment. However, the indiscriminate use of pesticides has been found ecologically unsound and leads to health hazards (Tillman and Mulrooney, 2000).

The use of naturally occurring seed protectants and usage of bio-control agents are powerful alternative. Use of locally available plant materials to protect seeds from insect pests is a common age-old practice and the extracts from different plants have been known to possess insecticidal properties against wide range of pests (Isman, 2008). The pest-controlling efficacy of many plant derivatives has already been proved against several storage pests and they can improve the seed quality. A number of synthetic and natural components have been suggested as potential candidates for seed treatment. The negative environmental impact of such protectants in terms of insecticidal hazards, is almost non-existent and therefore could be beneficial to our agricultural sector. However, their efficiency as well as effect on seed quality remains to be observed (Kumar *et al*., 2007).

Keeping this in view, the present study was formulated with the following objectives:

- To assess the influence of seed protectants on the quality and longevity of rice seeds
- To assess the effectiveness of seed protectants against the storage pests.

Review of literature

2. REVIEW OF LITERATURE

Ensuring high crop productivity demands for a physically and genetically pure, physiological sound and pathologically clean seed. Seed being a living entity, it undergoes various cytological, physical, physiological and biochemical changes leading to the loss of viability and ultimate death (Jyothi and Malik, 2013).

Stored grains are attacked by insect pests during the storage. Protecting rice seeds from pests during storage has remained as a great challenge for both the farmers and seed growers. According to Upadhya and Ahmad (2011), the infestation can occur during the storage or in most cases, the infestation is carried forward to the storehouses from the infested field crops and spread rapidly.

Literature on the changes in seed quality during storage, the storage pests, their infestation and associated loss, various seed protectants used for management of these pests and their bio-efficiency, are reviewed below under the following:

2.1 Seed quality during storage

2.1.1 Germination and seed longevity

2.1.2 Vigour

2.1.3 Seed Moisture

2.1.4 Seed infestation

2.2 Insect infestation and loss during seed storage

2.2.1 Storage pests in rice

2.2.2 Management strategies to combat insect infestation

2.2.2.1 Bio-efficacy of botanicals against stored pests

2.2.2.2 Bio-efficacy of Spinosad against stored pests

2.2.2.3 Bio-efficacy of diatomaceous earth against stored pests

2.2.2.4 Bio-efficacy of microbial inoculants against stored pests

2.1. Seed quality during storage

Roberts (1973) classified the seeds based on their contrasting physiological responses of survival during storage. He introduced two categories of seeds *i.e.*, orthodox and recalcitrant seeds. Orthodox seeds can be dried to low moisture contents (2-5%) without damage and with decrease in seed storage moisture content and temperature in a quantifiable and predictable way, their longevity increases over a wide range of storage environments. In contrast, recalcitrant seeds cannot survive desiccation below a comparatively high (between 12% and 31%) moisture content. Rice shows orthodox seed storage behaviour, meaning that the seeds can be dried and stored at low temperature and at low moisture content (Hay and Probert, 2013).

2.1.1 Germination and seed longevity

Sl. No	Crop	Experimental details	Reference
1	Paddy	The germination and other seed quality parameters were low in seeds stored in cloth bags compared to those in super bags and polythene bags. Seed viability was maintained only up to four months in cloth bags, as against 10 months in the latter cases owing to less moisture fluctuation in the moisture impervious bags.	Yogalakshmi *et al.*, 1996
2	Seeds irrespective of crops	Seed longevity depends upon a number of factors such as the genetic constitution, initial seed quality, storage environment, packaging material and pre-storage seed treatments. It has been ascertained that pre-storage treatments protect the seed from microbial	Gupta 2003; Jakhar *et al.*, 2003

		infestation and also enhance the storage potential of seeds.	
3	Soybean	Polymer film coated seeds recorded higher germination than the untreated seeds because the seed treatment act as physical barrier, to reduce the leaching of organic substances from the seed coverings and may restrict oxygen diffusion to the embryo.	Vanangamudi *et al*., 2003
4	Onion Paddy	Polymer film coated seeds stored in super grain bags registered higher germination and vigour when compared to seeds stored in jute bags. Even, the viability of seeds stored under jute bags was lost within six months, whereas, it was maintained up to 16 months in the super bags. Results demonstrated that the highest electrical conductivity of paddy seed is related to its membrane disintegration and finally loss of viability	Mumtaz *et al*., 2004; Siddarudh, *et al*., 2016
5	Soybean	Polythene bag and metal tin minimize deterioration of seed stored in them by maintaining germinability to an appreciable level of 58.70 – 86.00 per cent at the end of 12 months of storage period unlike in bamboo bin and clay pot, wherein the germination dropped to zero after four months of storage.	Pessu *et al.*, 2005
6	Cereals	Seed germination was significantly correlated with the starch metabolism in seeds of cereals. A significant reduction of α- amylase activity and total soluble sugar content in aged seeds, results in reduced germination.	Garcia *et al.*, 2006; Goyoaga *et al.*, 2011

7	Wheat	The germination of wheat varieties decreased with increase in relative humidity and at high RH, the longevity was only two months	Miah *et al*., 2006; Rani *et al*., 2013
8	Maize; Paddy	Moisture impermeable containers were found having lower moisture content as well as higher germination per cent. The lower germination in moisture pervious containers may be due to the increased accumulation of moisture, high relative humidity and temperature prevailing during the storage period.	Gc, 2006; Choudhury *et al*., 2014
9	Rice	Seeds stored in polythene bags registered higher germination than those in cloth bag at the end of 12 months of ambient storage. Similarly, seedling dry weight and vigour index were higher in polythene bag. Loss of germination and vigour was lower in seeds stored in polythene bags because of less moisture fluctuation, unlike in those stored in moisture pervious cloth bag.	Patil and Shekargouda, 2007
10	Paddy	Seeds under cold storage recorded better seed quality, physiological and biochemical parameters over those in ambient storage under room temperature. Long term storage of seed without deterioration is possible in low temperature storage.	Gupta, 2010; Saidanaik and Chetti, 2018
11	Sorghum	Seeds stored at room temperature had a low germination (28.00%), whereas, seeds in aluminium cans recorded the highest germination (41.33%) after a period of nine months, due to air tight condition	Owolade *et al*., 2011

12	Wheat	Effect of different packing materials (metal bin, earthen bin, plastic bag, cloth bag and gunny bag) and grain moisture content (10.00% and 16.00%) showed that the viability of wheat seed was maintained for ten months in moisture proof containers	Chatta *et al*., 2012
13	Pigeon pea; Chick pea	When fungicide treated seeds were stored in moisture impervious containers, it resulted in higher germination and storability as it eliminated dampness, deterioration, microorganisms and enhance the seed longevity.	Basavegowda *et al*., 2013; Shivagouda *et al*., 2014;
14	Soybean	Seeds stored in polythene bags recorded higher germination and retained viability up to eight months, against those in cloth bag storage, wherein the seeds lost viability within two months.	Verma and Verma, 2014
15	Rice	Negative correlation between seed germination per cent and high temperature and moisture of the grain during storage was observed, due to increase in enzyme activity at high moisture	Hussain *et al*., 2015
16	Rice	Seeds stored in cloth bags recorded the least germination and all other seed quality parameters throughout the storage period compared to those stored in polythene and super bags. At the end of 10 months, the germination per cent was 74.78 in case of cloth bag and the longevity of seeds was maintained only up to four months, whereas,	Sattigeri, 2015

		in polythene and super bag storage, the rice seeds maintained longevity up to 10 months.	
17	Lentil and Sorghum	Negative impact of high relative humidity and temperature on seed germination and viability was evident due to an increase in respiration and enzyme activity, resulting in loss of food reserves before germination.	Assefa and Srinivasan, 2016
18	Cluster bean	Treated seeds stored in super bags recorded high germination and viability up to 18 months; against those in cloth bags. This negative impact of stored fungi and insects when stored in cloth bags was attributed to high moisture absorption.	Umesha *et al*., 2017
19	Maize	The low MC in the hermetic storage significantly contributes in maintaining the germination, viability and vigour for more than a year, due to protection from biotic and abiotic factors of deterioration	Bhandari *et al*., 2017
20	Rice	The effect of different storage containers and storage conditions on the seed quality parameters was evaluated. Throughout the storage period of 18 months, irrespective the storage containers, the seeds in cold storage recorded better seed quality, physiological and biochemical parameters over those stored under ambient storage at room temperature. Among the containers, the seeds stored in vacuum packed bags maintained their quality with the least deterioration compared to samples stored in gunny and cloth bags, due to absorbance of more moisture in them.	Saidanaik and Chetti, 2018

21	Rice	When primed seeds were stored under ambient conditions of room temperature with high humidity, all the germination attributes considerably decreased. The germination percentage, germination index and seedling vigour index within 15 days decreased by 39.40, 64.40, and 83.30 per cent respectively. When the storage duration extended up to 60 days, the germination percent, germination index and seedling vigour index of primed seeds stored under high humidity decreased by 88.90, 95.30, and 99.60 per cent, respectively as compared with un-stored primed seeds. It was observed that the viability of primed rice seeds did not reduce under low temperature and vacuum storage but was significantly reduced under room temperature with high humidity. Under vacuum condition, the increase of storage temperature (30°C) did not reduce the longevity of primed seeds	Wang *et al.*, 2019

2.1.2 Seed vigour

Sl. No.	**Crop**	**Experimental details**	**Reference**
1	Cotton	Improved germination and vigour, and lowered electrical conductivity values was observed in seeds coated with Polykote and Vitavax. This was attributed to slower imbibition rate and reduced imbibition damage	Struve and Hopper, 1996

2	Rice	Increased leakage of exudates during seed deterioration might be due to reduced metabolic activity of untreated seeds, which preceded accumulation of free radicles and toxic substances leading to loss of germinability and vigour. Electrical conductivity of seed leachate is inversely proportional to quality.	Agarwal and Kharlukhi, 1985
3	Paddy	Linear abatement in all parameters was observed as the storage period prolonged. Significantly higher seedling length, seedling dry weight and vigour index were registered in polythene bag storage and it was the least in jute bag. Rate of deterioration was significantly higher in moisture pervious packing materials than in semi or impervious ones, especially after 12 months of storage under ambient conditions	Maurya *et al*., 2002; Raikar *et al*., 2011
4	Soybean	Higher seed germination, seedling length and dry weight and vigour indices were recorded in HDPE bags, due to less electrolyte leakage and membrane integrity. Cloth bag is not safe for soybean seed storage for longer time because the rate of moisture migration was higher in cloth bag than in HDPE bags.	Singh and Dadlani, 2003; Agha *et al*., 2004; Autade and Ghuge, 2018
5	Soybean	As a result of suppression of seed borne microflora and maintenance of strong membrane integrity, the germination per cent, seedling vigour, field emergence and storability were high, when seeds were treated.	Sunilkumar, 2004; Verma and Verma, 2014
6	Tomato	Seed treatment increases seed germination and seedling vigour up to 12 months when stored in aluminium foil under ambient conditions	Vinitha, 2006

7	Soybean	The vigour index of seeds decreased with the advancement of storage period and reached to zero after two months of storage at 80.00 per cent RH. Long term storage with 92.00 per cent germination was possible at 50-60 per cent RH and low moisture (10 %) under room temperature	Miah *et al.*, 2006
8	Pearl millet	The longevity and vigour of seedlings could also be prolonged two-folds when the seeds were stored under LTLH conditions, owing to more stable moisture leading to less enzyme activity	Gupta, 2007
9	Lablab; Marigold	Low viability and reduced seedling vigour of untreated seed samples stored in cloth bags were associated with higher EC values of seed leachate. The EC values recorded for treated seeds were significantly lower than those of the untreated seeds.	Kathiravan *et al.*, 2008; Tejashwi *et al.*, 2014
10	Paddy	About 18 per cent increase in vigour was recorded in seeds stored under controlled conditions as against seed stored under ambient conditions. A 5.90 per cent increase in seed vigour was recorded in treated seeds stored in poly-lined bags as against those in jute bags.	Gupta, 2010
11	Wheat	Influence of storage temperature and relative humidity on grain moisture content during storage showed that the significant decrease in germination and vigour occurred only at elevated temperatures. Seeds stored at 40° C registered a greater decrease in germination and vigour compared to seed stored at 25° C	Sterlac *et al.*, 2010
12	Paddy	The per cent seed germination and vigour index decreased due to hard seed and rotten seed in	Ora *et al.*, 2011

		cultivated hybrid rice varieties. Rotten seed and post-emergence mortality of seedling were directly associated with seed borne pathogenic infection.	
13	Wheat	Wheat seeds stored in aluminium and polyester bags recorded higher seed germination and seedling vigour, due to less electrical conductivity. The rate of seed deterioration was low in these packing when compared to those stored in plastic and cloth bags.	Naguib *et al*., 2011
14	Wheat	Higher germination per cent, shoot and root length, seedling dry weight with lowest electrical conductivity and days to germination were evident in seeds stored in tin containers than in earthen pots. The trend was observed in the seeds stored in earthen pots was attributed to higher moisture content. This enhances seed deterioration and in turn reduces the quality of seed.	Nabila *et al*., 2016
15	Cluster bean	Seeds stored in super grain bags recorded higher seedling vigour indices when compared to those stored in cloth bag. A close association between the enhanced solute leakage and disintegration of membrane integrity leading to the loss of seed viability and vigour in cloth bag storage was obvious. This change can be significantly controlled by proper packaging	Umesha *et al*., 2017
16	Rice	Storing seeds treated with fungicides in polythene bag increased seed germination and seedling vigour at the end of eight months storage period as against the seeds stored in gunny bag.	Harsha *et al*., 2018

17	Maize and Pearl millet	Viability and vigour of pearl millet and maize seeds can be maintained up to 12 months when stored under low temperatures. There is no drastic decrease in germination, as low temperature and humidity will limit the seed moisture content and rate of respiration.	Rahmawati and Aquil, 2020

2.1.3. Seed moisture content

Sl. No.	**Crop**	**Experimental details**	**Reference**
1	Lentil	Moisture proof containers, particularly those made of polyethylene, are most suitable for storage of seeds, compared to moisture pervious containers.	Chowdhury *et al.*, 1990; Janmejai *et al.*, 1999
2	Seeds irrespective of crops	For safer storage of seeds, the moisture content of the grain and the surrounding environment should be reduced and monitored	Devereau *et al.*, 2002; Hayma, 2003; Jayas and White, 2003;
3	Paddy	Hermetic storage of paddy helps maintain germination of 85.00 per cent or more for periods up to nine months in moisture proof containers, while, conventional storage in jute bags reduces germination down to 14.00 per cent to 76.00 per cent within three months	Sabio *et al*, 2006; Villers *et al.*, 2006
4	Seeds irrespective of crops	Conventional packaging materials are porous in nature and even dried seeds can regain moisture in these packaging materials under high ambient relative humidity. Super Bags are made up of high strength polyethylene (PE) with barrier	Ben *et al.*, 2006

		layers and has very low oxygen (≤ 4 cc/m /day) and water vapour transmission rate (≤5 gm day) and can be used to store seeds safely, as they resist moisture.	
5	Wheat	Hermetic storage of seeds at or below 12.50 per cent moisture content resulted in safe storage of seeds up to four years without the degradation of seeds and germination remaining above 88.00 per cent compared to conventional storage in jute bags.	Villers *et al*., 2008
6	Paddy	USA organic cocoon, Germax cocoon, IRRI made storage bag, rexin cocoon and thick poly bag maintained lower seed moisture content below the critical level (14.00%) and killed more than 90.00 per cent insects of the stored grains, compared to polythene and gunny bag	Alam *et al*., 2009
7	Seeds irrespective of crops	Temperature and moisture content of cereal grains are the two key factors affecting the resulting quality of the seed, biochemical reactions, dry matter losses, allowable storage times and overall storage management of the grain	Gonzales *et al*., 2009; Lawrence and Maier, 2010
8	Seeds irrespective of crops	In most South Asian countries including Pakistan, seeds are stored in conventional porous packaging materials and earthen bins. The large pore size of jute, cloth and woven polypropylene bags provide free access to the water vapour that were readily absorbed by the seeds and ultimately elevated seed moisture contents	Chatta *et al.*, 2012

9	Soybean	Seeds stored in cloth bag deteriorate faster as compared to seeds stored in polythene bag. The seeds stored in cloth bag recorded high moisture content, low germination per cent, root and shoot length and seedling vigour index as compared to seeds stored in polythene bag	Monira *et al*., 2012
10	Maize	Seed moisture content increased in traditional packaging materials with increasing RH. At higher level of RH, moisture contents increased slightly (1%-2%) in super bag, whereas this increase was much higher in traditional packaging materials (>9.00% higher than original SMC at 90.00% RH).	Bakhtavar *et al*., 2020

2.1.4 Seed infection

Sl.No.	**Crop**	**Experimental details**	**Reference**
1	Seeds of all crops	Moisture content plays a significant role in the storage of grain. When grain has more moisture, it heats up and can have mold spoilage	Brewbaker, 2003; Alborch *et al*., 2011
2	Maize	Infection of seeds by storage fungus results in discoloration, dry matter loss, chemical and nutritional changes and overall reduction of seed quality	Fandohan *et al*., 2003; Chuck-Hernandez *et al*., 2012
3	Maize	Major fungi associated with storage are, *Aspergillus flavus* and *Fusarium* spp. Fungal growth in maize presents a major risk for humans and animals, as they produce mycotoxins, especially aflatoxin. Aflatoxin production by the fungi in the grain depends on	Egal *et al*., 2005

		the storage conditions, including relative humidity, temperature and storage period	
4	Seeds irrespective of crops	Drying foods and seeds sufficiently to be in equilibrium with atmospheres below 85.00 per cent, prevents accumulation of mycotoxins during storage. Lack of management of the dryness of stores is responsible for wide distribution of mycotoxins in the food and feed system	Magan and Aldred, 2007; Chulze, 2010; Darwish *et al.*, 2014
5	Maize	The ambient temperature and relative humidity in the subtropics and tropics adversely affects storage of seed. Seeds require adequate conditioning to the environment for successful storage. The degree of environmental conditioning needed is determined by ambient conditions, the kind of seed to be stored, its quality at the beginning of storage, and the length of the storage period.	Suleiman *et al.*, 2013
6	Seeds irrespective of crops	During storage, many physicochemical and physiological changes will occur in seeds due to the spoilage caused by storage fungi and infestation by stored pests. These result in complete disorganizations in cell organelles with time, and leads to seed deterioration.	Seadh *et al.*, 2015; Sadaka *et al.*, 2016
7	Paddy	Three fungal genera were found to be associated with the seed samples during storage i.e., *Aspergillus* (*A. flavus*, *A. niger*), *Curvularia* (*C. lunata*), *Fusarium* (*F. moniliformae*). The fungus *C. lunata* was found to be the most predominant, with a frequency of	Arvindbhai, 2016

		25.71 per cent followed by *F. moniliformae* (8. 10%).	
8	Rice	Traditional storage of seeds under ambient conditions resulted in the highest levels of insect and fungal infestations caused by rice weevil, *Alternaria* sp., *Nigrospora oryzae* and *Fusarium* sp., while the controlled conditions resulted in less infestation. High germination (87.84%) was recorded in seeds stored under controlled conditions and it was also found effective in maintaining the physical qualities of rice grain	Katta *et al.*, 2019

2.2 Insect infestation and loss during seed storage

Insect damages include direct consumption of kernels, detritus of exuviae, webbing, and cadavers. These makes the grain unfit for human consumption and also reduce quality and quantity. Insect infestation vitiates the storage environment by increasing the temperature owing to increased respiration by insects, result in development of hotspots which are congenial for the proliferation of storage fungi and other harmful microflora (Rajashekar *et al.*, 2012).

Abedin *et al*. (2012) reported a significant loss in stored rice through the activities of both biotic and abiotic factors. The average in-store losses that occurred for *Aus*, *Aman* and *Boro* rice were 3.68, 3,80 and 4.12 per cent respectively with an aggregated average of 3.92 per cent.

The infestation from pathogens in grain can lead to kernel discoloration and moldy odours, resulting in economic losses associated with a decrease in grain quality. In addition, certain species of fungus like *Aspergillus*, *Fusarium*, and *Penicillium* will produce certain mycotoxins, which can make the grain unfit for human consumption and also cause detrimental effects when fed to animals (Mylona *et al*., 2012). The growth of mould may also increase the grain temperature, thereby not only creating a more favourable environment for pest attack, but potentially increasing temperature to scorch or even ignite the grain.

The attack of *Sitophilus* sp. on maize cobs may start in the mature crop and subsequent infestations in storage (through the transfer of infested grain into storage or from the pest flying into storage facilities) may cause weight loss up to 30.00-40.00 per cent (Radha, 2014).

According to Bolarin and Bosa (2015), the biological (internal) causes of seed deterioration include changes in respiration rate, ethylene production, compositional changes in colour, flavour, texture and nutritive value, water stress, physiological disorders and pathological breakdown. The rate of biological deterioration also depends on several environmental (external) factors such as temperature, relative humidity, air velocity and atmospheric composition and sanitation procedures.

Generally, about 10.00 to 25.00 per cent of the grains stored world-wide are damaged due to the infestation from storage-grain pests. Loss in weight is not the only damage caused by storage pests, but they also seriously decrease nutrients, lowering seed germination rate, reducing quality, and lowering their marketing value due to the mass of waste, webbing, and insect cadavers (Abdel-Raheem *et al*., 2015).

Ali *et al.* (2016) investigated the quantitative losses caused by *Trogoderma granarium* and *Tribolium castaneum* in wheat, rice and maize seeds and reported 2.43 per cent and 4.02 per cent damage in maize, 3.99 per cent and 5.69 per cent in rice and 5.43 per cent and 7.96 per cent in wheat respectively by *T. granarium* and *T. castaneum.*

Ahmad *et al.* (2017) reported that, as the storage period increased, there was increase in kernel damage per cent of wheat grains. The weight loss in grains recorded was 20.00 per cent, 8.00 per cent and 2.50 per cent by *Sitophilus oryzae*, *Trogoderma granarium*, *Tribolium castaneum* respectively. Also, the loss in germination was observed up to six months of storage. Due to infestation of *S. oryzae*, *T. granarium*, *T. castaneum*, 45.40 per cent, 35.40 per cent and 18.00 per cent loss in germination was observed respectively.

Alam *et al.* (2019) reported that, in farmers traditional storage practices of maize seeds, grain damage (75.85 %) and weight loss (70.23 %) by weevils was observed. Grain damage ranged from 62.00 to 82.00 per cent, whereas, the weight loss varied from 51.00 to 85.00 per cent between three to six months of storage.

The kernel damage in wheat ranged from 3.60 per cent to 13.60 per cent and mean weight loss due to insects was 1.50 per cent, while the mean seed germination was only 72.30 per cent. Hence, it was concluded that wheat stored under farmers storage facilities experiences up to 14.00 per cent loss due to the insects and there was also significant loss of seed germination (Kalsa *et al.*, 2019).

A study was conducted to evaluate the impact of different storage conditions on seed quality characters of rice cultivars. The traditional storage under ambient conditions, resulted in the highest levels of insect and fungal infestations caused by rice weevil, *Alternaria* sp., *Nigrospora oryzae* and *Fusarium* sp., while the controlled conditions resulted in less infestation. The high germination (87.84%) per cent was recorded in controlled conditions and it was also effective in maintaining the physical qualities of rice grain (Katta *et al.,* 2019).

India possesses highly conducive environment for the year-round occurrence of the storage pests as most of the parts in India belong to tropical and sub-tropical climatic conditions. Number of stored pests get access to seeds during various stages of processing and the major source of infestation are old bags, old containers, storage structures and cross over infestation (Tyagi *et al*., 2019).

2.2.1 Storage pests in rice

The rice moth, *Corcyra cephalonica* Stainton (Lepidoptera: Pyralidae) is one of the most destructive secondary pests of stored cereals in Asia, Africa, North America and Europe causing severe economic losses. It attacks not only rice, wheat, corn and sorghum but also groundnut, cotton seeds, coffee, cocoa beans under storage. The larval stage of the insect cause damage to broken grains by feeding inside them and forming silken webbing. When infestation is high the entire stock of grains may be converted into a webbed mass, resulting in both qualitative and quantitative damage (Atwal and Dhaliwal, 2008).

Saw toothed grain beetle, *Oryzaephilus surinamensis* L. (Colioptera: Silvanidae) is considered as one of the main stored grain pests. Both the insects and larvae attack the grains and their products. It infests dried products, stored meat and also flour and medicines. Adults and larvae can be observed in all types of rice that have already been infected with pests (Al-Iraqi, 2010).

The red flour beetle (*Tribolium castaneum*) (Coleopteran: Tenebrionidae) is one of the most destructive pests and is widely distributed. Rust-red flour beetles have an extremely large craving for a variety of foods, such as food products stored in soils, warehouses, grocery stores, and houses including meal, crackers, beans, spices, pasta dried pet food, dried flowers, chocolate, nuts and seeds, and even dried museum specimens, but, they were particularly abundant in cereal grain products. When they occur in large number, they will secrete a chemical mixture that includes quinones, which are carcinogenic, thereby affecting product quality (Mishra *et al*., 2012).

The lesser grain borer, *Rhyzopertha dominica* (Coleoptera: Bostrichidae), is the sole species commonly found within stored cereal grains. A large amount of frass is produced from adult feeding activities and the presence of larvae exuvae, feces, fragments of immature insects, and various by products affects the overall quality of the stored products. Adult and larval stages of *R. dominica* feed on the germ and endosperm of the seed (Edde, 2012).

The rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), is one of the primary pests and is cosmopolitan in nature. It is the most destructive pest of the stored cereals and causes severe loss in rice, maize, barley and wheat during storage. The presence of weevil damage can be identified by the presence of hole on the seed and it can cause losses directly by feeding the grain or indirectly by producing the hotspots, which result in loss of moisture and thereby making grain more suitable for the attack of pests (Das, 2013).

Angoumois grain moth, *Sitotroga cerealella* (Lepidoptera: Gelechiidae), has the global distribution and causes loss in rice, wheat, corn, sorghum and pearl millet. The larvae are the damaging stage of the insect, feeding on internal content of the seed resulting in loss in weight. The moth emerges out by leaving an emergence hole on the seed coat (Ahmed *et al.*, 2014).

Stored grains are found to be attacked by a number of insect pests. These pests infest the seeds/grains to fulfill their food and shelter requirements, which results in qualitative as well as quantitative losses. A number of insect pests gain access to the seed storage at various stages of processing *i.e.*, during the process of development and maturation, in threshing yard, during transit or while in storage. Some of them start damaging the seeds at the ripening stage of crops and continue during storage. The spread and distribution are facilitated by movement of grains from one area to another either passively or by active fight of insect pests as some of the adult insects are strong fliers. These may destroy the grains and contaminate the rest with undesirable odours and flavours. Majority of insect pests belongs to orders Coleoptera and Lepidoptera (Srivatsava and Subramanian, 2016).

Stored insect pests may be divided into primary and secondary pests. Primary pests have the ability to attack the whole, unbroken grains, while the secondary pests attack only damaged grain, dust and milled products. Among the insects attacking rice in storage, rice weevil, *Sitophilus oryzae* (L.), lesser grain borer, *Rhyzopertha dominica* (F.), granary weevil, *Sitophilus granaries* (L.), Khapra beetle, *Trogoderma granarium* (Everts) are the major primary pests and secondary pests which cannot initiate infestation and infest the whole grain but feed on as broken kernels, debris, high moisture weed seeds, and grain damaged by primary pests, *viz*., rust-red flour beetle, *Tribolium castaneum* (Herbst), rusty grain beetle, *Cryptolestes ferrugineus* (L.), saw toothed grain beetle, *Oryzaephilus surinamensis* (L.) and rice moth, *Corcyra cephalonica* (Singh *et al.*, 2017).

Khapra Beetle, *Trogoderma granarium* Evarts (Coleoptera: Dermestidae) is one of the most dreaded pests of whole and ground cereals, oilseeds, maltings, copra and other foodstuffs. It has a capability to cause huge loss in stored grains through voracious feeding and heating of grains. The ability of its larvae to withstand starvation for up to three years as well as its ability to live on food with very low moisture content results in huge loss to the stored grains (Kavallieratos *et al.*, 2019).

2.2.2 Management strategies to combat insect infestation

The use of physical control measures against stored insect pest include, control of temperature, moisture and relative humidity in the storage place and in stored containers. However, it is highly uneconomical and difficult as it requires technical expertise. It also includes use of heat and cold treatments for the management of storage pests, as heat kills some pests while cold blocks their development. It is reported that a temperature below 4°C results in death of insects especially the immature stages of almost all insect pests. Before the lethal cold treatment, the insects are exposed to a temperature of 10–20°C for some days and death occurs rapidly at freezing point. However, in heat treatment, exposing the grains to 50°C for 2 hours eliminates most insect pests. Grain heating can be carried out using a hot-air fluidized bed, infrared radiation or high frequency dielectric and microwave heating, in order to achieve a uniform grain temperature throughout the storage structure (Shankar and Abrol, 2015).

The destructive activities of insects and other storage pests have been adequately subdued by synthetic chemical control methods. However, there are many problems associated with the use of these chemical, including high persistence, poor knowledge of application, increasing cost of application, pest resurgence, genetic resistance by insects, also they cause the lethal effects on non-target organisms and toxicity to the users (Ileke and Oni, 2011).

According to Selvaraj *et al*. (2012), the fumigation is an effective method. But, it cannot be practiced by farmers as the storage structures are not airtight and the structures commonly built inside the residential areas.

Patel and Joshi (2014) reported that the amounts of fumigants absorbed is greatly increased by the presence of fat in grain kernel and also the exposure period. As the duration of exposure period increases, its residual effect on germination, vigour and other qualities of seeds gets increases.

Many botanicals possess insecticidal as well as repellent properties with little or no mammalian toxicity. The use of plant products as seed protectants is a traditional method and is off great interest in the recent past. Surface persistence of some of these last for long time with no adverse effects on seed quality as well as cooking quality, besides they are easily available at low cost (Kelida *et al.,* 2015).

In addition, many chemicals are banned considering their adverse effect on environment. Hence, management of storage pests has become a challenge. In view of negative effects of synthetic insecticides, substances of plant origin for the control of stored grain insects are quite promising as they are more biodegradable, less toxic to human beings and safe to environment (Kaoud *et al*., 2013; Ahad *et al.*, 2016).

2.2.2.1 Bio-efficacy of botanicals against stored pests

Tiwari (1993) studied the efficiency of plant powder of *Acorus calamus* in wheat as a grain protectant against *Sitophilus oryzae*. The treatment was helpful in reducing the kernel infestation at 0.05 per cent concentration. He also evaluated its efficiency against *Rhyzopertha dominica*, in the laboratory. The powdered roots of *Acorus calamus* were observed to be effective at one per cent (w/w), in an initial test. However, more than 80.00 per cent suppression of the insects was observed in admixture of 0.25 and 0.50 (w/w).

A study was conducted by Chander *et al.* (2003) against the *Rhyzopertha dominica* using the rhizome powder of *Curcuma* sp. and mustard oil treatment in rice. The powdered dust form of rhizome was found to be effective against lesser grain borer and it supressed 0.50 per cent of the progenies. Mustard oil in combination with various levels of rhizome powder supressed the progeny by more than 92.00 per cent. Complete mortality of adult insects was reported in rice seeds treated with 6 ml of oil and 4 g of rhizome powder.

Park *et al.* (2003) studied the insecticidal activities of *Acorus gramineus* rhizome derived materials against adults of *Sitophilus oryzae* (L.), *Callosobruchus chinensis* (L.) and *Lasioderma serricorne* (F.) using direct contact application. In the filter paper diffusion test, it was observed that (Z)-asarone caused 70.00 per cent and

90.00 per cent mortality of *S. oryzae* adults at 0.064 and 0.255 mg/cm2 after four days of treatment, respectively. Cent per cent mortality was achieved seven days after treatment.

Umoetok and Gerard (2003) studied the insecticidal activity of *Acorus calamus* and two synthetic insecticides (Pirimiphos-methyl and Rotenone) against *Sitophilus oryzae*, *Tribolium castaneum* and *Rhizopertha dominica* in a laboratory. The results revealed that there was significant higher per cent mortality of *R. dominica* (83.22 %) in *A. calamus* treatment than in the insecticides and control. However, it was not found effective against *S. oryzae* and *T. castaneum.*

Yao *et al.* (2008) studied the insecticidal activity of rhizome derived materials of *Acorus calamus* L. against adults of *Sitophilus zeamais* M. by using repellency method and contact toxicity method. The active constituent is characterized as the (Z)- asarone. In the repellency test, ethanol extract of *A. calamus* showed 93.92 per cent repellency at 629.08 $\mu g/cm^2$, 12 hr after treatment. (Z)-asarone showed 84.50 per cent repellency at 314.54 $\mu g/cm^2$ and 77.02 per cent at 78.63 $\mu g/cm^2$, 12 h after treatment. In filter paper diffusion test, (Z)-asarone showed cent per cent and 15.56 per cent mortality at 40.89 $\mu g/cm^2$ and 15.73 $\mu g/cm^2$, respectively.

Govindan and Nelson (2009) studied the insecticidal activity of various plant powders on mortality and adult emergence of *Sitophilus oryzae* L. After 7 days of exposure, high mortality (99.10 %) was recorded in *V. negundo* followed by *A. officinarum* (96.60%), *N. speciosum* (94.40%), while it was only 46.60 per cent in untreated control. In another experiment, minimum adult emergence was observed in *A. indica* (18.00), *A. officinarum* (18.00), *G. superpa* (20.00), when compared to 98.00 adult weevils in untreated control. The grain weight loss was minimum (10.58%) in *A. indica* compared to untreated control (39.21%) after 45^{th} day of treatment.

Jadhav (2009) conducted an experiment to know the LC_{50} of plant extracts of *Acorus calamus*, Annona and Tephrosia against the larvae of *Corcyra cephalonica*. The toxicity effect of all the three plant extracts resulted in blackening and death of the larvae. Freshly emerged larvae were more sensitive to *A. calamus* and recorded 55.97, 39.62, 33.11 and 23.50 per cent mortality at 24, 48, 72 and 96 h respectively.

The chloroform extracts of leaf, stem bark, stem wood and roots of *Glycosmis pentaphylla* were tested for the repellency against *Tribolium castaneum* adults. The highest repellency was reported for leaf (60.99 %), followed by stem bark, stem wood and root bark have the lowest repellency. Except for the root, strong repellency was observed for all the other extracts (Pramanik *et al.*, 2009).

In India, it is an age-old practice to mix the seeds with dried leaves, especially, the neem leaves before the seeds being stored. Hameed *et al.* (2012) conducted an experiment to know the toxicological effects of neem on the red flour beetle and observed that 45.00 per cent mortality at 168 hr exposure time with 2.50 per cent concentration and 16.67 per cent with 0.50 per cent dose at 24 hr exposure time. LC_{50} and LT_{50} values of neem was also calculated. LC_{50} was found to be 6273 @ 24 hr, 4822 @48 hr, 566 @ 72 hr and 79.785 @ 168 hr. LT_{50} value of neem was 190 hr.

Seed treatment of paddy with neem seed acetone extract @ 0.16 per cent active ingredient was found to be effective against developmental stages of *Corcyra cephalonica* and caused 100 per cent larval mortality (Pathak and Tiwari, 2012).

Five different plants extract *viz*., garlic, allamanda, neem, chirata and bishkatali with two dilutions (1: 1 & 1 :2), was used for rice seed treatment to manage the seed microflora. Garlic extract (1I: 1) dilution was found to be the best. It successfully reduced seed-borne infection and increased seed germination. Neem (1: 1) and chirata (1: 1) were found to be next best to garlic extract (Ahmed *et al.*, 2013).

Yankanchi *et al*. (2014) evaluated the insecticidal and repellent activities of crude methanol extract of *Clerodendrum serratum* L. against the rice weevil, *Sitophilus oryzae* L. Results showed that the crude methanol extract was having moderate insecticidal efficacy against *S. oryzae*, with a maximum mortality of 63.00 per cent at 32 mg/mL (w/v) after seven days of exposure. The methanol extract was highly repellent against *S. oryzae*, ranging from 23.00 and 77.00 per cent at 30 minutes to two- hour exposure and from 47.00 to 97.00per cent at four to 24 hours of exposure.

Yang *et al.* (2015) studied the repellent activities of leaf and stem crude extracts of *Glycosmis lucida*, *G. craibii* var. *glabra*, *G. craibii*, *G. oligantha*, *G.*

pentaphylla and *G. esquirolii,* by using bio-assays on petri dishes against *Tribolium castaneum* and *Liposcelis bostrychophila*. The leaf and stem extracts of all the *Glycosmis* sp. possessed significant repellent activities against *T. castaneum* and the highest repellency was exhibited by *G. lucida* (94.00% and 84.00%) at two hour and four hour exposure time. The leaves and stem extracts of *G. pentaphylla* also registered insect attractant properties. Moreover, they also exhibited repellent activity against the *L. bostrychophila*

A study was conducted by Iqbal *et al.* (2015) to evaluate the effect of various extracts of *Curcuma* sp. on growth inhibition of *Tribolium castaneum* in wheat flour using ethanol extract of rhizome, revealed that minimum number of larvae were produced (8.00) at 1000 µg/g application rate.

Erenso and Berhe (2016) studied the effects of neem leaf and kernel powder against the maize weevil, *Sitophilus zeamais*, at the rates 1, 2 and 3 per cent w/w. Results revealed significant differences among various concentrations, with respect to the time of storage. At 1, 2 and 3 per cent w/w concentration rate, the seed kernel powder killed about 63.30, 70.72 and 82.00 per cent of adult weevils. Leaf powder treatment, however, killed 61.13, 68.76 and 77.75 per cent adults. At higher concentration, both the leaf powder and seed powder were effective in controlling weevils.

Sami *et al.* (2016) reported increased larval mortality up to 37.00 per cent and 10.00 per cent in case of adults of *Tribolium castaneum*, when treated with neem powder.

Sorghum seeds were treated with sweet flag rhizome powder against the rice weevil. It resulted in least damage to the seeds. The treatment resulted in the highest adult mortality (97.00%), minimum weight loss (0.40%) and also it showed maximum germination (99.00%), vigour index I (2030) and field emergence (90.00%), whereas the untreated showed least mortality (only 7.00%), more weight loss (18.00%) and reduced germination (36.00%) (Gadewar *et al.,* 2017).

Nair *et al.* (2017) evaluated the toxic effects of aqueous, ethanol and acetonic extract of *G. pentaphylla* against the rice weevil, *Sitophilus oryzae* at various dosages

(1.00, 5.00, 10.00, 15.00 and 20.00%). The results showed that, at higher concentration of the extract, the mortality of the insects was also high and the acetone extract exhibited the highest mortality of insect *i.e.*, 84±0.45, followed by ethanol extract (78±0.45) and then the aqueous extract (70±0.40).

The effect of plant powders against the rice weevil was evaluated in wheat seeds and it was reported that neem seed kernel powder at 2.00 per cent showed maximum inhibition rate of weevils (92.58%). The treatment also showed minimum grain damage (21.16%), weight loss (1.15%), adult emergence (16.17). The untreated control registered maximum grain damage of 21.16 per cent, 13.83 per cent of weight loss and maximum adult emergence (157.58 adults) (Singh *et al.,* 2017).

The efficacy of different plant products against rice moth was investigated and it was reported that the seeds treated with neem seed powder @ 2.5 g/250g of seeds showed highest larval (83.00%) and pupal mortality (10.00%) and also minimum emergence of adults (7.00%). In the same study, the treatment of seeds with neem leaf powder was also found to be effective and it showed 77.00 per cent, 9.00 per cent in larval and pupal mortality and 14.00 per cent in adult emergence against 82.00 per cent in untreated (Jhala *et al.,* 2018).

2.2.2.2 Bio-efficacy of Spinosad against stored pests

The insecticidal activity of Spinosad is that, it will act on insect's nervous system by ingestion or contact. It is reported to have low mammalian toxicity (Sparkes *et al.*, 2001).

Nayak *et al.* (2005) evaluated the effectiveness of Spinosad against the storage grain pests of wheat. The seeds were treated with 0.1, 0.5, 1.0 mg (a.i.)/kg of seeds for 14 days exposure period. Spinosad showed highest toxicity against *Rhyzopertha dominica* (100% mortality rate) at 1.0 mg (a.i.)/kg, followed by *Sitophilus oryzae* (88%) and the least mortality rate was observed in *Tribolium castaneum* and *Oryzaephilus surinamensis.*

Maize seeds treated with Spinosad @ 1.00 and 2.00 mg a.i./kg resulted in cent per cent mortality of *R. dominica* even after four months and 95.00 per cent mortality of *S. oryzae* with no progeny production and damage to the seeds, when adults were exposed to treated grains for 10 days every month up to four consecutive months. After four months, the efficiency was decreased and a few progenies of weevils were produced causing 5.00 per cent damage to seeds. No damage and progeny production were observed in case of *R. dominica* even after four months (Sharma and Michaelraj, 2006).

Subramanyam *et al*. (2007) studied the persistence and insecticidal activity of Spinosad against stored insect pests in hard red winter wheat. The seeds were treated with 1 mg (a.i.)/kg of grain at the time of storage. The study revealed complete mortality of *Rhyzopertha dominica* observed in treated seeds, whereas, mortality in the untreated grains, ranged from 3.00 per cent to 44.00 per cent. Mortality of *Tribolium castaneum* adults in Spinosad treated grain, ranged from 49.00 per cent to 82.00 per cent.

Hertlein *et al*. (2011) concluded that Spinosad effectively controls the adult and immature stages of economically important beetles and moth pests associated with stored grains. It is highly stable and can give protection from six months to two years.

Hameed *et al.* (2012) conducted an experiment to elucidate the toxicological effects of neem, kanair and Spinosad (Tracer 240 SC) on the red flour beetle at 0.5, 1.0, 1.5, 2.0 and 2.5 per cent concentration levels and observed that Spinosad (Tracer 240 SC) shown maximum mortality, 55.00 per cent at 2.50 per cent dose in 168 hr exposure time and minimum at 16.66 per cent with 0.50 per cent concentration at 24 hr exposure time. Neem showed 45.00 per cent mortality at 168 hr exposure time with 2.50 per cent concentration and 16.67 per cent at 0.50 per cent dose at 24 hr exposure time and concluded that the bio-pesticide Spinosad was best in controlling the storage pests.

Narayanaswamy (2013) studied the biology of rice weevil and effect of different packaging materials impregnated with insecticides and found that HDPE bags treated with Spinosad 45 SC @ 100 ppm a.i. was most effective in managing the adult weevils.

Insecticidal efficacy and degradation of Spinosad 45 SC, Indoxacarb 15 SC @ 0.5, 1.0 and 3.0 mg (a.i.)/kg against *Trogoderma granarium, Rhyzopertha dominica* and *Sitophilus oryzae* was studied by treating wheat seeds. Seeds treated with Spinosad @ 1.0 and 3.0 mg (a.i.)/kg recorded cent per cent mortality of *R. dominica* up to 120 days. In case of *S. oryzae*, cent per cent was observed up to 90 and 30 days at 3.0 and 1.0 mg (a.i.)/kg, respectively (Pandey *et al.,* 2016).

Ajaykumar *et al.* (2018) conducted an experiment to study the residual toxicity of insecticides against *R. dominica* in stored maize under ambient storage condition. Observations revealed that Spinosad 45 SC @ 2ppm, recorded the highest mortality of 80.00 per cent compared to all other treatments and was effective in managing the insects up to nine months.

2.2.2.3 Bio-efficacy of diatomaceous earth against stored pests

The insecticidal activity of diatomaceous earth depends on its capacity to damage insect's cuticle. The action of dust was reduced at high relative humidity (>70.00 %). The mode of action of diatomaceous earth formulations is that it absorbs cuticular waxes from the epicuticular surface, causing abrasion of the cuticle, leading to death of insects due to desiccation (Korunic, 1996).

The rice seeds were treated with diatomaceous earth formulation, Fossil Shield against the infestation from *Tribolium castaneum* by Mewis and Ulrichus (2001). Results showed that at 1 g and 2 g concentrations, the population of beetles was significantly reduced to a co-efficient of 0.27 and 0.02 respectively after 42 days of exposure. The mortality rate of adult beetles was highest at 0.50g of Fossil Shield. Death of stored product insects treated with diatomaceous earth decreased with increased relative humidity, due to reduced transpiration through the cuticle. High relative humidity >60.00% can prevent the drying action of diatomaceous earth.

The diatomaceous earth formulations are inert and have low mammalian toxicity. They leave no toxic residues on grains and can give long-lasting protection to grains (Vayias *et al.,* 2006; Athanassiou *et al.*, 2007).

Kostyukovsky *et al*. (2010) conducted a study to evaluate the sensitivity of the stored insect pests to the commercial formulation of diatomaceous earth (Inerto) at concentration of 0.5, 1.0, 2.0 and 4.0 g per kg of wheat grains. At lower concentrations, the mortality of *S. oryzae* and *O. surinamensis* were 92 and 86 per cent respectively, whereas the highest mortality of *T. castaneum* and *R. dominica* was observed at 4 g/kg of DE, after nine weeks of treatment.

Inert materials like activated clay, kaolinite and diatomaceous earth have been found to be effective in controlling pulse beetle. Among these, diatomaceous earth was found to cause high mortality rates in the adult populations of *Callosobruchus maculatus* (Neda *et al.,* 2012).

Arthur and Fontenot (2013) evaluated the efficacy of Alpine, a formulation containing dinotefuran and diatomaceous earth as aerosol spray and a dust with diatomaceous earth at the rate of 5 g/m^2 and 10 g/m^2, against six stored insect species. The mortality of *T. castaneum, R. dominica* and *O. surinamensis* generally increased with exposure time and 90.00 per cent or more was observed after three days of exposure at both the dust rates.

Wakil *et al.* (2013) evaluated the effects of combining Thiamethoxam @ 0.25, 0.5 and 0.75 mg/kg of active ingredient with the diatomaceous earth (DE) formulation, Silico Sec @ 100 mg/kg against lesser grain borer. The bioassay showed that there was greater mortality in population and production of progeny of borers in treated seeds compared to the control.

Athanassiou *et al.* (2016) conducted an experiment in order to assess the insecticidal effect of a diatomaceous earth formulation (SilicoSec) against *Sitophilus oryzae* and *Tribolium confusum* on stored wheat @ 0.25, 0.50, 100 and 1.50 g/kg seeds. The effect of DE resulted in cent per cent mortality after seven days of exposure. After 14 days, all weevils were dead except at lowest concentration (0.25 g) and at 0.50 g, only 80.00 per cent were killed. In case of beetle, in all concentrations, significantly less number of adults were dead compared to weevil, whereas, after 14 days of exposure cent per cent mortality was recorded at two higher doses. Results revealed that DE can be successfully used as good alternatives to residual insecticides in stored grain.

The effectiveness of an improved form of diatomaceous earth, Grain-Guard was evaluated against *T. castaneum* and *R. dominica* on four grain commodities, wheat, rice, maize and sorghum. After 14 days of treatment, more than 80.00 per cent mortality of adults was observed on wheat than in other grains. During the first 60 days of exposure, adult mortality increased, later on, the steady decrease in mortality was observed (Saeed *et al.,* 2018). Later, cent per cent adult mortality was observed on wheat at 100ppm and on rice at 150ppm.

2.2.2.4 Bio-efficacy of microbial inoculants against stored pests

The virulence of ten isolates of *Beauueria bassiana* was tested against maize weevil, *Sitophilus zeamais* in the laboratory. All the tested isolates were capable of infecting *S. zeamais*. A total of five (I89-481, 190-520, W-477, 190-533, and 194-907) highly virulent, three (192-736, X93-906 and 192-761A) intermediate and two (I93-868 and 193-870) weakly virulent isolates were identified for weevil. Mortality of beetles depended on the conidial concentration and the lowest mortality of 88.00 per cent was recorded in 10^4/ml. The highest proportion of damaged seeds within the 14 days of the test period was recorded in the control treatment (Adane *et al.*, 1996).

The effect of fungal entomopathogen, *Beauveria bassiana* as a seed protectant in wheat was investigated by Padin *et al.* (2002). The results showed the significant reduction in storage pest infestation in treated samples when compared with untreated control. The weight loss due to insect feeding reduced by 81.50 per cent when compared to the untreated.

Blanc *et al.* (2002) carried out the bioassay to evaluate the insecticidal activity of Bt *tenebrionis* isolates on the activity of cigarette beetle (*L. serricorne*) and the screening dose of 10 µg/mg showed the maximum larval mortality of 83.00 per cent after seven days of exposure.

Cherry *et al*. (2004) evaluated twelve indigenous and exotic isolates of *B. bassiana* and *M. anisopliae* for their virulence and their ability to suppress populations of *C. maculatus* F. in stored cowpea. They reported that *B. bassiana* was the most virulent isolate followed by *M. anisopliae*.

Oluwafemi *et al.* (2009) studied the insecticidal property of *Bacillus thuringiensis* and a natural enemy, *Habrobracon hebetor* against the cereal stored pest *Plodia interpunctella*. The results showed that *Bt* or *H. hebetor* alone caused 41.67 per cent and 35.35 per cent larval mortality of *P. interpunctella* respectively, whereas, the Bt–parasitoid combined treatment significantly increased mortality of *P. interpunctella* (86.00%).

Sabbour and Solieman (2014) evaluated the effect of entomopathogenic fungus *Beauveria brongniartii* against the storage pests in sugar beet. Results indicated that the number of eggs laid /female significantly decreased to 22±1.7, 33±8.9, and 35±2.6 individuals for *P. cruciferae*, *P. hyoscami*, and *Cassida vittata* respectively as compared to 266±8.7 individuals in the control.

In a study conducted by Magda and Moharam (2014) using five bacterial strains of *Bacillus sp*. against rice weevil, application of bacteria on foam covering gunny bags provided promising oviposition deterrency and toxicity, suppressing weevil infestation.

Mallik and Raisat (2014) studied the combined effect of locally isolated *Bacillus thuringiensis* and turmeric powder on red flour beetle (*Tribolium castaneum*). It was concluded that isolates of *B. thuringiensis*, proved to be an effective biopesticide against the *T. castaneum,* as it resulted in high mortality of larvae and adults. Turmeric powder also showed its bio-pesticidal activity, but the response was slow. The combined insecticidal effect of turmeric powder and three *B. thuringiensis* isolates was more effective as compared to their individual effect.

In a study conducted by Bello *et al.* (2017), it was reported that in wheat seeds treated with *Beauveria bassiana,* there was a significant control of live insects of *Tribolium castaneum* and *Rhyzopertha dominica* up to the four months of storage. In addition, the loss in grains was lower in treated seeds compared to the control and chemical seed treatment.

Materials and methods

3. MATERIALS AND METHODS

The present study was carried out in the Department of Seed Science and Technology, College of Horticulture, Vellanikkara during 2019-2020. It envisaged to assess the influence of seed protectants on the quality and longevity of rice seeds as well as to assess their effectiveness against the storage pests. The details of materials and methods used during the course of the work are described below.

3.1 Location

The study was conducted in the Department of Seed Science and Technology, College of Horticulture, Kerala Agricultural University (KAU), Vellanikkara P.O., Thrissur. Vellanikkara is located 40 m above MSL at 10^{0} 54’ North latitude and 76^{0} 28’ East longitudes and experiences a warm humid tropical climate with relative humidity remaining above 75 per cent for most of the year.

3.2 Climate

The climatic conditions that prevailed during the experimental period is summarised in Fig. 1 and Fig. 2. During the study period, relative humidity (RH) varied between 73 per cent (December, 2019) and 96 per cent (August, 2019). Total rainfall received during the study period was 3042.9 mm, ranging between 4.4 mm (Dec, 2019) and 977.5 mm (Aug, 2019). The monthly mean maximum temperatures ranged from 29.50 °C in August 2019 to 34.1 °C in January 2020, while, the mean minimum temperature varied between 21.4 °C in October 2019 and 22.4°C in January 2020.

3.3 Experimental material

The experiment was conducted with the red kernelled rice variety Jyothi that was harvested during 22nd March 2019. Owing to the unavailability of seed protectants and inability to bring down the seed moisture content to less than 12 per cent after treatment with protectants due to unfavourable environmental conditions, the storage experiment commenced on 13th June 2019, *i.e.,* two-and- a-half months after harvest.

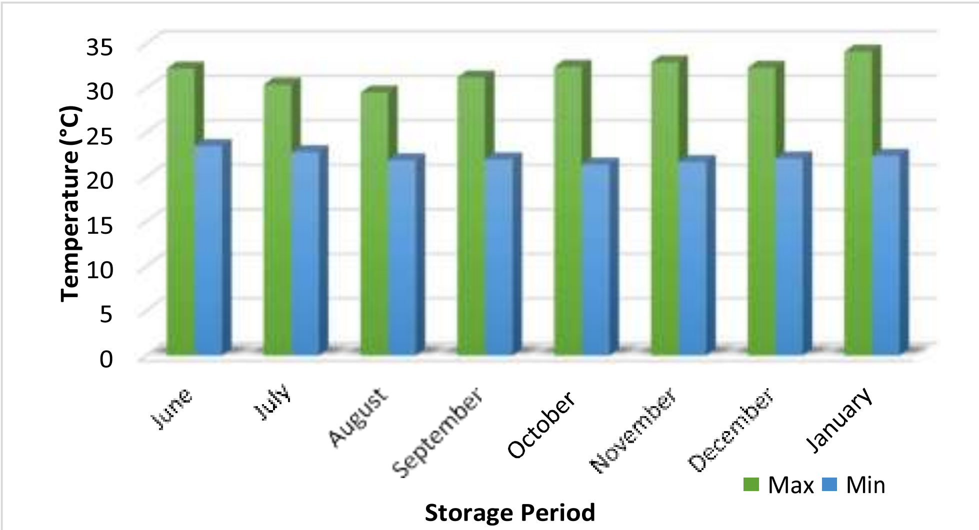


Fig 1. Mean maximum and minimum temperatures (^{O}C) experienced during the study period (June 2019 to January 2020)

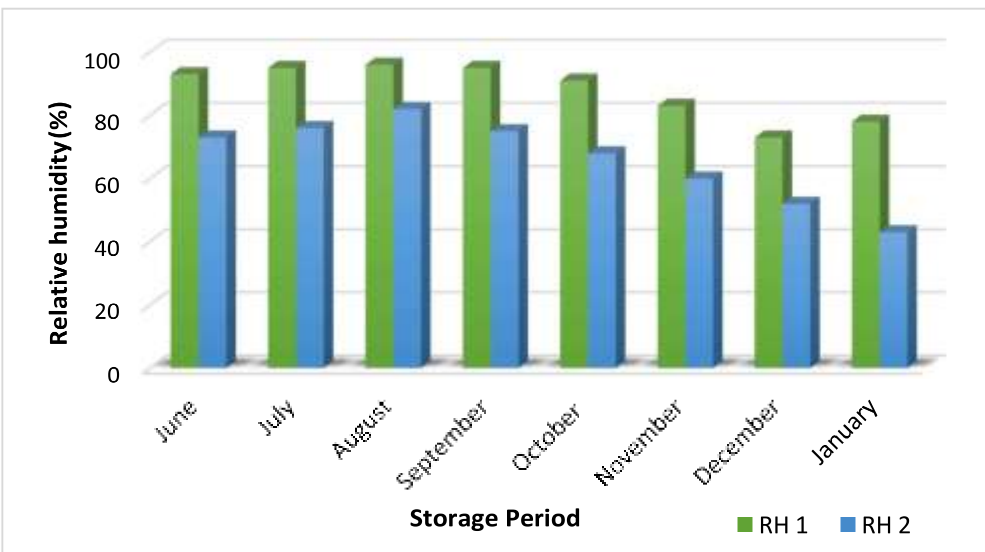


Fig 2. Relative humidity (%) experienced during the study period (June 2019 to January 2020)

3.4 Experimental method

The storage experiment was conducted following a completely randomized design (CRD) with ten treatments and three replications.

3.4.1 Treatment details

The seed protectants were selected on the basis of their reported insecticidal activity against the storage pests in rice. Rice seeds were treated with seed protectants at the rate listed in Table 1. Untreated seeds served as control (Plate 1).

Table 1: Treatment details

Treatment	Common name	Dose/kg of seed
T_1	Neem (*Azadirachta indica*) leaf powder	10 g
T_2	Neem (*Azadirachta indica*) seed kernel powder	10 g
T_3	Sweet flag (*Acorus calamus*) rhizome powder	10 g
T_4	Manja koova (*Curcuma angustifolia*) rhizome powder	10 g
T_5	Panal (*Glycosmis pentaphylla*) leaf powder	10 g
T_6	Spinosad	10 ppm
T_7	Diatomaceous earth	5 g
T_8	*Beauveria bassiana*	1×10^8 spores.ml^{-1}
T_9	*Bacillus thuringiensis*	1×10^8 spores.ml^{-1}
T_{10}	Untreated (control)	-

3.4.2 Procurement and preparation of seed protectants

Fresh leaves of neem (*Azadirachta indica* L.) and panal *(Glycosmis pentaphylla),* kernels of neem and rhizomes of sweet flag (*Acorus calamus* L.) and manja koova (*Curcuma angustifolia),* were collected and air-dried in the shade. They were powdered separately in an electric grinder and the resulting powder was passed through a 25-mesh sieve to obtain a fine dust to treat the seeds. Diatomaceous earth was collected from the Department of Seed Science and Technology, College of Horticulture, Vellanikkara.

The commercial formulation of novel insecticide Spinosad (Taffin 45 SC) was procured locally. The required dosage 10 ppm Spinosad was prepared by dissolving 4.44mg of Taffin in 200ml of water. This solution was sufficient to treat one kg of seeds. The entomopathogenic fungus *Beauveria bassiana* @ 1×10^8 spores.ml^{-1} was obtained from All India Co-ordinated Research Project on Biological Control of Crop Pests and Weeds (AICRP on BCCP & W), College of Horticulture, Vellanikkara and the biocontrol agent *Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1} was obtained from Department of Microbiology, College of Horticulture, Vellanikkara.

3.4.3 Procedure of seed treatment and storage

Freshly harvested rice seeds were dried to < 12 per cent moisture. The dried seeds in lots of 10 kg were treated with seed protectants given in Table. 1 by mixing the required quantity of seed protectants. Except in treatments T_6 (Spinosad @ 10 ppm), T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}) and T_9 (*Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}), the seeds were dry-dressed. In T_6, T_8, and T_9, the seeds were re-dried to < 12 per cent moisture content after treatment. The seeds (10 kg) were packed in jute bags and sealed. Each of the three replicates of a treatment, comprised of 30 such bags. Both treated and untreated seeds were stored under ambient conditions for a period of 12 months.

3.5 Observations recorded

The seed quality parameters were assessed before and after treatment with seed protectants. Subsequently, seed samples were drawn and tested for quality parameters following standard procedures at monthly intervals until the germination of seed falls below minimum standards for seed certification (MSCS <80%). However, seed microflora infection was assessed at the start and end of storage. The procedure followed with respect to recording of various observations is enumerated below.

3.5.1 Germination (%)

Germination test was conducted by adopting the top paper method as per ISTA (1965). Moist filter paper was used as the substratum. Seeds were placed on a layer of moist filter paper placed in Petriplates. These Petriplates were covered with lid and placed inside the germination cabinet at a temperature of 25±2°C and RH 90±3%. First count (%) and final count (%) was taken for each treatment of every replication. The first count was taken on 5^{th} day, while final count was taken on 14^{th} day from the day when germination test was performed. On 14^{th} day, the total number of normal seedlings were counted and expressed in per cent.

$$\text{Germination (\%)} = \frac{\text{Number of seeds germinated}}{\text{Total number of seeds kept for germination}} \times 100$$

3.5.2 Seedling root length (cm)

Ten normal seedlings were selected randomly from each replication of a treatment at the end of the germination test and the root length was measured from the collar region to the tip of the primary root. The mean root length was expressed in centimetre (cm).

3.5.3 Seedling shoot length (cm)

Ten normal seedlings used for measuring the root length were used to measure the shoot length. The shoot length was measured from the base of the primary leaf to collar region. The mean shoot length was expressed in centimetre (cm).

3.5.4 Seedling dry weight (mg)

The seedling dry weight estimation was done with the same ten seedlings used for the measurement of shoot and root length. They were taken in properly labelled butter paper bags and kept in hot air oven maintained at 80°C temperature for 24 h. The butter paper bags were then transferred to a desiccator to cool for 30 minutes. The weight of ten seedlings was recorded using a digital weighing balance and the average expressed in milligram (mg).

3.5.5 Vigour index

It is a secondary trait computed using the data recorded on germination (%) and seedling dry weight (g) or shoot length (cm).

3.5.5.1 Vigour index- I (VI-I)

Seedling vigour index-I was calculated as per the formula given by Abdul-Baki and Anderson (1973).

Vigour index-I = Germination (%) x Seedling length (cm)

3.5.5.2 Vigour index- II (VI-II)

Seedling vigour index II was calculated as per the formula given by Bewly and Black, (1994).

Vigour index-II = Germination (%) x Seedling dry weight (mg)

3.5.6 Moisture content (%)

Moisture content (MC) of the seed was measured during the storage period by using the low constant temperature procedure advocated by ISTA (1985). Five-gram seeds from each replication of each treatment were drawn from the sample and evenly distributed over the surface of the container made of non-corrosive glass of approximately 0.5 mm thickness. Both the container and its cover were weighed before and after filling. It was then placed in a hot-air oven maintained at 103±2 °C and dried for 17±1 h. The drying period was considered to have begun from the time when oven reaches 103 °C. At the completion of the prescribed time, the container was removed from the oven and placed in a desiccator to cool for 30-45 minutes. After cooling, the container along with its cover was weighed and the seed moisture content (%) was calculated using the following formula:

$$\text{Moisture content (\%)} = \frac{M2-M3}{M2-M1} \times 100$$

Where,

M1: weight of container with lid

M2: weight of container with lid + seeds before drying

M3: weight of container with lid + seeds after drying

3.5.7 Weight of damaged and undamaged seeds (g)

Two replicates of seed sample weighing 100 g from each replication of a treatment were drawn and the insect-damaged seeds (seeds with bored holes) were separated from the undamaged seeds and the weight of both the seeds were taken using weighing balance and the average expressed in grams.

The per cent reduction in weight of undamaged seeds was calculated using the formula:

$$\text{Reduction in weight of undamaged seeds (\%)} = \frac{\text{Initial weight} - \text{Final weight}}{\text{Initial weight}} \times 100$$

$$\text{Fold change in the weight of damaged seeds} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Initial weight}}$$

3.5.8 Number of beetles and number of larvae

From the sample drawn from each replication as described under 3.5.7. the number of beetles (red flour beetle, khapra beetle and rice weevil) and the number of larvae infesting the rice seeds were counted and the average expressed in numbers.

3.5.9 Number of infested seeds

From the samples drawn from each replication as described under 3.5.7. the number of seeds which are infested due to insect and pest attack was counted and the average expressed in numbers.

3.5.10 Seed microflora (%)

The seeds were examined for infection by storage fungi by using the standard moist blotter paper method and agar plate method at the start and end of the storage period, as recommended by ISTA (1999).

3.5.10.1 Blotter paper method

The detection of seed microflora was carried out by adopting ISTA's standard blotter test described by Neergaard (1979). Three layers of sterilized blotter papers were kept in sterilized petri plates. Sterilized water was added in the plates to soak the filter paper and the excess water was removed. Twenty-five seeds of rice were kept on the blotter paper equidistantly under aseptic conditions maintained by laminar air flow. After plating the seeds, the Petri dishes were incubated at 25±1°C under 12/12 hrs light and darkness cycle for 7 days. On the eighth day, the plates were observed for the presence of seed microflora under stereo binocular microscope. The number of infected seeds were counted and recorded in per cent. The fungal bodies were also identified based on the morphological characters of the conidia, conidiophores and fruiting bodies by making slides of the same.

3.5.10.2 Agar plate method

The agar plate method for detection of seed microflora requires surface sterilization. Potato Dextrose Agar (20 mL) was poured into sterilized Petriplates in aseptic conditions. The seeds were surface sterilized with 0.1% sodium hypochlorite and subsequently washed with sterile distilled water thrice. The washed seeds were then placed on sterile filter paper to remove the excessive water on its surface. After the media had set, the seeds were kept equidistantly in the PDA plate. The Petriplates were incubated under bell jar for six days. After incubation, the number of infected seeds were observed and recorded as per cent. The identification of the infection causing pathogen was also made by preparation of slide.

3.6 Statistical analysis

3.6.1 Analysis of data

Statistical analysis of the data on various seed quality parameters was performed following the factorial completely randomized design (CRD) with three replications, with various seed treatments (Factor A) and period of storage (Factor B), as per Fisher's method of analysis of variance (Gomez and Gomez, 1976). Square root transformation of data was done wherever applicable (Snedecor and Cochran, 1967).

3.6.1.1 ANOVA for Factorial design

The data recorded in the experiment was analyzed using three factorial ANOVA (CRD) so as to estimate the impact of seed treatments and period of storage on the dependent variables. It helps us to distinguish whether there are interactions between the different factors considered. The mean squares due to different sources of variation were worked out using the following analysis of variance (Gomez and Gomez, 1976).

Source	**df**	**Mean square**	**Expected mean squares**
Replication	(r-1)	M_r	M_r/M_e
Treatment	(2^n-1)	Mt	M_t/M_e
Main effect (A)	1	M_A	M_A/ M_e
Main effect (B)	1	M_B	M_B/ M_e
Factor (AB)	1	M_{AB}	M_{AB}/ M_e
Error	$(r-1)\ (2^n-1)$	M_e	-

The treatments were compared using the critical difference (C.D) estimate at P = 0.05.

3.6.1.2 Ranking of seed protectant treatments

Scoring of seed protectant treatments based on the longevity, mean germination (%), length of root and shoot of seedling, seedling vigour indices, moisture content (%) and storage infestation parameters studied was done according to a modification to the method suggested by Arunachalum and Bandyopadhyay (1984), for making decisions jointly on a number of dependent characters. High estimates of weight of undamaged seeds and seed quality attributes except seed moisture content were considered advantageous (P traits). However, high estimates for insect infestation parameters were considered disadvantageous (N traits). The seed protectant treatments were ranked in descending order of mean estimates for each P traits and vice-versa for each N traits the respectively based on the grouping obtained as per Duncan's Multiple Range Test (DMRT) test. Each group including the overlapping groups were assigned ranks. Individual score obtained by the seed protectant for each trait studied were added up to arrive at a total score. The protectants were then ranked in descending order of numerical values of total scores *i.e.,* the overall status of a seed protectant treatment was considered high if the total score across various parameters was low. Hence, the treatments with the least total score were ranked the best and the one that scored the highest was considered the most disadvantageous.

Plate 1: Seed Protectants used for treatment

Neem leaf powder

Neem seed kernel powder

Sweet flag rhizome powder

Manja koova rhizome powder

Panal leaf powder

Diatomaceous earth

***Baeveria bassiana* broth culture**

***Bacillus thuringiencis* broth culture**

Spinosad (Taffin 45 SC)

Results

4. RESULTS

Deterioration starts immediately after seeds attain physiological maturity and continues further, the rate depending upon several internal and external factors. Quality seeds are inevitable for good crop establishment. Safe storage of rice seeds over a long period has always remained a great challenge. Seeds have to be stored from harvesting to next planting season. Small and marginal farmers of tropics and subtropics, generally, use farm-saved seeds for planting in the subsequent season or store them for use in the next year. In India, especially in coastal states like Kerala, owing to sub-tropical conditions, maintenance of the quality of the seed throughout the storing period is always a challenge, as the conditions are highly conducive for the growth and proliferation of storage pests.

In consideration of the above, an attempt was made to evaluate the insecticidal efficacy of botanicals like neem leaf powder, neem seed kernel powder, sweet flag rhizome powder, manja koova rhizome powder, panal leaf powder, a novel insecticide Spinosad, inert diatomaceous earth, an entomopathogenic fungi *Beauveria bassiana* and a microbial agent *Bacillus thuringiensis,* against stored pests in rice and their effect on seed viability, seedling vigour and seed microflora. The experiment was conducted in the Department of Seed Science and Technology, Kerala Agricultural University (KAU), during 2019-2020.

The results obtained are enumerated hereunder.

4.1 Quality of seeds before storage

4.1.1 Quality of seeds before seed treatment.

The quality parameters of rice seeds of variety Jyothi used for the study, before subjecting to seed treatment are enumerated in Table 2.

The seed lot of variety Jyothi with a germination per cent of 95.33 per cent and a seed moisture content of 11.80 per cent, was used for seed treatment. The average root and shoot length and seedling dry weight of the seedlings was 11.20 cm, 10.50 cm and

0.25 g respectively. The vigour index I and vigour index II were 2068.00 and 2383.00, respectively. The seeds were also inspected for insect infestation. The weight of the damaged and undamaged seeds in 100 g of seed sample was found to be 1.30 g and 98.70 g respectively. The number of infested seeds and the larval and beetle population in 100 g seed sample were found to be 48.00, 0.00 and 2.00 respectively. Infection by seed pathogens and seed infection was absent.

Table 2: Quality of seeds before treatment with protectants in rice variety Jyothi

Sl. No.	Parameters	Value
1	Seed germination (%)	95.33
2	Seedling root length (cm)	11.20
3	Seedling shoot length (cm)	10.50
4	Seedling dry weight (g)	0.25
5	Vigour index I	2068.00
6	Vigour index II	2383.00
7	Seed moisture content (%)	11.80
8	Seed microflora (%)	0.00
9	Number of beetles/100 g	2.00
10	Number of larvae/100 g	0.00
11	Number of infested seeds/100 g	48.00
12	Weight of damaged seeds/100 g (g)	1.30
13	Weight of undamaged seeds/100 g (g)	98.70

4.1.2. Quality of treated seeds, before initiation of storage

The quality parameters of rice seeds after seed treatment are enumerated in Table 3a and Table 3b.

A marginal increase in germination per cent was observed in all instances after seed treatment, while a decrease was observed in untreated seeds (92.00%) during the same period. Among treated seeds, germination ranged from 95.00 per cent in T_4 (Manja koova rhizome powder @ 10 g/kg) to 96.60 per cent each in T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}) and T_9 (*Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}).

Germination was 96.30 per cent in T_2 (Neem seed kernel extract @ 10 g/kg), T_3 (Sweet flag rhizome powder @ 10 g/kg), T_6 (Spinosad @ 10pm) and T_7 (Diatomaceous earth @ 5 g/kg). Germination per cent in T_4 (Manja koova rhizome powder @ 10 g/kg: 95.00%) and T_{10} (Untreated control: 92.00%) had decreased after treatment. The untreated seeds registered the least germination.

In similar lines, variation in seedling root and shoot length was observed in treated seeds. Root length of seedling in treated seeds varied from 11.20 cm in T_1 (Neem leaf powder @ 10 g/kg), T_3 (Sweet flag rhizome powder @ 10 g/kg), T_5 (Panal leaf powder @ 10g /kg) and T_7 (Diatomaceous earth @ 5 g/kg) to 11.80 cm in T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}), while shoot length varied between 10.50 cm in T_1 (Neem leaf powder @ 10g.kg), T_3 (Sweet flag rhizome powder @ 10 g/kg), T_4 (Manja koova rhizome powder @ 10 g/kg) and 10.90 cm in T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}).

The seedling dry weight in treated seeds varied between 0.25g in T_1 (Neem leaf powder @ 10g.kg), T_4 (Manja koova rhizome powder @ 10 g/kg), T_5 (Panal leaf powder @ 10g /kg) and T_7 (Diatomaceous earth @ 5 g/kg), and 0.26g in (Neem seed kernel extract @ 10 g/kg), T_3 (Sweet flag rhizome powder @ 10 g/kg), T_6 (Spinosad @ 10pm), T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}) and T_9 (*Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}), while the untreated seeds registered 0.25g.

Vigour index I of treated seedlings varied between 2083.00 in T_1 (Neem leaf powder @ 10g.kg) to 2199.00 in T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}), while the untreated seeds registered a VI-I of 1935.00. Similarly, the seedling vigour index-II varied between 2400.00 in T_1 (Neem leaf powder @ 10g.kg) and T_5 (Panal leaf powder @ 10g /kg) to 2514.00 in T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}) and T_9 (*Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}). The untreated seeds had registered a VI-II of 2300.00.

The seed moisture content in treated seeds varied from 11.40 per cent in T_2 (Neem seed kernel powder @ 10 g/kg), T_5 (Panal leaf powder @ 10 g/kg) and T_7 (Diatomaceous earth @ 5 g/kg) to 11.80 per cent each in T_6 (Spinosad @ 10pm), T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}) and T_9 (*Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}), while the untreated seeds had a moisture content of 12.10 per cent.

Weight of undamaged seeds varied between 98.70g in T_1 (Neem leaf powder @ 10 g/kg) and T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}) and 99.00g each in T_2 (Neem seed kernel extract @ 10 g/kg), T_6 (Spinosad @ 10pm) and T_7 (Diatomaceous earth @ 5 g/kg).

Similarly, the weight of damaged seeds was found to varied between 1.00g in T_2 (Neem seed kernel extract @ 10 g/kg), T_6 (Spinosad @ 10pm) and T_7 (Diatomaceous earth @ 5 g/kg), and 1.30g in T_1 (Neem leaf powder @ 10 g/kg) and T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}). The untreated seeds registered 96.50g and 3.50g of undamaged and damaged seed, respectively per 100 gram of seed weight.

In T_2 (Neem seed kernel extract @ 10 g/kg) and T_7 (Diatomaceous earth @ 5 g/kg) the beetle infestation was absent, whereas, in all other treatments, one beetle each was observed. The number of beetles/100 g of seed sample was found to be two in untreated seeds. The number of infested seeds/100 g of seed sample varied between 41.00 in T_2 (Neem seed kernel extract @ 10 g/kg) and T_7 (Diatomaceous earth @ 5 g/kg), and 48.00 each in T_3 (Sweet flag rhizome powder @ 10 g/kg) and T_4 (Manja koova rhizome powder @ 10 g/kg). The untreated seeds registered 51 infested seeds/100 g seed sample.

Table 3a. Quality of treated seeds of rice variety Jyothi, before initiation of storage

Treatments (T)	Seed quality parameters							
	Germination (%)	Seedling root length (cm)	Seedling shoot length (cm)	Seedling dry weight (g)	Seedling vigour index-1	Seedling vigour index-II	Seed moisture content (%)	Seed microflora (%)
T_1	96.00	11.20	10.50	0.25	2083.00	2400.00	11.50	0.00
T_2	96.30	11.40	10.70	0.26	2128.00	2503.00	11.40	0.00
T_3	96.30	11.20	10.50	0.26	2089.00	2503.00	11.50	0.00
T_4	95.00	11.20	10.50	0.25	2061.00	2375.00	11.70	0.00
T_5	96.00	11.20	10.60	0.25	2092.00	2400.00	11.40	0.00
T_6	96.30	11.30	10.80	0.26	2128.00	2503.00	11.80	0.00
T_7	96.30	11.20	10.70	0.25	2108.00	2407.00	11.40	0.00
T_8	96.60	11.80	10.90	0.26	2199.00	2514.00	11.80	0.00
T_9	96.60	11.50	10.70	0.26	2149.00	2514.00	11.80	0.00
T_{10}	92.00	10.80	10.20	0.25	1935.00	2300.00	12.10	0.00

Table 3b. Quality of treated seeds of rice variety Jyothi, before initiation of storage

Treatments (T)	Seed quality parameters				
	Number of beetles/100 g	Number of larvae/100 g	Number of infested seeds/100 g	Weight of damaged seeds/100 g (g)	Weight of undamaged seeds/100 g (g)
T_1	1.00	0.00	45.00	1.30	98.70
T_2	0.00	0.00	41.00	1.00	99.00
T_3	1.00	0.00	48.00	1.10	98.90
T_4	1.00	0.00	48.00	1.30	98.70
T_5	1.00	0.00	43.00	1.10	98.90
T_6	1.00	0.00	42.00	1.00	99.00
T_7	0.00	0.00	41.00	1.00	99.00
T_8	1.00	0.00	43.00	1.30	98.70
T_9	1.00	0.00	42.00	1.20	98.80
T_{10}	2.00	0.00	51.00	3.50	96.50

There were no larvae in seeds and also seed infection was absent in both treated and untreated seeds.

4.2 Quality of treated seeds during storage

4.2.1 Analysis of Variance

Analysis of variance revealed the existence of significant difference in the impact of various seed protectants on seed quality parameters like germination, moisture content, seedling dry weight, vigour index I and II, during the storage period. Significant differences in weight of insect damaged seeds and undamaged seeds, number of beetles, larva and number of infested seeds, among the treatments, over the storage period, were also evident.

4.2.1.1 Germination (%)

The results on germination as influenced by the storage period, seed treatment and their interaction are furnished in Table 4.

Germination during the storage was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.1.1 Impact of seed treatment (T)

Irrespective of the storage period, germination varied significantly between the treatments. The highest mean germination was observed in T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}: 87.04%) and the least in T_1 (Neem leaf powder: 80.87%). T_8 was significantly superior to all other treatments. Treatment T_9 (*Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 84.08%) and T_7 (Diatomaceous earth @ 5 g/kg: 83.79%) were on par with each other and next best to T_8. T_5 (Panal leaf powder @ 10 g/kg: 83.35%), T_{10} (Untreated seeds: 83.17%) and T_2 (Neem seed kernel powder @ 10 g/kg :82.62%) were on par with each other. T_3 (Sweet flag rhizome powder @ 10 g/kg: 82.08%) and T_4 (Manja koova rhizome powder @ 10 g/kg: 92.26%) and seeds treated with Spinosad @ 10 ppm (T_6: 81.00%) were found to be on par with $T_{1,}$ which registered the least germination per cent.

4.2.1.1.2 Impact of storage period (S)

Irrespective of the seed treatments, germination varied significantly during the storage period.

Germination declined progressively and significantly as the period of storage increased. Germination per cent was the highest during the first month (S_1: 92.73%) of storage and the least at the end of storage period (S_7: 72.25%). Germination fell below the IMSCS (80%), five months after storage ($S_{5:}$ 80.28%).

4.2.1.1.3 Impact of interaction between Treatment (T) × Storage period (S)

There existed significant differences in germination owing to the interaction of seed treatment and storage period.

Germination declined progressively and significantly in various treatments over the period of storage. Germination in all the treatments were on par with each other during the initial period of storage *i.e.,* during S_1 and S_2. Thereafter, it was varied significantly between the treatments. Germination in T_8 (*Beauveria bassiana* @ 1×10^8 spores. ml^{-1}) varied from 96.23 per cent in S_1 to 78.67 per cent at S_7 and was found to be the highest throughout the storage period.

Germination in T_8 alone was above the minimum standards for seed certification (MSCS) at S_6 (T_8S_6: 80.64%) and thereafter, decreased to 78.67 per cent at S_7. At S_6, germination in treatment T_9 (T_9S_6: *Bacillus thuringiensis* @ 1×10^8 spores. ml^{-1}: 79.92 %), T_2 (T_2S_6- Neem seed kernel powder @ 10 g/kg: 78.32%) and T_5 (T_5S_6- Panal leaf powder @ 10 g/kg: 78.32%) were on par with $T_{8,}$ although the values were below IMSCS.

During the fifth month (S_5) of storage, germination in treatments T_1 (T_1S_5- Neem leaf powder @ 10 g/kg: 76.38%), T_3 (T_3S_5- Sweet flag rhizome powder @ 10 g/kg: 76.91%), T_6 (T_6S_5- Spinosad @ 10 ppm: 74.64%) and T_7 (T_7S_5- Diatomaceous earth @ 5 g/kg: 78.67%) fell below MSCS. Except these four, germination in all other treatments including the untreated seeds (T_7S_5-Control: 82.26%) was retained above 80.00 per cent at S_5.

Table 4: Impact of storage period, treatments and their interaction on seed germination (%) in rice

Treatments TR	Intervals of storage (MAS)							Mean
	S_1	S_2	S_3	S_4	S_5	S_6	S_7	
T_1	92.35^{uvwx} (9.61)	88.36^{qrstu} (9.40)	84.27^{mnoprq} (9.18)	81.72^{klmn} (9.04)	76.38^{efghij} (8.74)	73.61^{abcdefg} (8.58)	70.72^{abc} (8.41)	**80.87^{d}**
T_2	93.70^{vwx} (9.68)	89.30^{rstuvw} (9.45)	83.90^{mnopq} (9.16)	81.36^{klmn} (9.02)	80.64^{jklmn} (8.98)	78.32^{ghijkl} (8.85)	72.59^{abcdef} (8.52)	**82.62^{bc}**
T_3	94.09^{wx} (9.70)	90.63^{tuvw} (9.52)	87.42^{pqrstu} (9.35)	82.99^{lmnop} (9.11)	76.91^{fghijk} (8.77)	74.64^{bcdefgh} (8.64)	70.05^{a} (8.37)	**82.08^{cd}**
T_4	91.58^{tuvwx} (9.57)	89.30^{rstuvw} (9.45)	86.67^{opqrst} (9.31)	81.90^{lmno} (9.05)	81.72^{klmn} (9.04)	75.34^{defghi} (8.68)	70.39^{ab} (8.39)	**82.26^{cd}**
T_5	92.35^{uvwx} (9.61)	90.06^{stuvw} (9.49)	88.73^{qrstuv} (9.42)	82.26^{lmno} (9.07)	80.64^{jklmn} (8.98)	78.32^{ghijkl} (8.85)	71.74^{abcd} (8.47)	**83.35^{bc}**
T_6	91.01^{tuvw} (9.54)	89.68^{stuvw} (9.47)	87.23^{pqrstu} (9.34)	82.26^{lmno} (9.07)	74.64^{bcdefgh} (8.64)	72.59^{abcdef} (8.52)	70.72^{abc} (8.41)	**81.00^{d}**
T_7	91.96^{uvwx} (9.59)	91.39^{tuvwx} (9.56)	90.25^{stuvw} (9.50)	85.37^{nopqrs} (9.24)	78.67^{hijkl} (8.87)	74.99^{cdefgh} (8.66)	74.30^{abcdefgh} (8.62)	**83.79^{b}**
T_8	96.23^{x} (9.81)	93.70^{vwx} (9.68)	91.96^{uvwx} (9.59)	86.67^{opqrst} (9.31)	82.26^{lmno} (9.07)	80.64^{jklmn} (8.98)	78.67^{hijkl} (8.87)	**87.04^{a}**
T_9	92.35^{uvwx} (9.61)	90.63^{tuvw} (9.52)	88.36^{qrstu} (9.40)	84.08^{mnopq} (9.17)	81.72^{klmn} (9.04)	79.92^{ijklm} (8.94)	72.25^{abcde} (8.50)	**84.08^{b}**
T_{10}	91.58^{tuvwx} (9.57)	89.30^{rstuvw} (9.45)	87.42^{pqrstu} (9.35)	86.67^{opqrst} (9.31)	82.26^{lmno} (9.07)	74.64^{bcdefgh} (8.64)	71.40^{abcd} (8.45)	**83.17^{bc}**
Mean	**92.73^{a}**	**90.25^{b}**	**87.60^{c}**	**83.53^{d}**	**80.28^{e}**	**76.21^{f}**	**72.25^{g}**	

(Figures in parenthesis are square root transformed values)

Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)
MAS	0.040	0.020	0.014	TR	0.048	0.024	0.017	MAS × TR	0.126	0.064	0.045

4.2.1.2 Seedling root length (cm)

The results on impact of storage period, seed protectants and their interaction on seedling root length are furnished in Table 5.

Seedling root length was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.2.1 Impact of seed treatment (T)

Irrespective of the storage period, seedling root length varied significantly between treatments. Over the storage period, the root length varied between 9.52 cm (T_4- Manja koova rhizome powder @ 10 g/kg) and 10.90 cm (T_8 -*Beauveria bassiana* @1×10^8 spores.ml^{-1}). Root length in T_8 was significantly superior to all other treatments. T_9 (*Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 10.80 cm) and T_6 (Spinosad @ 10 ppm: 10.80 cm) were on par with each other and next best to T_8.

Seeds treated with T_7 (Diatomaceous earth @ 5 g/kg: 10.49 cm), T_2 (Neem seed kernel powder @ 10 g/kg: 10.29 cm), T_5 (Panal leaf powder @ 10 g/kg: 10.23 cm) were superior to T_{10} (Untreated control: 10.10 cm). T_3 (Sweet flag rhizome powder @ 10 g/kg: 9.71 cm), T_1 (Neem leaf powder @ 10 g/kg: 9.69 cm) and T_4 (Manja koova rhizome powder @10 g/kg: 9.52 cm) registered lower root length.

4.2.1.2.2 Impact of storage period (S)

Irrespective of the treatment, seedling root length varied significantly over the storage period.

Significant and progressive decline in seedling root length occurred in both treated and untreated seeds. The highest root length occurred during the first month (S_1: 10.86 cm) and the least was at the end of storage (S_7: 9.50 cm).

Table 5: Impact of storage period, treatments and their interaction on seedling root length (cm) in rice

Treatments (T)	Intervals of storage (MAS)							Mean
	S_1	S_2	S_3	S_4	S_5	S_6	S_7	
T_1	$10.07^{ghijklm}$	10.32^{lmnopq}	$10.07^{ghijklm}$	9.77^{efghi}	9.37^{bcd}	9.17^{ab}	9.12^{ab}	**9.69^{f}**
T_2	$10.67^{qrstuvwx}$	$10.60^{pqrstuvw}$	$10.53^{opqrstu}$	$10.30^{klmnopq}$	$10.30^{klmnopq}$	9.90^{fghij}	9.77^{efghi}	**10.29^{d}**
T_3	10.80^{stuvwx}	9.80^{fghi}	9.73^{defgh}	9.89^{fghij}	9.37^{bcd}	9.27^{b}	9.10^{ab}	**9.71^{f}**
T_4	$10.50^{nopqrst}$	9.90^{fghij}	9.70^{defg}	9.40^{bcde}	9.20^{b}	9.13^{ab}	8.80^{a}	**9.52^{g}**
T_5	10.83^{tuvwx}	10.83^{tuvwx}	10.73^{stuvwx}	10.23^{jklmno}	9.93^{fghijk}	9.76^{efgh}	9.28^{bc}	**10.23^{d}**
T_6	11.33^{zAB}	11.40^{B}	11.30^{yzAB}	10.97^{wxyz}	$10.57^{opqrstuv}$	10.23^{jklmno}	9.78^{fghi}	**10.80^{b}**
T_7	10.90^{uvwx}	11.00^{xyzA}	10.93^{vwxy}	$10.60^{pqrstuvw}$	10.13^{ijklmn}	10.08^{hijklm}	9.81^{fghi}	**10.49^{c}**
T_8	11.50^{B}	11.53^{B}	11.43^{B}	10.87^{tuvwx}	$10.53^{opqrstu}$	$10.37^{mnopqrs}$	$10.07^{ghijklm}$	**10.90^{a}**
T_9	11.43^{B}	11.43^{B}	11.37^{AB}	10.80^{tuvwx}	$10.33^{lmnopqr}$	$10.25^{jklmnop}$	$9.98^{fghijkl}$	**10.80^{b}**
T_{10}	$10.60^{pqrstuvw}$	$10.70^{rstuvwx}$	$10.50^{nopqrst}$	10.10^{hijklm}	9.83^{fghi}	9.65^{cdef}	9.30^{bc}	**10.10^{e}**
Mean	**10.86^{a}**	**10.75^{b}**	**10.63^{c}**	**10.29^{d}**	**9.96^{e}**	**9.78^{f}**	**9.50^{g}**	

Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)
MAS	0.055	0.028	0.020	TR	0.065	0.033	0.023	MAS × TR	0.173	0.087	0.062

4.2.1.2.3 Impact of interaction between Treatment (T) × Storage period (S)

Significant difference in root length of seedlings was observed due to interaction between seed treatment and storage period**.**

Root length varied between 11.50 cm in S_1 and 10.07 cm in S_7. Throughout the storage period, higher root length was recorded in T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}). The root length in T_8 in the first three months after storage were on par with each other (T_8S_1: 11.50 cm, T_8S_2: 11.53 cm, T_8S_3: 11.43 cm).

Root length in seeds treated with T_9 (*Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1} - T_9 S_1: 11.43 cm, T_9S_7: 9.98 cm) and T_6 (Spinosad @ 10 ppm- T_6S_1: 11.33 cm, T_6S_7: 9.78 cm) were on par with T_8 throughout the storage period. These treatments produced significant longer roots length over other treatments including the untreated seeds T_{10} ($T_{10}S_1$: 10.60 cm, $T_{10}S_{7:}$ 9.30 cm), during the storage period.

T_1 (Neem leaf powder @ 10 g/kg), T_4 (Manja koova rhizome powder @ 10 g/kg) registered shorter root length than T_{10} during storage. They differed significantly from T_{10} except during S_4 and S_7 in case of T_1 (T_1S_4: 9.77 cm; T_1S_7: 9.12 cm) and S_1 in case of T_4 (T_4S_1: 10.50 cm).

At the end (S_7) of the storage period, T_7 (T_7S_7- Diatomaceous earth @ 5 g/kg: 9.81 cm), T_6 (T_6S_7- Spinosad @ 10 ppm: 9.78 cm), T_2 (T_2S_7- Neem seed kernel extract @ 10 g/kg: 9.77 cm) T_8 and T_9 were on par with each other. Root length was least in T_4 (T_4S_7- Manja koova rhizome powder @ 10 g/kg: 8.80 cm), T_1 (T_1S_7- Neem leaf powder @ 10 g/kg: 9.12 cm) and T_3 (T_3S_7- Sweet flag rhizome powder @ 10 g/kg: 9.10 cm). T_5 (T_5S_7- Panal leaf powder @10 g/kg: 9.28 cm) and T_{10} ($T_{10}S_7$- Untreated control: 9.30 cm) were on par with each other.

4.2.1.3 Seedling shoot length (cm)

The results on the impact of seed treatments, storage period and their interaction on shoot length of seedlings are furnished in Table 6.

The shoot length of seedling was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.3.1 Impact of seed treatment (T)

Irrespective of the storage period, seedling shoot length varied significantly between treatments. The shoot length was maximum in T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}: 10.09 cm) and it was the least in T_4 (Manja koova rhizome powder @ 10 g/kg: 9.00 cm). T_8 exhibited significant superiority over other treatments. T_9 (*Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}: 9.94 cm) followed by T_6 (Spinosad @ 10 ppm: 9.65 cm) were the next best treatments. There was a significant difference between T_9 and T_6. T_{10} (untreated control: 9.40 cm), T_3 (Sweet flag rhizome powder @ 10 g/kg: 9.28 cm), T_1 (Neem leaf powder @ 10 g/kg: 9.13 cm) apart from T_4, recorded significant lower shoot length than others.

4.2.1.3.2 Impact of storage period (S)

Irrespective of the seed treatments, seedling shoot length varied significantly during the storage period.

Shoot length of seedlings in both treated and untreated control declined progressively as well as varied significantly over the storage period. The highest shoot length was observed during the first month of storage (S_1: 10.17 cm), and the least was recorded at the end of storage (S_7: 8.67 cm).

4.2.1.3.3 Impact of interaction between Treatment (T) × Storage period (S)

Significant difference was found in the seedling shoot length due to the interaction of seed treatments and storage period.

Among the treatments, the significantly superior shoot length was observed in T_8 (*Beauveria bassiana* @1×10^8 spores.ml^{-1}: T_8S_1: 10.94 cm; T_8S_7: 9.10 cm) and T_9 (*Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: T_9S_1: 10.50 cm; T_9S_7: 9.36 cm) throughout the storage period. Shoot length of *Beauveria bassiana* @1×10^8 spores.ml^{-1} treated seeds during the first three months of storage (T_8 S_1: 10.94 cm; T_8S_2: 10.85 cm and T_8S_3: 10.77 cm) were on par with each other.

Treatments T_8 and T_9 were on par with each other except during the 4th, 5th and 6th month of storage. During the initial period *i.e.,* in S_1, S_2 and S_3 shoot length in T_8 (T_8 S_1: 10.94 cm; T_8S_2: 10.85 cm and T_8S_3: 10.77 cm) was significantly superior than T_9 (T_9S_1: 10.50 cm; T_9S_2: 10.35 cm; T_9S_3: 10.13 cm). However, at the end of storage T_9 registered high significant root length over T_8. TreatmentsT_6 (T_6S_7- Spinosad @ 10 ppm: 8.97 cm), T_7 (T_7S_7- Diatomaceous earth @ 5 g/kg: 8.93 cm) and T_2 (T_2S_7- Neem seed kernel powder @ 10 g/kg: 8.90 cm) were on par with T_8 at the end of the storage period. These were significantly superior over other treatments including untreated seeds ($T_{10}S_1$: 10.25 cm; $T_{10}S_7$: 8.33 cm) during storage.

Treatments T_6 (T_6S_7- Spinosad @ 10 ppm: 8.97 cm), T_7 (T_7S_7- Diatomaceous earth @ 5 g/kg: 8.93 cm), T_2 (T_2S_7- Neem seed kernel powder @ 10 g/kg: 8.90 cm) and T_5 (T_5S_7- Panal leaf powder @ 10 g/kg: 8.77 cm) were significantly superior to untreated seeds for most of the storage period.

Seed treatment with Neem leaf powder @ 10 g/kg (T_1S_1: 9.90 cm; T_1S_7: 8.11 cm) and Manja koova rhizome powder @ 10 g/kg (T_4S_1: 9.76 cm; T_4S_7: 7.98 cm) recorded the least length throughout the storage period. The observations in these treatments were significantly lower than in untreated seeds.

Table 6: Impact of storage period, treatments and their interaction on seedling shoot length (cm) in rice

Treatments (T)	Intervals of storage (MAS)							Mean
	S_1	S_2	S_3	S_4	S_5	S_6	S_7	
T_1	9.90^{zABCDE}	$9.64^{rstuvwxy}$	$9.47^{lmnopqrs}$	9.17^{ghijk}	8.95^{efg}	8.65^{c}	8.11^{ab}	$\mathbf{9.13^{g}}$
T_2	10.03^{CDEF}	9.96^{BCDE}	$9.80^{vwxyzABC}$	$9.68^{rstuvwxyz}$	$9.50^{mnopqrst}$	9.13^{fghij}	8.90^{cdef}	$\mathbf{9.57^{d}}$
T_3	10.08^{DEF}	9.38^{klmnop}	$9.68^{rstuvwxyz}$	$9.61^{pqrstuvw}$	9.12^{fghij}	8.89^{cdef}	8.23^{ab}	$\mathbf{9.28^{f}}$
T_4	$9.76^{uvwxyzAB}$	$9.57^{opqrstuv}$	$9.88^{xyzABCDE}$	8.79^{cde}	8.69^{cd}	8.32^{b}	7.98^{a}	$\mathbf{9.00^{h}}$
T_5	10.03^{CDEF}	$9.87^{wxyzABCD}$	$9.80^{vwxyzABC}$	$9.45^{lmnopqr}$	9.27^{hijklm}	9.02^{efgh}	8.77^{cde}	$\mathbf{9.46^{e}}$
T_6	10.23^{FG}	10.03^{CDEF}	$9.89^{yzABCDE}$	$9.68^{rstuvwxyz}$	$9.46^{lmnopqr}$	9.30^{ijklmn}	8.97^{efg}	$\mathbf{9.65^{c}}$
T_7	10.04^{CDEF}	9.91^{zABCDE}	$9.74^{stuvwxyzAB}$	$9.59^{opqrstuv}$	9.36^{jklmno}	9.24^{hijkl}	8.93^{defg}	$\mathbf{9.54^{d}}$
T_8	10.94^{I}	10.85^{I}	10.77^{I}	9.93^{ABCDE}	$9.63^{qrstuvwx}$	$9.43^{lmnopqr}$	9.10^{fghi}	$\mathbf{10.09^{a}}$
T_9	10.50^{H}	10.35^{GH}	10.13^{EFG}	$9.85^{wxyzABCD}$	$9.73^{stuvwxyzA}$	$9.67^{rstuvwxyz}$	9.36^{jklmno}	$\mathbf{9.94^{b}}$
T_{10}	10.25^{FGH}	10.01^{CDEF}	$9.54^{nopqrstu}$	$9.47^{lmnopqrs}$	9.25^{hijklm}	8.92^{defg}	8.33^{b}	$\mathbf{9.40^{e}}$
Mean	$\mathbf{10.17^{a}}$	$\mathbf{9.96^{b}}$	$\mathbf{9.87^{c}}$	$\mathbf{9.52^{d}}$	$\mathbf{9.29^{e}}$	$\mathbf{9.06^{f}}$	$\mathbf{8.67^{g}}$	

Factors	**C.D.**	**SE(d)**	**SE(m)**	**Factors**	**C.D.**	**SE(d)**	**SE(m)**	**Factors**	**C.D.**	**SE(d)**	**SE(m)**
MAS	0.035	0.019	0.013	TR	0.045	0.023	0.016	MAS × TR	0.119	0.060	0.042

4.2.1.4 Seedling dry weight (g)

The results on the impact of storage period, seed treatments and their interaction on seedling dry weight are furnished in Table 7.

Seedling dry weight during the storage period was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.4.1 Impact of seed treatment (T)

Irrespective of the storage period, seed treatments exerted significant influence on the seedling dry weight.

Seedling dry weight varied between 0.219 g in T_4 (Manja koova rhizome powder) and 0.233 g each in T_6 (Spinosad @ 10 ppm: 0.233 g) and T_8 (*Beauveria bassiana* @1×10^8 spores.ml^{-1}: 0.233 g). In addition to T_6 and T_8, significant high dry weight of seedling was observed in T_2 (Neem seed kernel extract @ 10 g/kg: 0.230 g), T_7 (Diatomaceous earth @ 5 g/kg: 0.228 g), T_9 (*Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 0.228 g) and T_1 (Neem leaf powder @ 10 g/kg: 0.227 g). Seedling dry weight in these treatments were on par with each other.

In addition, treatments T_9, T_7 and T_1 were on par with all other treatments including T_{10} (Untreated control: 0.220 g), but differed significantly from T_4.

4.2.1.4.2 Impact of storage period (S)

Irrespective of the seed treatments storage period, exerted significant difference on the seedling dry weight. It decreased as the storage period increased. Significant high dry weight of seedlings was observed in the first month of storage (S_1: 0.255g) and the least dry weight was found at the end of storage (S_7: 0.184 g). It was also observed that, the seedling dry weight during S_3 (0.232 g), S_4 (0.232 g) and S_5 (0.228 g) were on par with each other.

Table 7: Impact of storage period, treatments and their interaction on seedling dry weight (g) in rice

Treatments (T)	Intervals of storage (MAS)							Mean
	S$_1$	**S$_2$**	**S$_3$**	**S$_4$**	**S$_5$**	**S$_6$**	**S$_7$**	
T$_1$	0.251^{nopqrstu}	0.250^{nopqrstu}	0.243^{lmnopqrstu}	0.232^{ijklmnopqr}	0.228^{ghijklmno}	0.200^{cdef}	0.185^{abcd}	**0.227^{ab}**
T$_2$	0.259^{stu}	0.257^{rstu}	0.244^{lmnopqrstu}	0.232^{ijklmnopqr}	0.233^{ijklmnopqrs}	0.200^{cdef}	0.185^{abcd}	**0.230^{a}**
T$_3$	0.263^{u}	0.234^{ijklmnopqrs}	0.217^{efghijk}	0.228^{ghijklmnop}	0.220^{efghijkl}	0.209^{defghi}	0.184^{abcd}	**0.222^{bc}**
T$_4$	0.251^{nopqrstu}	0.237^{klmnopqrstu}	0.219^{efghijkl}	0.225^{fghijklmn}	0.221^{efghijklm}	0.202^{cdefg}	0.179^{abc}	**0.219^{c}**
T$_5$	0.250^{nopqrstu}	0.242^{klmnopqrstu}	0.227^{fghijklmn}	0.229^{hijklmnop}	0.222^{efghijklm}	0.204^{cdefgh}	0.172^{ab}	**0.221^{bc}**
T$_6$	0.256^{qrstu}	0.252^{opqrstu}	0.241^{lmnopqrstu}	0.235^{ijklmnopqrst}	0.231^{ijklmnopqr}	0.223^{efghijklm}	0.198^{bcde}	**0.233^{a}**
T$_7$	0.252^{opqrstu}	0.251^{nopqrstu}	0.236^{jklmnopqrstu}	0.237^{klmnopqrstu}	0.230^{hijklmnopq}	0.209^{defghi}	0.180^{abc}	**0.228^{ab}**
T$_8$	0.261^{tu}	0.252^{opqrstu}	0.234^{ijklmnopqrs}	0.244^{lmnopqrstu}	0.231^{ijklmnopqr}	0.210^{defghij}	0.198^{bcde}	**0.233^{a}**
T$_9$	0.254^{pqrstu}	0.247^{mnopqrstu}	0.237^{klmnopqrstu}	0.232^{ijklmnopqr}	0.236^{jklmnopqrstu}	0.204^{cdefgh}	0.186^{abcd}	**0.228^{ab}**
T$_{10}$	0.251^{nopqrstu}	0.244^{lmnopqrstu}	0.226^{fghijklmn}	0.229^{hijklmnop}	0.223^{efghijklm}	0.201^{cdef}	0.170^{a}	**0.220^{bc}**
Mean	**0.255^{a}**	**0.247^{b}**	**0.232^{c}**	**0.232^{c}**	**0.228^{c}**	**0.206^{d}**	**0.183^{e}**	

Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)
MAS	0.004	0.002	0.001	TR	0.005	0.002	0.002	MAS × TR	0.012	0.006	0.004

4.2.1.4.3 Impact of interaction between Treatment (T) × Storage period (S)

Significant difference in the dry weight of seedlings was observed due to the interaction of seed treatments and storage period.

The seedling dry weight varied significantly between the treatments throughout the storage. As the storage period increased, there was a significant decrease in dry weight of seedlings. During storage, although T_8 (T_8S_{1}- *Beauveria bassiana* @1×10^8 spores.ml^{-1}: 0.261 g), T_2 (T_2S_{1}- Neem seed kernel powder @10 g/kg: 0.259 g) and T_6 (Spinosad @ 10 ppm: 0.265 g) registered high dry weight, they were on par with other treatments for most of the storage period.

At the end (S_7) of the storage, T_8 (T_8S_7- *Beauveria bassiana* @1×10^8 spores.ml^{-1}: 0.198 g) and T_6 (T_6S_7- Spinosad @ 10 ppm treated seeds: 0.198 g) registered highly significant dry weight compared to T_{10} ($T_{10}S_7$- Untreated control: 0.170 g). All treated seeds were on par with each other at S_7.

However, the dry weight was comparatively low in treatment T_4 (T_4S_7- Manja koova rhizome powder @ 10 g/kg: 0.179 g), T_5 (T_5S_7- Panal leaf powder @ 10 g/kg: 0.172 g) and the least in T_{10} (Untreated control).

4.2.1.5 Seedling vigour index I (VI-I)

The results on the impact of seed treatments, storage period and their interaction on seedling vigour index are furnished in Table 8.

Seedling vigour index I was found to be significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.5.1 Impact of seed treatment (T)

Irrespective of the storage period, seed treatments exerted significant influence on the seedling vigour index I.

Among the treatments, vigour index I varied between 1533 in T_1 (Neem leaf powder @ 10 g/kg) to 1838 in T_8 (*Beauveria bassiana* @1×10^8 spores.ml^{-1}).

T_8 registered significantly higher VI-I over all other treatments. T_9 (*Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 1752), T_7 (Diatomaceous earth @ 5 g/kg: 1686), T_6 (Spinosad: 1668), T_2 (Neem seed kernel powder @ 10 g/kg: 1650), T_5 (Panal leaf powder @ 10 g/kg: 1649) were the next best treatments. These treatments except T_2 and T_5 were significantly different from each other as well as from all other treatments. T_2 and T_5 were on par with each other.

Significantly lower VI-I was observed in T_1 (Neem leaf powder @ 10 g/kg: 1533), T_4 (Manja koova rhizome powder @ 10 g/kg: 1555) and T_3 (Sweet flag rhizome powder: 1573). These differed significantly from T_{10} (Untreated control: 1627). T_4 and T_3 were on par with each but significantly different from T_1.

4.2.1.5.2 Impact of storage period (S)

Irrespective of the seed treatment, the seedling vigour index I varied significantly over the storage period. VI-I decreased progressively, differing significantly as storage period increased. The highest seedling vigour index I was observed during the first month of storage (S_1: 1952) and the least estimate was recorded at the end of storage (S_7: 1314).

4.2.1.5.3 Impact of interaction between Treatment (T) × Storage period (S)

The interaction of seed treatments and storage period (T × S) significantly influenced the seedling vigour index I.

For up to three months, the vigour index I of T_8 (*Beauveria bassiana* @1×10^8 spores.ml^{-1}) was significantly superior over other treatments (T_8S_1: 2162, T_8S_2: 2097, T_8S_3: 2042). T_9 (T_9S_1- *Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 2025), T_3 (T_3S_1- Sweet flag rhizome powder @ 10 g/kg: 1963), T_6 (T_6S_1- Spinosad @ 10 ppm: 1962), T_2 (T_2S_1- Neem seed kernel powder @ 10 g/kg: 1938), T_5 (T_5S_1- Panal leaf powder @10 g/kg: 1927) and T_7 (T_7S_1- Diatomaceous earth @ 5 g/kg: 1926) were on par with each other and differed significantly from other treatments. T_1 (T_1S_1- Neem leaf powder @ 10 g/kg: 1844) and T_4 (T_4S_1- Manja koova rhizome powder: 1857) registered significantly low VI-I, but were on par with T_{10} ($T_{10}S_1$- Untreated control: 1911).

At the end of storage, vigour index I in T_8 (T_8S_7- *Beauveria bassiana* @1×10^8 spores.ml^{-1}: 1508), T_9 (T_9S_7- *Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 1399) and T_7 (T_7S_7- Diatomaceous earth @ 5 g/kg: 1393) was high and on par with each other. However, T_9 and T_7 were also found to be on par with T_2 (T_2S_7- Neem seed kernel extract @ 10 g/kg: 1356), T_6 (T_6S_7- Spinosad @ 10 ppm: 1325) and T_5 (T_5S_7- Panal leaf powder @ 10 g/kg: 1294).

T_{10} (Untreated control: 1258), T_1 (Neem leaf powder @ 10 g/kg: 1217), T_3 (T_3S_7- Sweet flag rhizome powder @ 10 g/kg: 1213) and T_4 (T_4S_7- Manja koova rhizome powder @ 10 g/kg: 1180) recorded low vigour at S_7.

4.2.1.6 Seedling vigour index II (VI-II)

The results on the impact of storage period, seed treatments and their interaction on seedling vigour index II are presented in Table 9.

Seedling vigour index II during the storage period was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.6.1 Impact of seed treatment (T)

Irrespective of the storage period, seedling vigour index II was significantly influenced by the seed treatments.

Significantly superior seedling vigour index II was recorded in T_8 (*Beauveria bassiana* @1×10^8 spores.ml^{-1}: 2042) and the least vigour index II was recorded in T_4 (Manja koova rhizome powder @ 10 g/kg: 1826). T_9 (*Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 1934) followed by T_7 (Diatomaceous earth @ 5 g/kg: 1926), T_2 (Neem seed kernel powder @ 10 g/kg: 1921), T_6 (Spinosad @ 10 ppm: 1911) and T_5 (Panal leaf powder @ 10 g/kg: 1858), were on par with each other and found to be next best to T_8.

Table 7: Impact of storage period, treatments and their interaction on seedling vigour index I in rice

Treatments (T)	Intervals of storage (MAS)							Mean
	S_1	S_2	S_3	S_4	S_5	S_6	S_7	
T_1	1844^{BCDEFG}	1763^{yzABCD}	$1647^{qrstuvwx}$	1547^{lmnopq}	1398^{efghij}	1312^{bcdef}	1217^{abc}	**1533^{g}**
T_2	1938^{GHIJK}	1836^{BCDEFG}	$1708^{tuvwxyz}$	$1625^{pqrstuv}$	1597^{nopqrs}	1491^{ijklmn}	1356^{defgh}	**1650^{de}**
T_3	1963^{HIJK}	1739^{wxyzAB}	$1695^{stuvwxyz}$	1619^{pqrstu}	1423^{ghijk}	1356^{defgh}	1213^{ab}	**1573^{f}**
T_4	1857^{CDEFGH}	1739^{wxyzAB}	$1696^{stuvwxyz}$	1604^{opqrst}	1492^{ijklmn}	1315^{bcdefg}	1180^{a}	**1555^{fg}**
T_5	1927^{FGHIJ}	1863^{DEFGH}	1821^{ABCDEF}	1621^{pqrstu}	1549^{lmnopq}	1471^{ijklm}	1294^{bcde}	**1649^{de}**
T_6	1962^{HIJK}	1922^{FGHIJ}	1851^{CDEFG}	$1700^{stuvwxyz}$	$1495^{ijklmno}$	1419^{fghij}	1325^{cdefg}	**1668^{cd}**
T_7	1926^{FGHIJ}	1910^{EFGHI}	1868^{DEFGHI}	$1723^{uvwxyzA}$	1533^{klmnop}	1449^{hijkl}	1393^{efghij}	**1686^{c}**
T_8	2162^{M}	2097^{LM}	2042^{KL}	1803^{zABCDE}	$1660^{rstuvwxy}$	1597^{nopqrs}	1508^{jklmno}	**1838^{a}**
T_9	2025^{JKL}	1975^{IJK}	1899^{EFGHI}	$1735^{vwxyzAB}$	$1638^{pqrstuvw}$	1593^{nopqrs}	1399^{efghij}	**1752^{b}**
T_{10}	1911^{EFGHI}	1850^{CDEFG}	1750^{xyzABC}	$1696^{stuvwxyz}$	1571^{mnopqr}	1387^{efghi}	1258^{abcd}	**1632^{e}**
Mean	**1952^{g}**	**1869^{f}**	**1798^{e}**	**1656^{d}**	**1547^{c}**	**1439^{b}**	**1314^{a}**	

Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)
MAS	16.313	8.242	5.828	TR	19.497	9.851	6.966	MAS × TR	51.585	25.063	18.429

T_4 (Manja koova rhizome powder @ 10 g/kg: 1826), T_3 (Sweet flag rhizome @ 10 g/kg: 1846) and T_1 (Neem leaf powder @ 10 g/kg: 1856) were on par with T_{10} (Untreated control: 1855) and registered lower vigour index II. The estimate was the least in T_4.

4.2.1.6.2 Impact of storage period (S)

Irrespective of the seed treatment, significant difference was observed in the seedling vigour index II during the period of storage. Vigour index II decreased significantly over the storage period. The highest estimate was recorded during the first month of storage (S_1:2363) and least vigour was recorded at the end (S_7: 1330) of storage.

4.2.1.6.3 Impact of interaction between Treatment (T) × Storage period (S)

The interaction of seed treatments and storage period exerted significant influence on seedling vigour index II.

During the first month of storage (S_1), vigour index II was found to be the highest in T_8 (T_8S_1- *Beauveria bassiana* @1×10^8 spores.ml^{-1}: 2514), followed by T_3 (T_3S_1- Sweet flag rhizome powder @ 10 g/kg: 2469), T_2 (T_2S_1- Neem seed kernel powder @ 10 g/kg: 2426) and T_9 (T_9S_1- *Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 2342). T_{10} ($T_{10}S_1$- Untreated control: 2298), T_4 (T_4S_1- Manja koova rhizome powder @ 10 g/kg: 2303), T_5 (T_5S_1- Panal leaf powder @ 10 g/kg: 2309) and T_1 (T_1S_1- Neem leaf powder @ 10 g/kg: 2320) registered lower estimates.

As the storage proceeded, the vigour index II declined over the storage period. At the end of storage (S_7), significantly superior vigour index II was recorded in T_8 (T_8S_7- *Beauveria bassiana* @1×10^8 spores.ml^{-1}: 1560). T_6 (T_6S_7- Spinosad @ 10 ppm: 1402), followed by T_9 (T_9S_7- *Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 1348), T_2 (T_2S_7- Neem seed kernel powder @ 10 g/kg: 1344) and T_7 (T_7S_7- Diatomaceous earth @ 5 g/kg: 1338) were next best to T_8.

Table 9: Impact of storage period, seed treatments and their interaction on seedling vigour index II

Treatments (T)	Intervals of storage (MAS)							Mean
	S_1	S_2	S_3	S_4	S_5	S_6	S_7	
T_1	2320^{xyzAB}	$2211^{tuvwxyzA}$	$2046^{opqrtuvw}$	$1894^{lmnopqr}$	$1740^{ghijklmn}$	$1476^{abcdefg}$	1305^{abcd}	**1856^{cde}**
T_2	2426^{zAB}	2296^{wxyzAB}	$2048^{opqrstuvw}$	$1884^{klmnopqr}$	$1879^{klmnopq}$	1570^{defghi}	1344^{abcde}	**1921^{bcd}**
T_3	2469^{AB}	$2122^{qrstuvwxy}$	$1896^{lmnopqr}$	$1892^{klmnopqr}$	$1691^{ghijklm}$	1563^{defgh}	1289^{abcd}	**1846^{de}**
T_4	2303^{wxyzAB}	$2120^{qrstuvwxy}$	$1899^{lmnopqr}$	$1844^{jklmnopq}$	$1833^{ijklmnop}$	1522^{cdefg}	1261^{abc}	**1826^{e}**
T_5	2309^{wxyzAB}	$2177^{stuvwxyz}$	$2010^{opqrstuv}$	$1888^{klmnopqr}$	$1794^{hijklmno}$	1596^{efghi}	1235^{ab}	**1858^{bcde}**
T_6	2330^{xyzAB}	$2257^{vwxyzAB}$	$2102^{pqrstuvwxy}$	$1933^{mnopqrs}$	$1727^{ghijklmn}$	1624^{fghijk}	1402^{abcdef}	**1911^{bcd}**
T_7	2315^{xyzAB}	2292^{wxyzAB}	$2135^{qrstuvwxy}$	$2023^{opqrstuv}$	$1809^{hijklmno}$	1570^{defghi}	1338^{abcde}	**1926^{bc}**
T_8	2514^{B}	2360^{yzAB}	$2152^{rstuvwxy}$	$2114^{qrstuvwxy}$	1902^{mnopqr}	$1694^{ghijklm}$	1560^{defgh}	**2042^{a}**
T_9	2342^{xyzAB}	$2240^{uvwxyzA}$	$2090^{pqrstuvwx}$	$1952^{mnopqrst}$	$1931^{mnopqrs}$	$1632^{fghijkl}$	1348^{abcde}	**1934^{b}**
T_{10}	2298^{wxyzAB}	$2177^{stuvwxyz}$	$1971^{nopqrstu}$	$1988^{nopqrstu}$	$1833^{ijklmnop}$	1501^{bcdefg}	1215^{a}	**1855^{cde}**
Mean	**2363^{a}**	**2225^{b}**	**2035^{c}**	**1941^{d}**	**1814^{e}**	**1575^{f}**	**1330^{g}**	

Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)
MAS	39.615	20.016	14.153	TR	47.349	23.923	16.916	MAS × TR	125.275	63.295	44.756

These treatments were on par with each other. VI-II was the least in T_{10} ($T_{10}S_7$- Untreated control: 1215). Similar low vigour estimates were observed in T_5 (T_5S_7- Panal leaf powder @ 10 g/kg: 1235), T_4 (T_4S_7- Manja koova rhizome powder @ 10 g/kg: 1261), T_3 (T_3S_7- Sweet flag rhizome powder @10 g/kg: 1289) and T_1 (T_1S_7- Neem leaf powder @10 g/kg: 1305).

4.2.1.7 Seed moisture content (%)

Results on the impact of seed treatment, storage period and their interaction on the moisture content of seeds are furnished in Table 10.

Seed moisture content was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.7.1 Impact of seed treatment (T)

Irrespective of the storage period, seed moisture content was significantly influenced by various seed treatments.

Seed moisture content ranged between 13.54 per cent ($T_{7:}$ Diatomaceous earth @ 5 g/kg) and 14.28 per cent ($T_{9:}$ *Bacillus thuringiensis* @1×10^8 spores.ml^{-1}).

T_7 was on par with T_6 (Spinosad @ 10 ppm: 13.58 %), T_5 (Panal leaf powder @ 10 g/kg: 13.68 %), T_2 (Neem seed kernel powder @ 10 g/kg: 13.70 %) and T_3 (Sweet flag rhizome powder @ 10 g/kg: 13.80 %) and differed significantly from T_1 (Neem leaf powder @ 10 g/kg: 13.90 %), T_4 (Manja koova rhizome powder @ 10 g/kg: 14.19 %), T_8 (*Beauveria bassiana* @1×10^8 spores.ml^{-1}: 14.08 %), T_{10} (Untreated control: 14.24 %) and T_9. Untreated control was on par with treatments $T_{9,}$ T_8 and $T_{4.}$

4.2.1.7.2 Impact of storage period (S)

Irrespective of the seed treatment, the storage period significantly influenced he seed moisture content.

Significant increase in moisture content of seeds was observed over the storage period. The least seed moisture estimate was observed during first month (S_1: 12.47 %) and the highest moisture content was observed at the end of storage (S_7: 14.83 %).

4.2.1.7.3 Impact of interaction between Treatment (T) × Storage period (S)

Significant difference in the seed moisture content was evident due to the interaction of seed protectants and storage period.

At first month, seed moisture content varied between 12.13 per cent in T_6 (T_6S_1- Spinosad @ 10 ppm) and 13.33 per cent in T_4 (T_4S_1- Manja koova rhizome powder @10 g/kg). T_6 was on par with all the treatments including untreated control ($T_{10}S_1$- Untreated control: 12.53 %) but differed significantly from T_4 (T_4S_1- Manja koova rhizome powder @10 g/kg: 13.33 %).

At the end of the storage period, relatively low moisture content was recorded in seeds treated with T_7 (Diatomaceous earth @ 5 g/kg: 14.43 %), T_3 (T_3S_7- Sweet flag rhizome powder @ 10 g/kg: 14.67 %), T_5 (T_5S_7- Panal leaf powder @ 10 g/kg: 14.67 %) and T_2 (T_2S_7- Neem seed kernel extract @ 10 g/kg: 14.73 %), while it was high in seed treatments T_9 (T_9S_7- *Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 15.17 %), T_{10} ($T_{10}S_7$- Untreated control: 15.00 %), T_8 (T_8S_7- *Beauveria bassiana* @1×10^8 spores.ml^{-1}: 15.00 %) and T_1 (T_1S_7- neem leaf powder @ 10 g/kg: 15.00 %). However, all treatments were on par with each other.

Table 10: Impact of storage period, treatments and their interaction on seed moisture content (%) in rice

Treatments (T)	Intervals of storage (MAS)							Mean
	S_1	S_2	S_3	S_4	S_5	S_6	S_7	
T_1	12.33^{abc}	$13.30^{bcdefghijk}$	$13.33^{cdefghijkl}$	$14.00^{hijklmnopqrstuvw}$	$14.63^{pqrstuvwx}$	$14.47^{nopqrstuvwx}$	15.00^{wx}	**13.90^{bc}**
T_2	12.43^{abcd}	$12.93^{abcdefg}$	$13.53^{efghijklmno}$	$13.87^{ghijklmnopqrst}$	$14.06^{ijklmnopqrstuvw}$	$14.40^{mnopqrstuvwx}$	$14.73^{rstuvwx}$	**13.70^{ab}**
T_3	12.53^{abcde}	$13.13^{abcdefghij}$	$13.83^{ghijklmnopqrs}$	$14.13^{jklmnopqrstuvwx}$	$13.67^{ghijklmnopq}$	$14.30^{klmnopqrstuvwx}$	$14.67^{qrstuvwx}$	**13.80^{ab}**
T_4	$13.33^{cdefghijkl}$	$13.77^{ghijklmnopqr}$	$13.93^{ghijklmnopqrstuv}$	$14.10^{ijklmnopqrstuvw}$	$14.47^{nopqrstuvwx}$	14.83^{stuvwx}	14.90^{tuvwx}	**14.19^{d}**
T_5	12.33^{abc}	$13.26^{bcdefghijk}$	$13.53^{efghijklmno}$	$13.60^{fghijklmnop}$	$14.03^{hijklmnopqrstuvw}$	$14.40^{mnopqrstuvwx}$	$14.67^{qrstuvwx}$	**13.68^{ab}**
T_6	12.13^{a}	$13.00^{abcdefgh}$	$13.40^{defghijklmn}$	$13.47^{efghijklmno}$	$13.90^{ghijklmnopqrstu}$	$14.40^{mnopqrstuvwx}$	$14.77^{rstuvwx}$	**13.58^{a}**
T_7	12.26^{ab}	$12.93^{abcdefg}$	$13.30^{bcdefghijk}$	$13.30^{bcdefghijk}$	$14.13^{jklmnopqrstuvwx}$	$14.20^{klmnopqrstuvwx}$	$14.43^{mnopqrstuvwx}$	**13.51^{a}**
T_8	12.57^{abcdef}	$14.00^{hijklmnopqrstuvw}$	$14.37^{lmnopqrstuvwx}$	$13.53^{efghijklmno}$	$14.53^{opqrstuvwx}$	$14.57^{opqrstuvwx}$	15.00^{wx}	**14.08^{cd}**
T_9	12.26^{ab}	$13.73^{ghijklmnopqr}$	$14.37^{lmnopqrstuvwx}$	$14.63^{pqrstuvwx}$	$14.77^{rstuvwx}$	15.00^{wx}	15.17^{x}	**14.28^{d}**
T_{10}	12.53^{abcde}	$13.06^{abcdefghi}$	$14.53^{opqrstuvwx}$	$14.60^{pqrstuvwx}$	14.93^{uvwx}	15.00^{wx}	15.00^{wx}	**14.24^{d}**
Mean	**12.47^{a}**	**13.31^{b}**	**13.81^{c}**	**13.92^{c}**	**14.31^{d}**	**14.56^{e}**	**14.83^{f}**	

Factors	**C.D.**	**SE(d)**	**SE(m)**	**Factors**	**C.D.**	**SE(d)**	**SE(m)**	**Factors**	**C.D.**	**SE(d)**	**SE(m)**
MAS	0.154	0.078	0.055	TR	0.184	0.093	0.066	MAS × TR	0.486	0.246	0.174

4.2.1.8 Seed microflora infection (%)

The result on per cent seed microflora in various treatments, at the end of the storage period is furnished in Table 11.

Seed treatments exerted a significant impact on the seed microflora infection (%). Seed microflora infection was examined by blotter method and agar plate method (Plate 2).

In blotter method, the seed microflora infection (%) varied from 18.92 per cent in T_2 (Neem seed kernel extract powder @ 10 g/kg) to 50.55 per cent in T_{10} (Untreated control). Presence of seed microflora (%) was also found to be low in T_6 (Spinosad @ 10 ppm: 24.21 %), T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}: 25.60 %), T_3 (Sweet flag rhizome powder @ 10 g/kg: 26.12 %) T_1 (Neem leaf powder @ 10 g/kg: 26.21 %), and T_9 (*Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}: 27.25 %). These treatments were found to be on par with each other but differed significantly from T_2 as well as others. Seed microflora in treatment T_7 (Diatomaceous earth @ 5 g/kg: 28.84 %), T_5 (Panal leaf powder @ 10 g/kg: 27.88 %) and T_4 (Manja koova rhizome powder @ 10 g/kg recorded: 32.14 %) were also high. These treatments differed significantly from T_{10}.

In agar plate method, the seed microflora infection (%) varied from 24.90 per cent in T_2 (Neem seed kernel extract powder @ 10 g/kg) to 60.84 per cent in the T_{10} (Untreated control) at the end of storage. T_6 (Spinosad @ 10 ppm: 28.41 %), T_8 (*Beauveria bassiana* @ 1×10^8 spores.ml^{-1}: 29.38 %) and T_7 (Diatomaceous earth @ 5 g/kg: 32.26 %) were on par with each other but differed significantly from T_2 that registered significant low estimates over all the treatments.

The per cent seed microflora was high in treatments T_1 (Neem leaf powder @ 10 g/kg: 36.60 %) and T_3 (Sweet flag rhizome powder @ 10 g/kg: 26.12 %). These were on par with each other, but differed significantly from T_{10} that registered the highest estimate for this parameter.

Presence of fungi *Aspergillus niger* and *Aspergillus flavus* was evident in the seeds evaluated (Plate 2).

Table 11. Effect of treatments on per cent seed microflora in rice, at the end of storage

Treatments	Seed microflora (%)	
	Blotter method	Agar plate method
T_1	26.21[bc] (5.12)	36.60[de] (6.05)
T_2	18.92[a] (4.35)	24.90[a] (4.99)
T_3	26.12[bc] (5.15)	36.96[de] (6.08)
T_4	32.14[d] (5.67)	40.58[e] (6.37)
T_5	27.88[c] (5.28)	35.64[cd] (5.97)
T_6	24.21[b] (4.92)	28.41[b] (5.33)
T_7	28.84[c] (5.37)	32.26[bc] (5.68)
T_8	25.60[bc] (5.06)	29.38[b] (5.42)
T_9	27.25[bc] (5.22)	35.04[cd] (5.92)
T_{10}	50.55[e] (7.11)	60.84[f] (7.80)
CD	**0.273**	**0.310**
SE(m)	**0.092**	**0.104**

(Values in parenthesis are square root transformed values)

4.2.1.9 Number of beetles per 100 g of seed

Results on the impact of seed treatments, storage period and their interactions on number of beetles in seed are furnished in Table.12.

Number of beetles were significantly influenced by seed treatments, storage period and their interaction.

4.2.1.9.1 Impact of seed treatment (T)

Irrespective of the storage period, the seed treatments exerted significant influence on number of beetles in the seed sample.

The estimates ranged between T_{10} (Untreated control: 15.57) and T_7 (Diatomaceous earth @ 10 g/kg: 6.05). T_7 was on par with T_2 (Neem seed kernel powder @ 10 g/kg: 7.57), T_6 (Spinosad @ 10 ppm: 8.05), T_1 (Neem leaf powder @ 10 g/kg: 8.14) and T_9 (*Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}: 8.38), recorded lesser number of beetles, but are on par with other treatments. T_8 (*Beauveria bassiana* @1×10^8 spores.ml^{-1}: 8.81), T_5 (Panal leaf powder @ 10 g/kg: 8.95), T_3 (Sweet flag rhizome powder @ 10 g/kg: 9.57) and T_4 (Manja koova rhizome powder @ 10 g/kg: 9.71) were the next best and were also on par with each other. The treated seeds registered significant low estimates compared to untreated seeds.

Storage pests like red flour beetle (*Tribolium castaneum)* and lesser grain borer (*Rhyzopertha dominica*) and rice weevil (*Sitophilus oryzae*) were (Plate 3) observed to infest the seed lot in the various treatments.

4.2.1.9.2 Impact of storage period (S)

Irrespective of the seed treatments, the storage period exerted significant influence on the number of beetles in the seed sample.

The number of beetles increased significantly with an increase in storage period. The least estimate was recorded during the first month (S_1: 1.37) and the maximum was recorded at the end of storage (S_7: 16.47). The beetle infestation during the second (S_2: 4.83) and third month (S_3: 6.32) of storage were on par with each other.

4.2.1.9.3 Impact of interaction between Treatment (T) × Storage period (S)

The interaction of seed treatment and storage period exerted a significant influence on the number of beetles in the seed sample.

The estimates varied between 0.67 each in T_7 (Diatomaceous earth @5 g/kg), T_1 (Neem leaf powder @10 g/kg) and T_2 (Neem seed kernel powder @ 10 g/kg) to 2.33 in T_{10} ($T_{10}S_1$- Untreated control). The number of beetles in both the treated and untreated seeds were on par with each other during the first three months after storage (S_1, S_2 and S_3). Thereafter, significant differences were observed between treated and untreated seeds.

At the end of storage (S_7), although the occurrence of beetles was found to be low in T_7 (T_7S_7- diatomaceous earth @ 5 g/kg treatment: 11.33), T_6 (T_6S_7- Spinosad @ 10 ppm: 13.66) and T_2 (T_2S_7- Neem seed kernel extract @ 10 g/kg:14.00), the estimate in all the treatments was found to be significantly low compared to T_{10} ($T_{10}S_7$- Untreated control: 31.00).

4.2.1.10 Number of larvae per 100 g of seed

The presence of larvae was not noticed in the various seed samples drawn, throughout the storage in both untreated and treated seeds.

4.2.1.11 Number of infested seeds per 100 g of seed

Results on the impact of seed treatments, storage period and their interaction on the number of infested seeds were represented in Table 13.

Number of infested seeds per 100 g of seed varied significantly with treatments, storage period and their interaction.

Table 12: Impact of storage period, treatments and their interaction on number of beetles per 100 g of seed in rice

Treatments (T)	Intervals of storage (MAS)							Mean
	S_1	S_2	S_3	S_4	S_5	S_6	S_7	
T_1	0.67^{a}	4.33^{abcdef}	$5.67^{abcdefghi}$	$7.33^{abcdefghijkl}$	$11.33^{efghijklmn}$	$12.33^{fghijklmn}$	15.33^{klmno}	**8.14^{ab}**
T_2	0.67^{a}	4.33^{abcdef}	4.66^{abcdef}	$7.33^{abcdefghijkl}$	$8.66^{abcdefghijklm}$	$13.33^{ghijklmn}$	$14.00^{ijklmno}$	**7.57^{ab}**
T_3	1.66^{abc}	4.66^{abcdef}	$7.00^{abcdefghijk}$	$8.66^{abcdefghijklm}$	$11.66^{efghijklmn}$	15.66^{lmno}	17.66^{no}	**9.57^{b}**
T_4	1.33^{ab}	$5.33^{abcdefgh}$	$6.33^{abcdefghij}$	$9.66^{bcdefghijklmn}$	$14.00^{ijklmno}$	14.33^{jklmno}	17.00^{mno}	**9.71^{b}**
T_5	1.66^{abc}	$5.67^{abcdefghi}$	$7.00^{abcdefghijk}$	$7.66^{abcdefghijkl}$	$12.66^{fghijklmn}$	$13.33^{ghijklmn}$	14.66^{jklmno}	**8.95^{b}**
T_6	1.33^{ab}	3.67^{abcde}	4.33^{abcdef}	$7.33^{abcdefghijkl}$	$12.33^{fghijklmn}$	$13.66^{hijklmn}$	$13.66^{hijklmn}$	**8.05^{ab}**
T_7	0.67^{a}	2.33^{abcd}	3.67^{abcde}	$6.66^{abcdefghij}$	$7.00^{abcdefghijk}$	$10.66^{defghijklmn}$	$11.33^{efghijklmn}$	**6.05^{a}**
T_8	1.66^{abc}	$5.00^{abcdefg}$	$6.33^{abcdefghij}$	$9.66^{bcdefghijklmn}$	$10.00^{cdefghijklmn}$	14.33^{jklmno}	14.66^{jklmno}	**8.81^{b}**
T_9	1.66^{abc}	4.66^{abcdef}	$6.33^{abcdefghij}$	$7.33^{abcdefghijkl}$	$9.66^{bcdefghijklmn}$	$13.66^{hijklmn}$	15.33^{klmno}	**8.38^{ab}**
T_{10}	2.33^{abcd}	$8.33^{abcdefghijklm}$	$12.00^{efghijklmn}$	15.33^{klmno}	17.66^{no}	22.33^{o}	31.00^{p}	**15.57^{c}**
Mean	**1.37^{a}**	**4.83^{b}**	**6.32^{b}**	**8.70^{c}**	**11.50^{d}**	**14.37^{e}**	**16.47^{f}**	

Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)
MAS	1.268	0.641	0.453	TR	1.516	0.766	0.541	MAS × TR	4.010	2.026	1.433

4.2.1.11.1 Impact of seed treatment (T)

Irrespective of the storage period, the seed treatments exerted significant influence on the number of infested seeds in the seed sample.

The estimated ranged between 113 (T_7: Diatomaceous earth @ 10 g/kg) and 196 in T_{10} (Untreated control). T_7 was found to be on par with T_2 (Neem seed kernel powder @ 10 g/kg: 124), but varied significantly from other treatment.

Significant low estimates for this trait were also recorded in T_3 (Sweet flag rhizome powder @ 10 g/kg: 133), T_6 (Spinosad @ 10 ppm: 133) and T_1 (Neem leaf powder @ 10 g/kg: 139) and were on par with each other.

Significantly higher number of infested seeds were recorded in T_{10} (Untreated control: 196). T_9 (*Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}: 153), T_4 (Manja koova rhizome powder @ 10 g/kg: 153), T_8 (*Beauveria bassiana* @1×10^8 spores.ml^{-1}: 145) and T_5 (Panal leaf powder @ 10 g/kg: 151), which were on par with each other, but differed significantly from T_{10}.

4.2.1.11.2 Impact of storage period (S)

Irrespective of the seed treatment, storage period exerted significant influence on the number of beetles in the seed sample.

Although the estimates varied between treatments, the significant increase in the number of infested seeds over the storage period (S_2: 96.6, S_3: 120.8, S_4: 137.0, S_5: 165.3, S_6: 200.8) was observed. The least number of infested seeds was during S_1: 67.1 and the highest was found at the end of the storage (S_7: 220.3).

4.2.1.11.3 Impact of interaction between Treatment (T) × Storage period (S)

The interaction of seed treatments and storage period exerted significant influence on the number of infested seeds in the seed sample.

Table 13: Impact of storage period, treatments and their interaction on number of infested seeds per 100 g of seed in rice

Treatments (T)	Intervals of storage (MAS)							Mean
	S_1	S_2	S_3	S_4	S_5	S_6	S_7	
T_1	52.7^{ab}	101.3^{defghijkl}	122.7^{ghijklmnopqr}	133.0^{jklmnopqrs}	159.0^{qrstuvwxyz}	195.3^{yzABCDE}	211.3^{BCDEF}	**139.00^{cde}**
T_2	60.0^{abcd}	77.3^{abcdef}	110.0^{fghijklmno}	117.0^{fghijklmnopq}	148.0^{nopqrstuvw}	164.0^{rstuvwxyz}	192.7^{xyzABCDE}	**124.00^{ab}**
T_3	77.7^{abcdef}	85.7^{abcdefgh}	100.0^{defghijkl}	112.3^{fghijklmnop}	134.7^{klmnopqrst}	207.0^{ABCDEF}	215.7^{BCDEF}	**133.00^{bc}**
T_4	63.0^{abcde}	106.7^{fghijklmn}	128.7^{ijklmnopqrs}	145.3^{mnopqrstuv}	179.7^{uvwxyzABC}	221.7^{CDEF}	226.7^{DEF}	**153.00^{f}**
T_5	75.3^{abcdef}	125.3^{ghijklmnopqrs}	142.0^{lmnopqrstu}	153.0^{pqrstuvwxy}	161.0^{rstuvwxyz}	176.0^{tuvwxyzAB}	223.3^{DEF}	**151.00^{ef}**
T_6	76.7^{abcdef}	97.7^{cdefghijk}	115.3^{fghijklmop}	126.0^{hijklmnopqrs}	135.7^{klmnopqrst}	185.7^{vwxyzABCD}	191.0^{xyzABCDE}	**133.00^{bc}**
T_7	44.3^{a}	57.0^{abc}	89.0^{bcdefghi}	103.7^{efghijklm}	140.3^{klmnopqrstu}	168.0^{stuvwxyzA}	186.7^{vwxyzABCD}	**113.00^{a}**
T_8	54.7^{ab}	83.0^{abcdefg}	118^{fghijklmnopq}	150.3^{opqrstuvwx}	197.7^{yzABCDEF}	201.0^{zABCDEF}	213.0^{BCDEF}	**145.00^{def}**
T_9	76.3^{abcdef}	104.3^{efghijklm}	129.0^{ijklmnopqrs}	140.3^{klmnopqrstu}	164.3^{rstuvwxyzA}	217.3^{BCDEF}	239.3^{FG}	**153.00^{f}**
T_{10}	90.3^{bcdefghij}	128.0^{hijklmnopqrs}	154.0^{pqrstuvwxy}	189.3^{wxyzABCD}	232.3^{EFG}	272.0^{GH}	303.7^{H}	**196.00^{g}**
Mean	**67.10^{a}**	**96.60^{b}**	**120.80^{c}**	**137.00^{d}**	**165.30^{e}**	**200.80^{f}**	**220.30^{g}**	

Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)	Factors	C.D.	SE(d)	SE(m)
MAS	6.293	3.180	2.248	TR	7.522	3.801	2.687	MAS × TR	19.902	10.055	7.110

During (S_1), the number of insect infested seeds varied between 44.30 (T_7S_1- Diatomaceous earth @ 5 g/kg) and 90.30 ($T_{10}S_1$- Untreated control). In addition to T_7, low estimates for this parameter were observed in T_1 (T_1S_1- Neem leaf powder @ 10 g/kg: 52.70) and T_8 (T_8S_1- *Beauveria bassiana* @ 1×10^8 spores.ml^{-1}: 52.70), while it was found to be comparatively high in T_3 (T_3S_1- Sweet flag rhizome powder @ 10 g/kg: 77.70), T_6 (T_6S_1- Spinosad @ 10 ppm: 76.70) and T_9 (T_9S_1- *Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}: 76.30). In spite of this variability in the number of insect infested seeds, the treated seeds differed significantly from the untreated seeds.

At the end of the storage (S_7), T_7 (T_7S_7- Diatomaceous earth @ 5 g/kg: 186.70), T_6 (T_6S_7- Spinosad @ 10 ppm: 191.00), T_2 (T_2S_7- Neem seed kernel powder @ 10 g/kg: 192.70) registered comparatively lower estimates of the number of insect infested seeds in the seed sample. However, the difference between these treatments and other treated seeds was only marginal. The estimate in the treated seeds differed significantly from the untreated control T_{10} ($T_{10}S_7$- Untreated control: 303.70).

4.2.1.12 Weight of damaged seeds (g) per 100 g of seed

Results on the impact of seed treatments, storage period and their interaction on the weight of undamaged seeds are presented in Table 14.

Weight of damaged seeds per 100 g of seed was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.12.1 Impact of seed treatment (T)

Irrespective of the storage period, the seed treatments exerted significant influence on the weight of undamaged seeds in the seed sample.

Significant high weight of damaged seeds was recorded in the T_{10} (Untreated control: 8.34 g). Among the treatments, the weight of damaged seeds varied between 1.89 g in T_7 (Diatomaceous earth @5 g/kg) to 2.92g in T_4 (Manja koova rhizome powder @ 10 g/kg).

Table 14: Impact of storage period, treatments and their interaction on weight of damaged seeds (g) per 100 g of seed in rice

Treatments (T)	Intervals of storage (MAS)							Mean
	S_1	S_2	S_3	S_4	S_5	S_6	S_7	
T_1	1.30^{abcde}	$1.83^{abcdefghij}$	$2.35^{abcdefghijklmnop}$	$2.81^{defghijklmnopq}$	$2.87^{defghijklmnopq}$	$3.32^{hijklmnopqr}$	$3.67^{lmnopqr}$	$\mathbf{2.60^{cd}}$
T_2	1.00^{ab}	$1.54^{abcdefg}$	$1.81^{abcdefghij}$	$1.96^{abcdefghijkl}$	$2.39^{abcdefghijklmnop}$	$2.76^{cdefghijklmnopq}$	$3.04^{efghijklmnopq}$	$\mathbf{2.07^{ab}}$
T_3	1.12^{abcd}	$2.10^{abcdefghijklm}$	$2.42^{abcdefghijklmnopq}$	$2.36^{abcdefghijklmnop}$	$3.05^{efghijklmnopq}$	$3.42^{ijklmnopqr}$	4.10^{qrs}	$\mathbf{2.66^{cd}}$
T_4	$1.90^{abcdefghijk}$	$2.60^{bcdefghijklmnopq}$	$2.70^{cdefghijklmnopq}$	$2.84^{defghijklmnopq}$	$2.74^{cdefghijklmnopq}$	$3.78^{mnopqrs}$	3.89^{opqrs}	$\mathbf{2.92^{d}}$
T_5	1.12^{abcd}	$2.13^{abcdefghijklmn}$	$2.33^{abcdefghijklmnop}$	$3.20^{ghijklmnopqr}$	$3.17^{fghijklmnopq}$	$3.10^{fghijklmnopq}$	$3.65^{klmnopqr}$	$\mathbf{2.67^{cd}}$
T_6	1.06^{abc}	$1.81^{abcdefghij}$	$2.35^{abcdefghijklmnop}$	$2.80^{cdefghijklmnopq}$	$2.80^{cdefghijklmnopq}$	$2.94^{defghijklmnopq}$	$3.14^{fghijklmnopq}$	$\mathbf{2.42^{bc}}$
T_7	0.98^{a}	1.43^{abcdef}	$1.66^{abcdefgh}$	$1.74^{abcdefghi}$	$2.10^{abcdefghijklm}$	$2.61^{bcdefghijklmnopq}$	$2.68^{bcdefghijklmnopq}$	$\mathbf{1.89^{a}}$
T_8	1.34^{abcde}	$2.20^{abcdefghijklmno}$	$2.29^{abcdefghijklmno}$	$3.20^{ghijklmnopqr}$	$3.20^{ghijklmnopqr}$	3.86^{nopqrs}	3.96^{pqrs}	$\mathbf{2.87^{cd}}$
T_9	1.12^{abcd}	$2.43^{abcdefghijklmnopq}$	$2.43^{abcdefghijklmnopq}$	$2.68^{bcdefghijklmnopq}$	$3.30^{hijklmnopqr}$	$3.51^{jklmnopqr}$	3.91^{opqrs}	$\mathbf{2.77^{cd}}$
T_{10}	4.95^{rst}	5.49^{st}	6.41^{tu}	7.71^{uv}	8.90^{v}	12.00^{w}	12.92^{w}	$\mathbf{8.34^{e}}$
Mean	$\mathbf{1.59^{a}}$	$\mathbf{2.35^{b}}$	$\mathbf{2.67^{b}}$	$\mathbf{3.13^{c}}$	$\mathbf{3.45^{c}}$	$\mathbf{4.13^{d}}$	$\mathbf{4.50^{d}}$	

Factors	**C.D.**	**SE(d)**	**SE(m)**	**Factors**	**C.D.**	**SE(d)**	**SE(m)**	**Factors**	**C.D.**	**SE(d)**	**SE(m)**
MAS	0.259	0.131	0.092	TR	0.309	0.156	0.111	MAS × TR	0.819	0.414	0.292

Treatments T_7 and T_2 (Neem seed kernel extract @10 g/kg: 2.07 g) were on par with each other and in addition, T_7 registered significantly lower weight of damaged seeds than other treatments. However, T_2 was found to be on par with T_6 (Spinosad @10 ppm: 2.42 g). Significantly higher values were observed in untreated seeds as well as seed protectant treatments T_4, T_8 (*Beauveria bassiana* @1×10^8 spores.ml^{-1}: 2.87 g) and T_9 (*Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 2.77 g). Untreated seeds differed significantly from T_4, T_8 and T_9.

4.2.1.12.2 Impact of storage period (S)

Irrespective of the seed treatments, the storage period exerted significant influence on the weight of undamaged seeds in the seed sample.

The weight of damaged seeds varied between 1.59 g in S_1 to 4.50g in S_7. It was found to gradually increase with increase in storage period in both treated and untreated seed lots. The estimates during the second and third month (S_2: 2.35 g; S_3: 2.67 g), fourth and fifth month (S_4: 3.13 g; S_5: 3.45 g) as well as the sixth and seventh month (S_6: 4.13 g; S_7: 4.50 g) were found to be on par with each other.

4.2.1.12.3 Impact of interaction between Treatment (T) × Storage period (S)

The interaction of seed treatment and storage period exerted significant influence on the weight of undamaged seeds in the seed sample. An increase in weight of damaged seeds was observed over the storage period.

Weight of damaged seeds at S_1, ranged between 0.98 g in T_7 (T_7S_1- Diatomaceous earth @ 5 g/kg) and 4.95 g in T_{10} (Untreated control). T_2 (T_2S_7- Neem seed kernel extract @ 10 g/kg: 1.00g) and T_6 (T_6S_1- Spinosad @ 10 ppm: 1.06g), recorded lower estimates next to T_7. However, all the seed treatments registered significantly lower estimates of weight of damaged seeds compared to the untreated control. The seed protectant treatments were found to be on par with each other.

Highly significant estimates of weight of damaged seeds was recorded in T_{10} throughout the storage period. As observed in S_1, at the end of storage (S_7), the treatments T_7 (T_7S_7: Diatomaceous earth @ 10 g/kg: 2.68 g), T_2 (T_2S_7- Neem seed kernel powder @ 10 g/kg: 3.04 g) and T_6 (T_6S_7- Spinosad @ 10 ppm: 3.14 g) registered comparatively lower weight of damaged seeds, while, higher estimates were observed in T_4 (T_4S_7- Manja koova rhizome powder @ 10 g/kg: 4.10 g), T_8 (T_8S_7- *Beauveria bassiana* @1×10^8 spores.ml^{-1}: 3.96 g) and T_9 (T_9S_7- *Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}: 3.91 g). All the treatments however, were on par with each other and significantly superior to T_{10} ($T_{10}S_7$- Untreated control: 12.92 g).

4.2.1.13 Weight of undamaged seeds (g) per 100 g of seed

Results on the influence of seed treatments, storage period and their interaction on the weight of undamaged seeds are presented in Table 15.

Weight of undamaged seeds per 100 g of seed was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.13.1 Impact of seed treatment (T)

Irrespective of the storage period, the seed treatments exerted significant influence on the weight of damaged seeds in the seed sample.

Among the seed protectant treatments, weight of undamaged seeds varied from 98.11 g in T_7 (Diatomaceous earth @ 5 g/kg) to 97.08 g in T_4 (Manja koova seed treatment @ 10 g/kg). Maximum weight of undamaged seeds was recorded in T_{10} (Untreated control: 91.66 g).

Treatments T_7 and T_2 (Neem seed kernel extract @ 5 g/kg: 97.93 g) were on par with each other and in addition, T_7 registered significantly higher weight of undamaged seeds than other treatments. However, T_2 was found to be on par with T_6 (Spinosad @ 10 ppm: 97.58 g). Significantly lower values were observed in untreated seeds as well as seed protectant treatments T_4, T_8 (*Beauveria bassiana* @1×10^8 spores.ml^{-1}: 97.13 g) and T_9 (*Bacillus thuringiensis* @1×10^8 spores.ml^{-1}: 97.23 g). Untreated seeds differed significantly from T_4, T_8 and T_9.

4.2.1.13.2 Impact of storage period (S)

Irrespective of the seed treatments, the weight of undamaged seeds in the seed sample varied significantly and decreased progressively during the storage period.

The weight of undamaged seeds varied between 98.41g in S_1 to 95.50 g in S_7. It gradually decreased with decrease in storage period in both treated and untreated seed lots. The estimates during the second and third month (S_2: 97.65 g; S_3: 97.33 g), fourth and fifth month (S_4: 96.87 g; S_5: 96.55 g) as well as the sixth and seventh month (S_6: 95.87 g; S_7: 95.50 g) were on par with each other.

4.2.1.13.3 Impact of interaction between Treatment (T) × Storage period (S)

The interaction of seed treatment and storage period exerted significant impact on the weight of undamaged seeds in the seed sample.

Weight of undamaged seeds at S_1, ranged between 95.05 g in T_{10} (Untreated control). and 99.02 g T_7 (T_7S_1- Diatomaceous earth @ 5 g/kg). T_2 (T_2S_7- Neem seed kernel extract @ 10 g/kg: 99.00 g) and T_6 (T_6S_1- Spinosad @ 10 ppm: 98.94 g), recorded higher estimates next to T_7. However, all the seed treatments registered significantly higher estimates of weight of damaged seeds compared to the untreated control. The seed protectant treatments were found to be on par with each other.

Significantly lower estimates of the weight of undamaged seeds was recorded in T_{10} throughout the storage period. As observed in S_1, at the end of storage (S_7), the treatments T_7 (T_7S_7: Diatomaceous earth @ 10 g/kg: 97.30 g), T_2 (T_2S_7- Neem seed kernel powder @ 10 g/kg: 96.96 g) and T_6 (T_6S_7- Spinosad @ 10 ppm: 96.86 g) had registered comparatively higher weight of undamaged seeds, while lower estimates were observed in T_4 (T_4S_7- Manja koova rhizome powder @ 10 g/kg: 96.11 g), T_8 (T_8S_7- *Beauveria bassiana* @1×10^8 spores.ml^{-1}: 96.03 g) and T_9 (T_9S_7- *Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}: 96.09 g). All the treatments however, were on par with each other and significantly superior to T_{10} ($T_{10}S_7$- Untreated control: 87.08 g).

Table 15: Impact of storage period, treatments and their interaction on weight of undamaged seeds (g) per 100 g of seed in rice

Treatments (T)	Intervals of storage (MAS)							Mean
	S_1	S_2	S_3	S_4	S_5	S_6	S_7	
T_1	98.70^{stuvw}	$98.16^{nopqrstuvw}$	$97.65^{ijklmnopqrstuvw}$	$97.18^{ghijklmnopqstu}$	$97.12^{ghijklmnopqrst}$	$96.68^{fghijklmnop}$	$96.33^{fghijkl}$	$\mathbf{97.40^{cd}}$
T_2	99.00^{vw}	$98.46^{qrstuvw}$	$98.19^{nopqrstuvw}$	$98.04^{lmnopqrstuvw}$	$97.60^{hijklmnopqrstuvw}$	$97.24^{ghijklmnopqstu}$	$96.96^{ghijklmnopqrs}$	$\mathbf{97.93^{ab}}$
T_3	98.86^{tuvw}	$97.90^{klmnopqrstuvw}$	$97.58^{hijklmnopqrstuvw}$	$97.63^{ghijklmnopqrstuvw}$	$96.95^{ghijklmnopqrs}$	$96.57^{fghijklmno}$	95.90^{efg}	$\mathbf{97.34^{cd}}$
T_4	$98.10^{mnopqrstuvw}$	$97.40^{ghijklmnopqrstuvw}$	$97.30^{ghijklmnopqrstuvw}$	$97.16^{ghijklmnopqst}$	$97.26^{ghijklmnopqrstuv}$	$96.22^{efghijk}$	96.11^{efghi}	$\mathbf{97.08^{d}}$
T_5	98.87^{tuvw}	$97.87^{klmnopqrstuvw}$	$97.67^{ijklmnopqrstuvw}$	$96.80^{fghijklmnopq}$	$96.83^{ghijklmnopqr}$	$96.89^{ghijklmnopqrs}$	$96.35^{fghijklm}$	$\mathbf{97.33^{cd}}$
T_6	98.94^{uvw}	$98.19^{nopqrstuvw}$	$97.64^{ijklmnopqrstuvw}$	$97.20^{ghijklmnopqstu}$	$97.20^{ghijklmnopqstu}$	$97.05^{ghijklmnopqrs}$	$96.86^{ghijklmnopqr}$	$\mathbf{97.58^{bc}}$
T_7	99.02^{w}	98.57^{rstuvw}	$98.34^{pqrstuvw}$	$98.26^{opqrstuvw}$	$97.90^{klmnopqrstuvw}$	$97.39^{ghijklmnopqrstuvw}$	$97.30^{ghijklmnopqrstuv}$	$\mathbf{98.11^{a}}$
T_8	98.66^{stuvw}	$97.80^{jklmnopqrstuvw}$	$97.71^{jklmnopqrstuvw}$	$96.79^{fghijklmnopq}$	$96.79^{fghijklmnopq}$	96.14^{efghij}	96.03^{efgh}	$\mathbf{97.13^{cd}}$
T_9	98.87^{tuvw}	$97.57^{hijklmnopqrstuvw}$	$97.56^{hijklmnopqrstuvw}$	$97.32^{ghijklmnopqrstuvw}$	$96.70^{fghijklmnop}$	$96.49^{fghijklmn}$	96.09^{efghi}	$\mathbf{97.23^{cd}}$
T_{10}	95.05^{def}	94.51^{de}	93.59^{cd}	92.29^{bc}	91.10^{b}	88.00^{a}	87.08^{a}	$\mathbf{91.66^{e}}$
Mean	$\mathbf{98.41^{a}}$	$\mathbf{97.65^{b}}$	$\mathbf{97.33^{b}}$	$\mathbf{96.87^{c}}$	$\mathbf{96.55^{c}}$	$\mathbf{95.87^{d}}$	$\mathbf{95.50^{d}}$	

Factors	**C.D.**	**SE(d)**	**SE(m)**	**Factors**	**C.D.**	**SE(d)**	**SE(m)**	**Factors**	**C.D.**	**SE(d)**	**SE(m)**
MAS	0.259	0.131	0.092	TR	0.309	0.156	0.110	MAS × TR	0.818	0.413	0.292

Discussion

5. DISCUSSION

Use of insecticides for management of the stored seeds is most common, but, chemical means of pest management are not advisable in rice as the seeds are often used as food or feed at the end of the season or on losing viability. Under such conditions, the ingested residues of the pesticides could lead to health hazard and their effect on seed quality and viability is hardly known. An alternative for pest control under storage is the use of plant products which are cheap, readily available, target specific and safe to the environment and human beings.

Considering the above, the present study was undertaken to elucidate the efficacy of botanicals as seed protectants and their impact on quality and longevity of rice seed during storage. Neem leaf powder, neem seed kernel powder, sweet flag rhizome powder, manja koova rhizome powder and panal leaf powder each @ 10 g/kg of seed, insecticide spinosad @ 10 ppm/ kg of seed, inert diatomaceous earth @ 5 g/kg of seed, entomopathogenic fungi *Beauveria bassiana* and microbial agent *Bacillus thuringiensis* @ 1×10^{8} spores.ml^{-1} each, were the protectants used to treat the seeds. The experiment was conducted in the Department of Seed Science and Technology, Kerala Agricultural University (KAU), during 2019-2020. The results obtained are discussed herewith.

4.1 Quality of seeds before seed treatment.

The seed lot used to initiate the study was of good quality and fit for storage studies. The germination per cent was 95.33 per cent and the moisture content in the seed (11.80%) was well below the level recommended for safe storage in rice seed (<13.00%). The insect infestation parameters were negligible and no seed microflora was detected. The longevity and storability of the seed has been observed to be a function of initial seed quality (Gupta, 2003; Hatherley and Elmore, 2004).

4.2. Quality of seeds on seed treatment, before initiation of storage

Following treatment with seed protectants, variation in the quality parameters of seed observed, even before the onset of storage period. In a few treatments, the parameters remained unchanged after treatment, while in most treatments, a marginal increase in seed quality was observed. Seeds treated with *Beauveria bassiana* @ $\times 10^8$ spores.ml^{-1} (T_8) registered the highest germination (%), seed vigour indices (VI-I and VI-II), length of shoot and root and seedling dry weight, while seed infestation was the least in diatomaceous earth and neem seed kernel powder treated seeds. The weight of damaged seed, the number of infested seed, beetles and larvae in 100 g of seed sample and the per cent seed microflora were the least in these treatments.

However, in untreated seeds (T_{10}), a decline in quality was observed. The germination (%), length of shoot and root, vigour indices and weight of seedlings in untreated seeds decreased, while, the number of infested seeds, weight of damaged seeds, number of beetles and larvae, each per 100 gram of seed sample, was higher than the rest. Sunilkumar (2004) and Verma and Verma (2014) reported that the germination per cent, seedling vigour and storability were high, when seeds were treated, owing to the suppression of seed borne microflora and maintenance of strong membrane integrity.

Redrying the treated samples ensured that the moisture level was conducive (<12.00%) for safe storage. Ali *et al*. (2017) reported that starchy and oily seeds should be stored below 12.00 and nine per cent moisture content respectively, for safe storage of seeds.

4.3 Quality of treated seeds during storage

4.3.1 Analysis of Variance

Analysis of variance revealed the existence of significant differences in the impact of treatments, storage period and their interactions on seed quality parameters like germination, moisture content, seedling dry weight, vigour index I and II, during the storage period. Significant differences were also evident in weight of insect damaged seeds and undamaged seeds, number of beetles, larva and number of infested seeds.

4.2.1.1 Germination (%)

Germination during the storage was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.1.1 Impact of seed treatment (T)

Irrespective of the storage period, germination (%) was found to be significantly influenced by the treatments.

Seed treatment with *Beauveria bassiana* resulted in significant enhancement in germination. The results were in agreement with earlier studies by Antony (2016) in cowpea; Jaber and Enkerli (2016) in broad bean and Espinoza *et al.* in Chilli (2019).

Lee *et al*. (2010) concluded that the microbial inoculation of seeds with entomopathogenic fungi like *B. bassiana* and *Metarrhizium anisophilae,* favours germination and early emergence of seeds. According to Parsa *et al*. (2018), between treating seeds with entomopathogenic fungi *Metarhizium* species and *B. bassiana,* the latter was found to be more advantageous owing to its increased ability to become endophytic when seeds are soaked, possibly due to differential competition outcomes with seed-borne fungal endophytes. Pandey and Mishra (2018) observed that *B. bassiana* does not leave any significant amount of mycotoxins. However, unlike in the present study, it was found to affect 50 per cent of germination in green gram seeds. Hence, it can be inferred that the use of the entomopathogenic fungi can be more effective in protection of stored food grains.

Germination in seeds treated with *B. thuringiensis* and diatomaceous earth, neem seed kernel powder, manja koova rhizome powder and panal leaf powder was found to be on par with the untreated seeds. These were next best to seed treatment with *B. bassiana.*

4.2.1.1.2 Impact of storage period (S)

Irrespective of the seed treatment, germination per cent declined significantly as storage period increased.

The decline was evident in both the treated and untreated seeds. Such progressive decrease in germination and the quality of seed with the increase in ageing period is inevitable (Ramamoorthy *et al.*, 1989 and Kumar *et al.*, 1997 in pea seeds; Hussaini *et al.*, 1998 in maize; Manoharan, 1999 in chilli; Aswathi, 2015 in cowpea; Shakuntala, 2009 in sunflower; Tabatabaei, 2013 in barley; Sharma, 2017; Saidanaik and Chetti, 2018 and Singh, 2019 in rice).

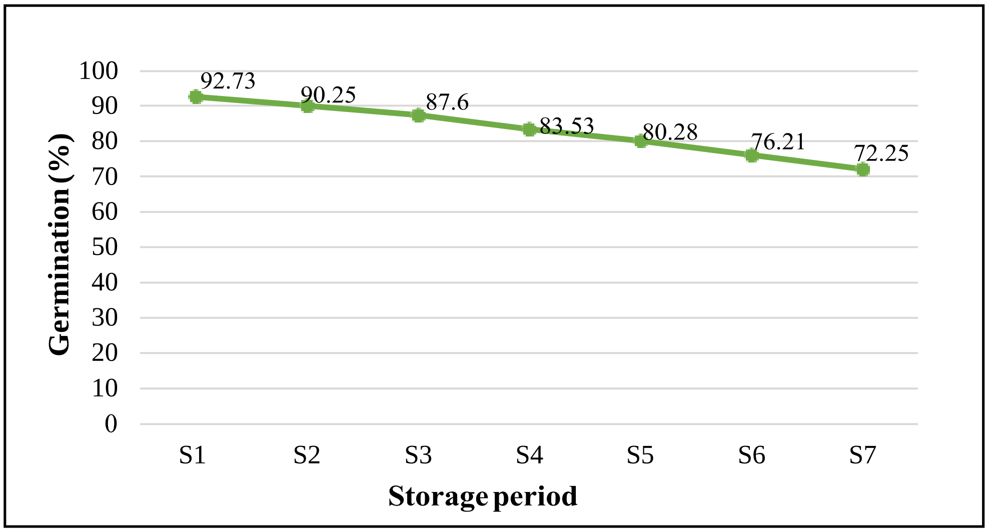
4.2.1.1.3 Impact of interaction between Treatment (T) × Storage period (S)

Germination was significantly influenced by both the treatments as well as the storage period. A significant progressive decline in germination in both treated and untreated seed over the storage period was evident (Fig. 3). This is due to the irreversible ageing process in seeds, causing the deteriorative changes in physical, physiological and biochemical characters in the seed (Abdul-Baki and Anderson, 1972; Moller *et al*., 2007; Dahuja and Yadav, 2015; Mahjabin *et al*., 2015; Garoma *et al.,* 2017; Gowda *et al.,* 2020). According to Gowda *et al*. (2020), the decline in seed germination during ageing could be related to lowering of enzyme activity within the seed leading to reduction in other seed quality parameters.

Seeds treated with *B. bassiana* exhibited significantly enhanced germination during storage (Fig. 4) compared to other treatments. It also helped to extend seed longevity by one month in comparison with seeds treated with seed protectants such as neem seed kernel powder, manja koova rhizome powder, panal leaf powder and *B. thuringiensis,* as well as untreated seeds. Longevity of seeds treated with *B. bassiana* was for six months after treatment, while in the other seed protectant treatments enumerated above, it was for a period of five months only (S_6). Considering the time- lag between harvest and the start of the experiment, the longevity of *B. bassiana* treated seed amount to 81/2 months, while that of untreated seeds was 71/2 months. However, treatment with neem leaf powder, sweet flag rhizome powder, spinosad and diatomaceous earth, had reduced seed longevity by two months in comparison to seeds treated with *B. bassiana*, *i.e.,* seed longevity in these treatments were for four months only. Asgharipour and Armin (2010) found that allelo-chemicals emancipated as residues, exudates and leaches from many plants from leaves, stems roots, fruit and seeds are reported to interfere with growth of other plants.

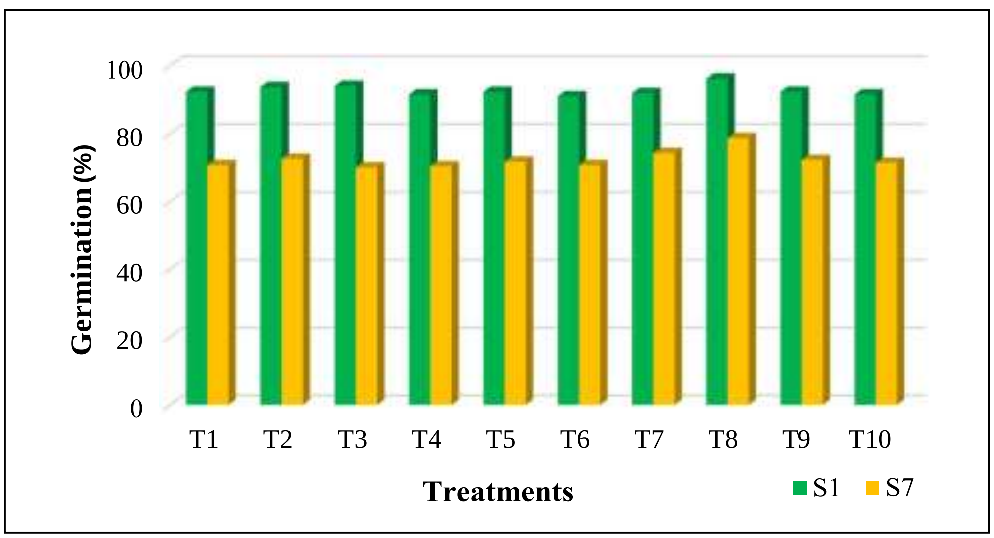
Jagadeesh *et al*. (2019), reported that the bio-inoculants can survive up to six months on pigeon pea seeds with minimum number of colonies and resulted in higher germination compared to control. Similar results were observed when wheat seeds were treated with *Trichoderma harzianum* and *Pseudomonas fluorescens* and stored for a period of six months (Chawla, 2005).

The quick loss of viability may be attributed to the elevated relative humidity and temperature prevailing during the storage period. It must be noted that the seeds were packed in jute bags. Since jute bags are not moisture impervious, the seed imbibes moisture to reach equilibrium and as a result, the seed moisture content increases leading to the deterioration of seeds. According to Sharma *et al*. (2007), seed longevity was greatly influenced by storage conditions, such as relative humidity and temperature and lowering of these parameters significantly increased the storage life of seeds. A negative correlation between seed germination, vigour and viability with high temperature and moisture content have been reported by Raikar *et al.* (2011) and Hussain *et al*. (2015) in rice seeds stored in jute bags under ambient conditions. Assefa and Srinivasan (2016) reported that the prevailing relative humidity and temperature of atmosphere greatly influenced the longevity of the seeds, since moisture content of the seeds fluctuates more in the moisture pervious containers than in the moisture vapour proof containers. As a result, there was an increase in respiration and enzyme activity, resulting in loss of food reserves before germination.



S_1- One month after storage
S_2- Two months after storage
S_3- Three months after storage
S_4- Four months after storage
S_5- Five months after storage
S_6- Six months after storage
S_7- Seven months after storage

Fig 3. Germination of seeds of rice variety Jyothi over the storage period



S_1- First month after storage
S_7- End of the storage period
T_1- Neem leaf powder @10 g/kg
T_2- Neem seed kernel powder @10 g/kg
T_3- Sweet flag rhizome powder @10 g/kg
T_4- Manja koova rhizome powder@10 g/kg
T_5- Panal leaf powder @ 10 g/kg
T_6- Spinosad @10 ppm
T_7- Diatomaceous earth @ 5 g/kg
T_8- *Beaveria bassiana* @ 1×10^8 spores.ml^{-1}
T_9- *Bacillus thuringiensis*@1×10^8 spores.ml^{-1}
T_{10}- Untreated control

Fig 4. Germination of treated seeds of rice variety Jyothi at the start (S_1) and end (S_7) of storage

4.2.1.2 Seedling root length (cm)

Seedling root length was found to be significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.2.1 Impact of seed treatment (T)

Irrespective of the storage period, seedling root length varied significantly between treatments. Seedling roots were the longest and significantly high in seeds treated with *B. bassiana* (Fig. 5). Seeds treated with *B. thuringiensis and* Spinosad produced long roots next to those treated with *B. bassiana*. The roots in these treatments were significantly longer compared to other seed protectant treatments including untreated control.

Enhancement in seedling root length on inoculation with *B. bassiana* in laboratory conditions as well as in greenhouse and field conditions were reported by Labidi *et al.,* (2015) in beans, in banana (Paparu *et al.,* 2009), french bean (Akello *et al.,* 2017). They attributed the positive impact on seedling growth to the antagonistic effect on seed borne pathogens and insects.

Treatment with manja koova leaf powder, neem leaf powder and sweet flag rhizome powder however, was found disadvantageous. Hassan *et al.* (2012) had observed that the certain botanicals exert a negative influence on germination and seedling growth due to their phytotoxic effects.

4.2.1.2.2 Impact of storage period (S)

Irrespective of the seed treatment, the root length of seedlings declined over the storage period. Similar results were also reported by Paul *et al.,* (1996) in mung bean and Beedi *et al.,* (2017) in chickpea seeds. The loss of membranal integrity and increased respiration rate during ageing resulted in seed deterioration as evident by poor seed quality and increased electrical conductivity of seed leachates over the storage period.

4.2.1.2.3 Impact of interaction between Treatment (T) × Storage period (S)

Significant difference in root length of seedlings was observed due to interaction between seed treatment and storage period**.** Results revealed that seed treatment with bio-inoculants was beneficial for better root growth. The seeds treated with *B. bassiana*, *B. thuringiensis* and Spinosad recorded significant long roots throughout the storage period. The results are in line with the reports of Pathak *et al*. (2016) in wheat. Raja *et al*. (2018) observed that seed infusion of microbial consortia resulted in increased seedling length in black gram seeds, as they have the ability to produce growth promoting agents. Qureshi *et al.* (2012) found that the co-inoculation of *Rhizobium* and *Bacillus* sp. increased the root length, root mass, number of nodules as compared to control in black gram. They attributed this positive effect on seedling growth to the increased auxin production. According to Behie and Bidochka (2014), endophytic fungus forms a mutually beneficial symbiotic relationship with the plant and boosts plant defences, while the plant in return acts as its host.

Seed treatment with neem seed kernel powder, diatomaceous earth and panal leaf powder were also found beneficial over storing seeds untreated. This may be due to reduced insect infestation and seed microflora. However, seed treatment with manja koova rhizome powder, neem leaf powder, sweet flag rhizome powder or storing them untreated were found disadvantageous. Root length in these treatments did not differ significantly for most part of the storage period.

However, in both treated and untreated seeds, the root length decreased as storage period increased. This is due to the damage caused to membranes, enzymes, proteins and nucleic acids and such degenerative changes resulted in the complete disorganization of membrane cell organelles (Roberts, 1972; Carvalho *et al.,* 2009). The deteriorative processes, leads to a reduction in the energy supply required for seedling growth (Gidrol *et al*., 1998). Similar findings were also reported by Kapoor *et al*. (2011) in rice seeds. Although the decline in root length cannot be negated by seed treatment, the results obtained point out that treating seeds before storage with bio-inoculants like *B. bassiana* and *B. thuringiensis* or with seed protectant chemical spinosad would be beneficial over storing them untreated.

4.2.1.3 Seedling shoot length (cm)

The shoot length of seedling was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.3.1 Impact of seed treatment (T)

Irrespective of the storage period, seedling shoot length varied significantly between treatments. High significant shoot length was observed in seeds treated with *B. bassiana*, while, the untreated seeds and those treated with sweet flag rhizome powder, neem leaf powder and manja koova rhizome powder registered poor shoot length (Fig. 5). As reported in the present study the ability of entomopathogenic fungi (EPF) to promote plant growth parameters was observed by several earlier workers (Bamisile *et al.*, 2018; Jaber and Enkerli, 2016, Tall and Meyling, 2018). Of late it has been observed that the entomopathogenic fungi also play a role as growth promoters and plant disease antagonism. The use of EPF is a common practice for integrated pest management. Inoculation with entomopathogenic fungi *B. bassiana* and *M. anisopliae* [(Metschn.) Sorokin 1883] were found to reduce seed contamination, improved seed germination and growth. Inoculation with entomopathogenic fungus produced seedlings with greater height and weight in chilli (Espinoza *et al.*, 2019).

Seed treatment with *B. thuringiensis* followed by Spinosad and Neem seed kernel powder and diatomaceous earth also enhanced the seedling shoot length. Efficacy of Spinosad and neem kernel powder in enhancing shoot length in cowpea was also reported by Antony (2016). However, according to Kumar (2012), germination per cent and seedling growth in pigeon pea were higher in seeds treated with thiram @ 5 g/kg of seeds and sweet flag rhizome powder as compared to the present study. They attributed this to the higher efficacy of these seed protectants against bruchid infestation.

4.2.1.3.2 Impact of storage period (S)

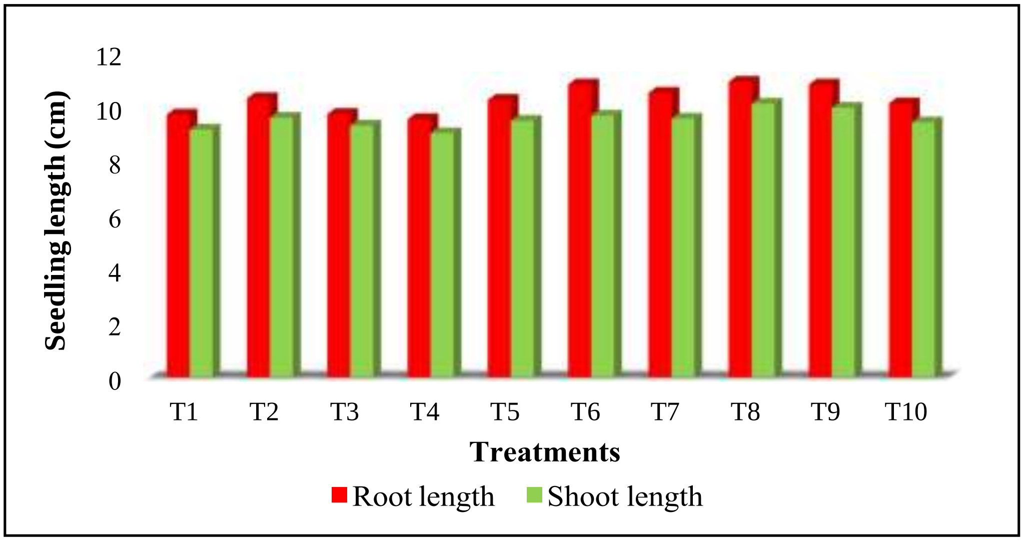
Irrespective of the seed treatments, the shoot length of seedlings decreased significantly as the seeds aged. Saidanaik and Chetti (2018) and Singh (2019) found similar results in rice seeds, due to an increase in seed leachates and loss of membrane integrity during storage, leading to deterioration. Sulthana *et al.* (2016) had also reported that seed germination and seedling growth parameters were decreased with increase in storage duration, as the cause of attaining of dormancy by seed.

4.2.1.3.3 Impact of interaction between Treatment (T) × Storage period (S)

Significant difference was found in the seedling shoot length due to the interaction of seed treatments and storage period**.**

The present findings revealed that, significant enhancement in seedling shoot length was found when seeds were treated with bioinoculants *i.e*., *B. bassiana* and *B. thuringiensis*, throughout the storage period. Minaxi *et al.* (2012) reported that bacterial seed treatment with *Bacillus* sp. improved seed germination and seedling growth parameters in cowpea seeds, as it improved their ability to live under stress environment. O'Callaghan (2016) concluded that seed treatment with beneficial microorganisms induce improvement in seed and subsequent seedling growth by reducing the mycotoxins producing fungal populations.

Seed treatment with spinosad, neem seed kernel powder, diatomaceous earth and neem seed kernel powder also resulted in better seedling growth compared to untreated seeds. Umesha *et al.* (2017) observed superior performance of seeds treated with Spinosad @ 0.04 ml/kg, neem leaf powder @ 1:20 ratio, sweet flag rhizome powder @ 10 g/kg over untreated seeds in cluster bean. They also found that, these treatments could be useful to prolong the storage life of cluster bean seeds. Seed treatment with Spinosad exhibited higher seed quality parameters (germination, root length, shoot length, mean seedling dry weight, seedling vigour index I and II and TDH activity with low electrical conductivity) up to 18 months under ambient conditions. They attributed this advantage to less membrane damage in seed due to lower insect infestation.



T_1- Neem leaf powder @10 g/kg
T_2- Neem seed kernel powder @10 g/kg
T_3- Sweet flag rhizome powder @10 g/kg
T_4- Manja koova rhizome powder@10 g/kg
T_5- Panal leaf powder @ 10 g/kg
T_6- Spinosad @10 ppm
T_7- Diatomaceous earth @ 5 g/kg
T_8- *Beaveria bassiana* @ 1×10^8 spores.ml^{-1}
T_9- *Bacillus thuringiensis*@1×10^8 spores.ml^{-1}
T_{10}- Untreated control

Fig 5. Impact of treatments on root length (cm) and shoot length (cm) of rice seedlings

However, a significant decrease in the seedling shoot length was also found in both treated and untreated seeds as the period of storage progressed. Similar results were found by Khaldun and Haque (2009) in cucumber seeds. Devi (2014) reported that the decline in shoot length of soybean seeds at the end of the storage period might be attributed to age induced decline in seed germination as well as damage caused by fungi, insects and toxic metabolites which might have hindered the seedling growth. Miah *et al*. (2006) found that the vigour index of seeds decreased with the advancement of storage period and reached to zero after two months of storage at 80.00 per cent RH. Long term storage with 92.00 per cent germination was possible at 50-60 per cent RH and low moisture (10.00%) under room temperature. Sultana *et al.* (2016) reported 97.26 per cent reduction in seedling shoot length of rice when they were stored in gunny bags for six months due to increase in seed moisture content and insect population.

4.2.1.4 Seedling dry weight (g)

Seedling dry weight during the storage period was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.4.1 Impact of seed treatment (T)

Irrespective of the storage period, seed treatments exerted significant influence on the seedling dry weight. Seed treatment with Spinosad, *B. bassiana*, neem seed kernel powder, diatomaceous earth, *B. thuringiensis* and neem leaf powder resulted in significant increase in seedling dry weight. In addition, seedling dry weight in all the treatments, including the untreated seeds, were higher than those treated with manja koova rhizome powder.

According to Sattigeri (2015), better performance observed due to treatment with spinosad is because of hyper excitation of the insect nervous system, primarily targeting binding sites on nicotinic acetylcholine receptors (nAChRs) of the insect nervous system, which is distinct from other insecticides. This property there by prevents damage to the seed. Similar results were reported by Parimala and Maheswari (2011) in maize. Lopez and Sword (2015) found an increase in certain growth parameters of cotton plants, such as dry weight owing to reduced insect infestation in the seeds treated with *B. bassiana*

Deshpande *et al.* (2010) reported higher germination, seedling length, seedling dry weight and vigour index in soybean seeds treated with neem seed powder treatment, due to existence of some insecticidal property leading to higher insect mortality with least population build up and reduced seed weight loss. Similar results were reported by Bhuiyan *et al.* (2010) in lentils, Sandeep *et al.* (2013) in sweet corn seeds and Dwivedi and Shukla (2019) in wheat seeds.

4.2.1.4.2 Impact of storage period (S)

Irrespective of the seed treatment, the dry weight of seedlings varied significantly over the storage period. The dry weight of seedlings declined over the storage period. However, dry weight during the mid-storage period was found to be on par with each other. Decrease in dry weight during storage of seed have been reported by earlier workers (Maurya *et al*., 2002; Singh and Dadlani, 2003; Agha *et al*., 2004; Autade and Ghuge, 2018; Raikar *et al*., 2011; Rahmawati and Aquil, 2020).

Filho *et al.* (2016) reported that the accumulation of the dry matter during the seedling phase may have been caused by increased seed moisture and susceptibility of the seeds to deterioration during the storage period owing to increase in temperature during storage. The negative impact of high relative humidity and temperature on seed quality has been reported by Sterlec *et al*. (2010) and Mbofung *et al*. (2013).

4.2.1.4.3 Impact of interaction between Treatment (T) × Storage period (S)

Significant difference in the dry weight of seedlings was observed due to the interaction of seed treatments and storage period

As the storage period increased, there was a significant decrease in dry weight of seedlings. Elevated temperature and relative humidity were recorded during the storage period, especially at the start of the experiment. Surki *et al.* (2012) reported that with the increase in storage temperature and storage time, the mechanisms directly connected with the processes of translocation and transformation of cotyledonary reserves into substances that can be assimilated by the embryonic axis were affected, restricting the accumulation of dry matter coming from the cotyledons. Marques *et al*. (2014) reported that, the temperature is an important factor for seeds storage, affecting the speed of biochemical processes and interfering with the water content, resulting in other processes, such as an increase of enzymes activities (hydrolytic enzymes) and free fatty acids. Also, temperature increases the enzymatic and metabolic reactions rate, causing acceleration of the seed deterioration. Jyothi *et al.* (2017) revealed that both bioagent or chemical treated seeds resulted in enhanced dry weight of seedlings when stored in polythene bags, whereas it declines when stored in jute bags owing to increase in the moisture content of seeds.

Seedling dry weight in treated and untreated seeds were found to be on par with each other over the storage period. At the end of the storage, dry weight of seedling from seeds treated with *B. bassiana* and Spinosad (0.198 g each) were found to be on par with the other seed protectant treatments. However, a significant high seedling dry weight was observed in Spinosad and *B. bassiana* treated seeds over untreated seeds.

The dry weight was comparatively low in seeds treated with panal leaf powder (0.172 g) and the least in untreated control (0.170 g). Not many reports are available on the effect of manja koova (*Curcuma angustifolia*) rhizome powder on seed germination and growth parameters. However, the inhibitory effect of related species *Curcuma longa* (turmeric) have been reported. Although, turmeric (*Curcuma* spp.) has numerous biological activities, including anticancer, antibacterial, antifungal and insecticidal properties, its potential growth- inhibitory effect has been well documented. The methanol extracts of turmeric varieties were found to inhibit the seed germination and seedling growth of radish, cress, lettuce and *Bidens pilosa* (Akther *et al.*, 2018).

The results thus indicated that treating seeds before storage with Spinosad or bio-inoculants like *B. bassiana* would be beneficial over storing them untreated. It would help decrease the decline in seedling dry weight, as the storage period prolongs.

Mishra and Sinha (2000) reported the enhanced growth and weight of rice seedling with bioagent application. Raikar *et al*. (2011) reported that the seed treatment with chemicals and botanicals will reduce the qualitative and quantitative losses besides maintaining the quality of seed for a longer period, as they have insecticidal and fungicidal properties.

4.2.1.5 Seedling vigour index I (VI-I)

Seedling vigour index I was found to be significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.5.1 Impact of seed treatment (T)

Irrespective of the storage period, seed treatments exerted significant influence on the seedling vigour index I.

Significant superior seedling vigour index I was realised on seed treatment with *B. bassiana* (Fig.6). Russo *et al.* (2019) found the significant increase in germination and yield of soybean when seeds were inoculated with *B. bassiana*, due to its ability to act as an antagonist against the potential deleterious pathogens. Similar results were obtained in field studies of corn following inoculation with entomopathogenic fungi (Kabaluk and Ericsson, 2007).

Seed treatment with *B. thuringiensis*, diatomaceous earth, Spinosad and neem seed kernel powder were next best to seed treatment with *B. bassiana*, each differing significantly from the other in their impact on vigour of the seedling. Seed treatment with neem leaf powder resulted in drastic reduction in VI-I. Similar impact was also observed on seed treatment with rhizome powders of manja koova and sweet flag. Vigour of seedlings in these treatments was lower than that observed in untreated seeds.

As observed in the study, Umesha *et al*. (2017) also reported significant enhancement vigour indices (VI-I and VI-II) of cluster bean on seed treatment with spinosad @ 0.04 ml/kg. However, unlike in the present study, they also observed that treatment with neem leaf powder and *Acorus calamus* also resulted in significant increase in VI-I over the control. Significant improvement in germination and seedling vigour of soybean on treatment with neem leaf powder @ 10 g/kg seed, was reported by Patel *et al.* (2017).

4.2.1.5.2 Impact of storage period (S)

Irrespective of the seed treatment, the seedling vigour index I varied significantly over the storage period. Seedling vigour (VI-I) declined progressively irrespective of seed treatment, the decrease being significant as ageing of the seed progressed (Fig.8). Copeland (1988) highlighted that the deteriorative changes in seed which include membrane degradation, accumulation of toxic metabolites, decreased enzymatic activity, lipid auto-oxidation, failure of repair mechanisms and genetic degradation, increases with the advancement in seed ageing. Consequently, these factors lead to reduction in viability or germinability and vigour of stored seeds.

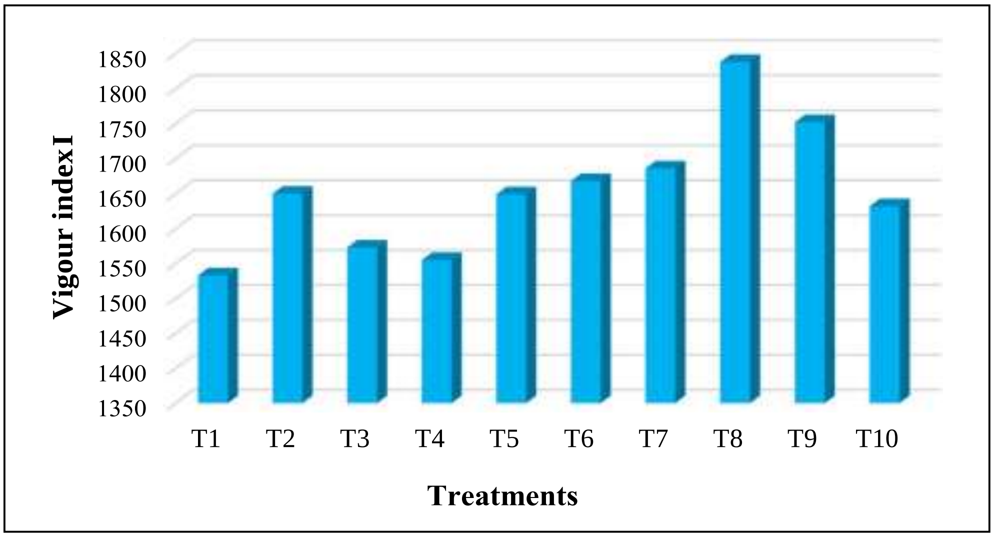
Seadh *et al.* (2019) concluded that an increase in the insect infestation over the storage period resulted in damage in membrane, enzyme, proteins and nucleic acid, ultimately leading to loss of vigour and finally results in death of seeds. Similar results were also reported by Jungtheerapanich *et al.* (2017).

4.2.1.5.3 Impact of interaction between Treatment (T) × Storage period (S)

The interaction between seed treatment and storage period significantly influenced the seedling vigour index I during storage.

The vigour of seedlings (VI-I) decreased significantly with the increase in storage period in both treated and untreated seeds. Decline in seed vigour due to ageing, irrespective of seed treatments such as seed invigoration, priming or treatment with seed health protectants have been reported earlier invariably in all seed storage experiments (Parimala, 2003; Amrutha *et al*., 2015; Sharma, 2017; Singh, 2019; Singh *et al.*, 2020).

Apart from ageing, the high relative humidity and temperature may have been instrumental in accelerating the deteriorative process in the seed during the storage period, which in turn may have led to decrease in vigour across all treatments. It is to be noted that the seeds were packed in jute bags, which have been categorised as moisture pervious packing material. As a result, the moisture content of the seeds would increase and may have further, increased the seed deterioration.



T_1- Neem leaf powder @10 g/kg
T_2- Neem seed kernel powder @10 g/kg
T_3- Sweet flag rhizome powder @10 g/kg
T_4- Manja koova rhizome powder@10 g/kg
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T_7- Diatomaceous earth @ 5 g/kg
T_8- *Beaveria bassiana* @ 1×10^8 spores.ml^{-1}
T_9- *Bacillus thuringiensis*@1×10^8 spores.ml^{-1}
T_{10}- Untreated control

Fig 6. Impact of treatments on seedling vigour index I during storage in rice

Similar results were reported in rice (Agarwal and Kharlukhi, 1985; Raikar *et al*., 2011) and in soybean (Singh and Dadlani, 2003; Agha *et al*., 2004; Autade and Ghuge, 2018). Lawrence and Maier (2010) reported that temperature and moisture content of the cereals are the two key factors affecting the quality of the seed, due to increase in biochemical reactions and loss in dry matter. Powell *et al*. (1984) reported that the low vigour or physiologically aged grain or legume seeds had increased leakage of solutes that attract fungi and also the presence of dead tissue provides a food base for infection.

A significant advantage in treating seeds with *B. bassiana* was evident throughout storage. Vigour in seeds treated with *B. thuringiensis* was also high throughout the storage period. After the initial three months of storage, treatment with *B. bassiana* however, was found to be on par with those treated with *B. thuringiensis* and at the end of storage it was also found to be on par with seeds treated with diatomaceous earth.

As the storage period increased, VI-I in *B. thuringiensis* treated seeds were found to be on par with seeds treated with Spinosad, diatomaceous earth as well as panal leaf powder. The estimate was found to be significantly low in seeds treated with powdered rhizome of manja koova, and sweet flag, powder of neem leaf, as well as in untreated control.

Endophytic *B. bassiana* was reported to enhance growth in cotton crop grown in green house following seed treatment with endophytic *B. bassiana* (Lopez and Sword, 2015). They found that *B. bassiana* systemically colonized all tissues of the host plant on seed treatment. Such colonisation following seed treatment with *B. bassiana* have been reported in several crops (Brownbridge *et al.*, 2012; Akutse *et al.*, 2013; Quesada-Moraga and Vey, 2004). Reports on the duration of survival of endophytic *B. bassiana* in the host plant is found to be varying. Jaber and Enkerli (2016) found that the fungi were retained in different plant parts for a period of one month after seed treatment while, previous studies indicated that the endophytic colonization of plants with *B. bassiana* lasted for three months in jute (Biswas *et al*., 2012), eight months in coffee (Posada *et al.*, 2007), and nine months in pine

(Brownbridge *et al*., 2012). The extent and persistence of endophytic colonization in the present study remains to be analysed.

4.2.1.6 Seedling vigour index II (VI-II)

Seedling vigour index II during the storage period was found to be significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.6.1 Impact of seed treatment (T)

Irrespective of the storage period, seedling vigour index II was significantly influenced by the seed treatment. Seed treatment with *B. bassiana* resulted in significant increment in seedling vigour (VI-II) when compared to all the other treatments (Fig.7).

Seed treatment with *B. thuringiensis,* diatomaceous earth, neem seed kernel powder, Spinosad and panal leaf powder were found to be the best. VI-II in seeds treated with diatomaceous earth, neem seed kernel powder, spinosad and panal leaf powder were also found to be on par with untreated seeds. Considerable negative impact on vigour (VI-II) of seedlings on account of seed treatment with manja koova rhizome powder, sweet flag rhizome and neem leaf powder was observed. These were however, found to be on par with untreated seeds.

The result thus pointed towards the advantage of seed treatment with *B. bassiana* to realise vigorous seedlings. Total control of seed deterioration is impossible but by providing ideal conditions, the rate of degenerative process could be slowed down to a certain extent (Umesha *et al.,* 2017).

Ownley *et al.* (2008) observed that the application of *B. bassiana* as an endophyte in tomato and cotton plants produced a significant increase in plant growth, plant stand and plant height due to reduced disease severity. Lopez and Sword (2015) and Jaber and Enkerli (2016) had also recorded similar results in their studies.

4.2.1.6.2 Impact of storage period (S)

Irrespective of the seed treatment, seedling vigour index II was significantly influenced by the storage period. Seedling vigour index II progressively declined during storage (Fig.8). Being a biological material, the loss of viability and vigour of seed during storage is an inexorable, irreversible and inevitable natural phenomenon. Accounting the diverse biological activities occurring in the seed during storage, the deteriorative processes results in impairment of germination and vigour (Umesha *et al.*, 2017). Patel *et al*. (2017) observed a steep decline in seedling vigour, when onion seeds were stored under ambient conditions, due to increase in moisture absorption. Rahmawati and Aquil, (2020) reported that vigour of pearl millet and maize seedlings could be maintained by cold storage, as low temperature and humidity will limit the seed moisture content and rate of respiration.

4.2.1.6.3 Impact of interaction between Treatment (T) × Storage period (S)

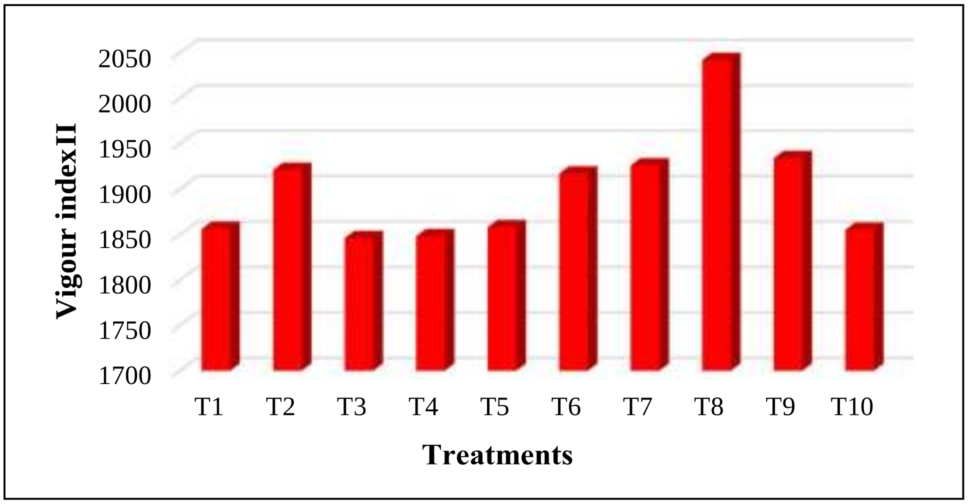
The interaction of seed treatments and storage period exerted significant influence on seedling vigour index II.

A decline in seed vigour was observed during the storage period in both treated and untreated seeds. The vigour and seed quality associated with reduction in germination over storage can be slowed down by seed treatment. Robust seeds with enhanced vigour can combat the yield-limiting effects by establishing seedlings more uniformly across a wide range of environmental conditions (Finch-Savage and Bassel, 2016).

Significant decline in seedling vigour index II was observed in all treatments as storage period progressed. Although seed treatment with *B. bassiana*, sweet flag rhizome powder, neem seed kernel powder and *B. thuringiensis* were found to register consistently high vigour index estimates throughout the storage period, they were found to be on par with all other treatments for most of the storage period *i.e.*, up to S_6. However, at the end of storage (S_7), the advantage of treating seeds with *B. bassiana* was evident. VI-II in this treatment was significantly superior over untreated control as well as those treated with panal leaf powder and rhizome powder of manja koova and sweet flag. Owing to suppression of seed borne microflora and maintenance of strong membrane integrity in the seeds treated with seed protectant, higher germination per cent, seedling vigour, field emergence and longevity is realised (Sunilkumar, 2004; Verma and Verma, 2014).

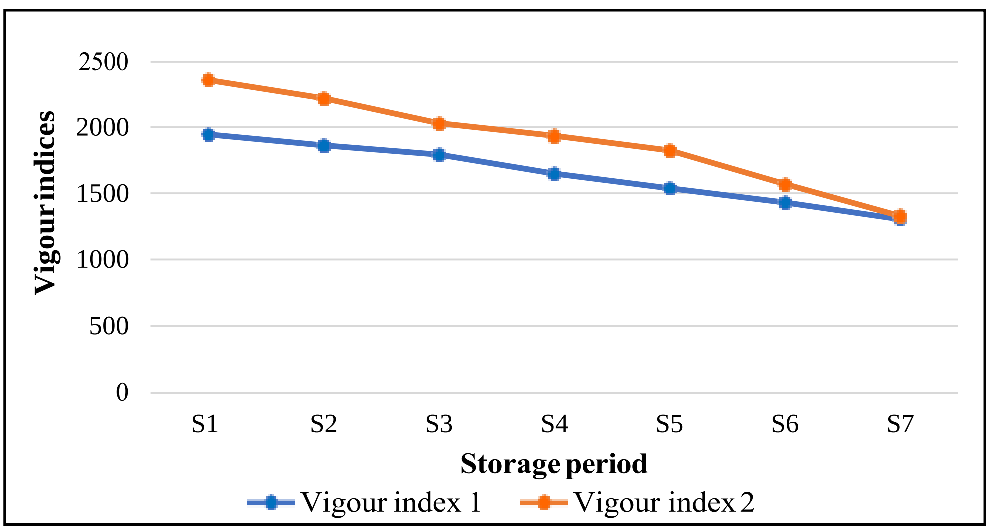
Recent studies have thrown light on the growth promoting activity of *B. bassiana* apart from its role against insect pests. In chilli, *B. bassiana* was reported to produce longer roots and higher biomass in addition to lower per cent of contaminated seeds *in vitro* and *in vivo.* (Espinoza *et al.*, 2019). Rice seeds primed with bioinoculants resulted in enhanced seed germination and seedling vigour in low vigour lots (Ramamoorthy *et al*., 2000). Din *et al.* (2014) reported that rice seeds treated with bacterial bio-agents increased the seedling vigour, due to its antagonistic ability and maintenance of membrane integrity.

Kathiravan *et al*. (2008) and Tejashwi *et al*. (2014) reported that spinosad seed treatment was effective in maintaining seed vigour due to less damage to seed membrane and lower electrical conductivity. The growth enhancing effect of several botanicals have also been reported in soybean (Babu and Ravi, 2008), in ambrette (Shakila and Rajeshwari, 2008), in black gram (Satish and Bhaskaran, 2013; Amrutha *et al*., 2015), in rice (Padmashri *et al*., 2017) and in maize (Sinha and Kumar, 2019). Antony (2016) observed that cowpea seeds treated with botanicals exhibited higher germination, more seeding length and seedling dry weight.



T_1- Neem leaf powder @10 g/kg
T_2- Neem seed kernel powder @10 g/kg
T_3- Sweet flag rhizome powder @10 g/kg
T_4- Manja koova rhizome powder @10 g/kg
T_5- Panal leaf powder @ 10 g/kg
T_6- Spinosad @10 ppm
T_7- Diatomaceous earth @ 5 g/kg
T_8- *Beaveria bassiana* @ 1×10^8 spores.ml^{-1}
T_9- *Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}
T_{10}- Untreated control

Fig 7. Impact of treatments on seedling vigour index II during storage in rice



S_1- One month after storage
S_2- Two months after storage
S_3- Three months after storage
S_4- Four months after storage
S_5- Five months after storage
S_6- Six months after storage
S_7- Seven months after storage

Fig 8. Decline in seedling vigour I and II of rice over the storage period

4.2.1.7 Seed moisture content (%)

Seed moisture content was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.7.1 Impact of seed treatment (T)

Irrespective of the storage period, the moisture content in seeds was significantly influenced by the various seed treatments.

The seed moisture content in all the treatments increased significantly with the increase in storage period, although the seeds were dried sufficiently enough to enable safe storage (<13.00%) initially. Among the treatments, seed treated with diatomaceous earth registered the least moisture content. Moisture content in this treatment was on par with seeds treated with spinosad, panal leaf powder, neem seed kernel powder and sweet flag rhizome powder. Ceruti *et al*. (2008) reported that when pearl millet seed was treated with diatomaceous earth, the moisture content of the seed decreased to 10.2 per cent, from the initial 11.0 per cent. He concluded that, the grain moisture content is maintained in a safe range when grain is protected with DE, avoiding deterioration. Befikadu (2019) reported that the reduction in moisture content observed in DE treated seeds could be due to sorptive nature of DE dust.

Significant high seed moisture content was recorded in seeds treated with manja koova rhizome powder, *B. bassiana*, untreated control and *B. thuringiensis*. A negative correlation between seed moisture content and germination, vigour and other quality parameters have been reported (Chaudary *et al*., 2014; Ali *et al*., 2017; Bhandari *et al*., 2017; Whitehouse *et al*., 2018; Mahatara *et al*., 2020). However, in the present study no such relationship was evident. Superior germination estimates and seedling growth parameters was exhibited by seeds treated with *B. bassiana.* The treatment had also resulted in of seed longevity by a period of one month over other treatments. In similar lines, seed treated with *B. thuringiensis* also resulted in enhanced seed and seedling quality. However, the seed treatments with manja koova rhizome powder, neem leaf powder and untreated seeds, that had registered low seed quality parameters during most of the storage period were found to be high in moisture content.

4.2.1.7.2 Impact of storage period (S)

Irrespective of the seed treatment, the storage period significantly influenced the seed moisture content.

A significant increase in the moisture content of seeds was obvious with the progress in storage period as they were stored in moisture pervious jute bags. In addition, the relative humidity of the storage environment was high for most part of the storage period (Fig.2). Chatta *et al.* (2012) reported that the large pore size of jute, cloth and woven polypropylene bags provide free access to the water vapour that were readily absorbed by the seeds and ultimately results in elevated seed moisture contents. Alam *et al*. (2009) reported that paddy seeds stored in USA organic cocoon, Germax cocoon, IRRI made storage bag, rexin cocoon and thick poly bag maintained lower seed moisture content below the critical level (14.00%), when compared gunny bags.

4.2.1.7.3 Impact of interaction between Treatment (T) × Storage period (S)

Significant difference in the seed moisture content was evident due to the interaction of seed protectants and storage period.

The moisture content in both treated and untreated seeds increased significantly over storage. It is to be noted that the relative humidity during storage was considerably high. As seed is hygroscopic in nature, its quality is being affected due to variations in the environmental conditions viz., relative humidity, temperature, moisture content, gaseous exchange, packaging material etc. (Doijode, 1990). Elias (2004) reported that rice grain, despite not being a good thermal conductor, as a living organism with porous intra and inter granular structures and a chemical composition which grants them hygroscopicity, are constantly trading heat and humidity with the environment air. In this manner, by the conventional storage system this grain is exposed to the air’s psychometric characteristic variations.

Throughout the storage period, seeds treated with diatomaceous earth and Spinosad had registered considerably lower estimates of seed moisture while, comparatively higher values were registered in seeds treated with *B. thuringiensis*, *B. bassiana* as well as untreated control. Although, initially, moisture content in seeds treated with spinosad was significant and low compared to treatment with manja koova rhizome powder, towards the end of storage period moisture content in all treated seeds as well as untreated seeds were on par with each other.

Absence of significant differences between treatments towards the end of storage may be due to the packaging of seeds in jute bags which are considered to be moisture pervious packaging material. The loss in seed vigour and quality is due to storage of seeds in jute bags and the increase in seed moisture content across all treatments towards the end of storage can be attributed to increase in temperature and relative humidity in the storage atmosphere. Bhandari *et al.* (2017) reported that seed germination and seedling vigour in maize seeds can be maintained for more than a year in hermetic storage, as it protects seeds from biotic and abiotic factors, due to low moisture content. Similar results were found in wheat (Naguib *et al*., 2011; Nabila *et al*., 2016). Wang *et al.* (2019) reported that when the primed seeds were stored under ambient conditions of room temperature and high relative humidity, there was a decrease in all the seed quality attributes. He concluded that the primed seeds should be stored under low temperature conditions.

As seeds in the present study were stored in jute bags, moisture is permitted inside and resulted in increased moisture content. Similar results were reported by Joshi *et al*. (2014). Warham (1986); Pham and Ramegouda (2007) reported that the moisture content of the seeds fluctuates more in the moisture pervious containers than in moisture proof containers.

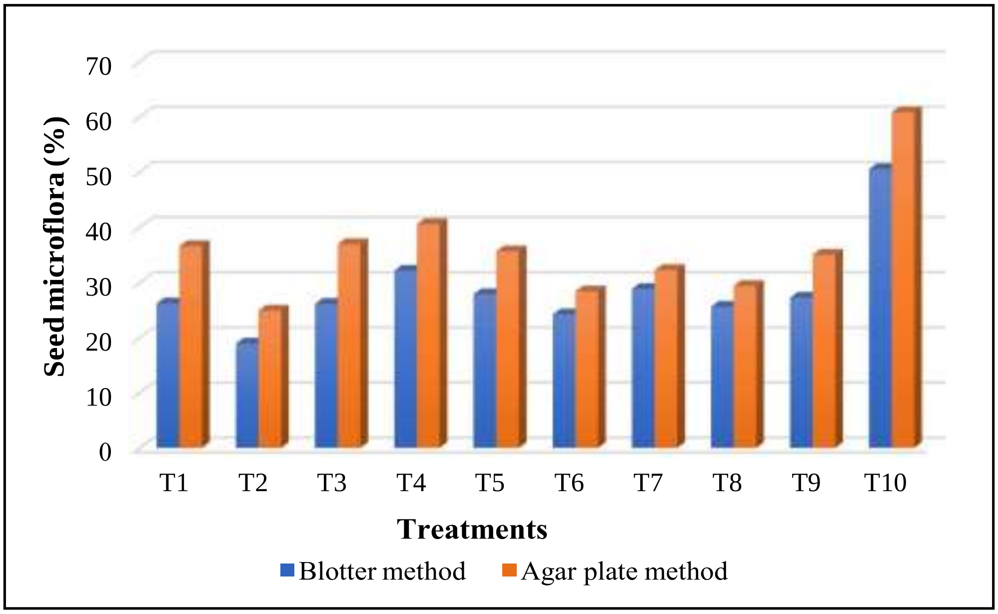
The fluctuation in moisture content might be due to the variation in atmospheric humidity and seeds absorb moisture from atmosphere, when they were stored in moisture pervious containers like gunny bags and cloth bags. Ben *et al.* (2006) reported that conventional packaging materials are porous in nature and even dried seeds can regain moisture in these packaging materials under high ambient relative humidity.

Results thus indicated that the increase in moisture content over the storage period is probably an important factor in reducing the seed viability and germinability and is in agreement with the earlier findings by Gupta (2010) in rice. The negative correlation between seed germination, vigour and viability with moisture content in storage was reported by Nagarajan and Kavitharaju (1976) in maize and sorghum seeds, Gupta (2003) in soybean seeds and Hussain *et al.* (2015) in rice seeds and several others.

4.2.1.8 Seed microflora infection (%)

Seed treatments exerted significant impact on the seed microflora infection (%). An increase in the seed microflora infection (%) was observed at the end of the storage (Fig. 9) and similar findings have been reported earlier as well (Christensen and Kauffmann, 1969 and Krishnamurthy and Raveesha, 1996). The per cent increase in microflora detected via blotter method and agar plate method varied between 18.92 and 50.55, and 24.90 and 60.84, respectively. In both cases, the least per cent infection was observed in seeds treated with neem seed kernel powder followed by those treated with spinosad (Blotter: 24.21%; Agar: 28.41%) and *B. bassiana* (Blotter: 25.60%; Agar: 29.38%), while it was the highest estimate was recorded in untreated seeds (Blotter: 50.55%; Agar: 60.84%).

The advantage in treating seeds with neem seed kernel powder in controlling the storage fungi was evident. The antifungal properties of neem seed kernel extract against many storage fungi have been reported in previous studies (Kumar *et al.,* 2011; Krishnamurthy *et.al.,* 2008). Hassan *et al.* (2015) studied the efficacy of neem seed powder against fungal pathogens and found that it was effective in controlling *Aspergillus niger, A. flavus* and *Rhizopus* species. Seed treatment with spinosad or *B. bassiana* also helped to reduce seed microflora occurrence, although the effectiveness was lower than treatment with neem seed kernel powder. Extensive studies on efficacy of fungicidal properties *of B.bassiana* is lacking. However, it has been reported that the endophytic fungus produces several toxic secondary metabolites such as bassianin, bassiacridin, beauvericin, bassianolide, beauverolides, tenellin and oosporein that infect insect cuticle, causing insect mortality (Quesada-Moraga and Vey, 2004).



T_1- Neem leaf powder @10 g/kg

T_2- Neem seed kernel powder @10 g/kg

T_3- Sweet flag rhizome powder @10 g/kg

T_4- Manja koova rhizome powder@10 g/kg

T_5- Panal leaf powder @ 10 g/kg

T_6- Spinosad @10 ppm

T_7- Diatomaceous earth @ 5 g/kg

T_8- *Beaveria bassiana* @ 1×10^8 spores.ml^{-1}

T_9- *Bacillusthuringiensis*@1×10^8 spores.ml^{-1}

T_{10}- Untreated control

Fig 9. Effect of treatments on seed microflora (%) infection of rice seeds at the end of the storage

Storing seeds untreated led to the highest per cent seed microflora when compared storing treated seeds. Masum *et al.* (2009) and Gaurilcikiene *et al.* (2012) indicated that seed treatments are important in controlling seed borne infections in wheat and pea and that the effect of seed treatments varies. The variation could be attributed to variation in effectiveness of the compound applied and phytotoxicity of the active ingredients in the seed treatments (Mancini and Romanazzi, 2014).

Irrespective of the seed treatments, the fungal colonies observed were *A. niger* and *A. flavus*. These fungi, have not been reported to cause any disease in rice crop. However, the presence of *Aspergillus* spp., especially *A. niger* and *A. flavus* on seeds of rice in higher frequencies resulted in lower germination as it is present as a saprophyte (Uma and Wesely, 2013). Frandoloso (2018) reported that the fungi seed contamination allowed the degradation of the seed, as the increase in optimal conditions for microorganisms' development favours seeds degradation. The infestation from pathogens in grains can lead to kernel discoloration and moldy odours, resulting in economic losses associated with a decrease in grain quality. The growth of mold may also increase the grain temperature, thereby not only creating a more favourable environment for pest attack, but also potentially increasing temperature to scorch or even ignite the grain (Mylona *et al*., 2012).

4.2.1.9 Number of beetles per 100 g of seed

Number of beetles were found to be significantly influenced by seed treatments, storage period and their interaction.

4.2.1.9.1 Impact of seed treatment (T)

Irrespective of the storage period, the seed treatments exerted significant influence on number of beetles in the seed. The untreated seeds recorded significantly high beetle infestations, compared to the treated seeds. All the treatments were found to be effective in reducing the insect population, however, the least was observed in seeds treated with diatomaceous earth. The result is in agreement with the previous studies, wherein partial or substantial mortality of adult insects due to the seed treatment with diatomaceous earth have been reported. Diatomaceous earth has been found effective against *Sitophilus oryzae* L. (Stadler *et al.*, 2012; Sadhegi *et al*., 2012), *S. zeamais* Mots. (Ceruti *et al.,* 2008), *S. granarius* L. (Saez and Mora, 2007), *Rhyzopertha dominica* F. (Chanbang *et al.,* 2008; Nwaubani *et al.,* 2014) and *Tribolium castaneum* Herbts (Arthur and Fontenot, 2013; Kabir 2013).

Arthur (2002) reported that the principal mode of action for inert dusts is that they cause insects to desiccate. Diatomaceous earth adheres and absorb the waxy fats and oils (Lipids) from the insect cuticle and physically damage the cuticle. This inhibits the ability of the insect to retain water and they tend to die from desiccation.

4.2.1.9.2 Impact of storage period (S)

Irrespective of the seed treatments, the storage period exerted significant influence on number of beetles in the seed sample.

A significant increase in the number of beetles was observed with the advancement of the storage period. This may be attributed to the increase in moisture content of the seeds over storage, creating an environment more conducive for the attack of storage pests. Hell and Mutegi (2011) recommended that harvested cereals should be dried to safe moisture levels of 10-13 per cent during storage to prevent growth of mycotoxigenic fungi and to reduce insect infestation. However, in the present study, seed moisture content exceeded the safe limit over storage. It ranged between 13.51 per cent ($T_{7:}$ Diatomaceous earth @ 5 g/kg) and 14.28 per cent ($T_{9:}$ *Bacillus thuringiensis* @1×10^{8} spores.ml^{-1}). This enhancement in seed moisture during storage was found to be influenced by the highly humid environment that prevailed during the storage period. Danso *et al*. (2017) reported that the micro-physical condition coupled with high grain moisture content could enhance insect pests and fungal proliferation in maize cobs when stored under ambient relative humidity above70 per cent.

4.2.1.9.3 Impact of interaction between Treatment (T) × Storage period (S)

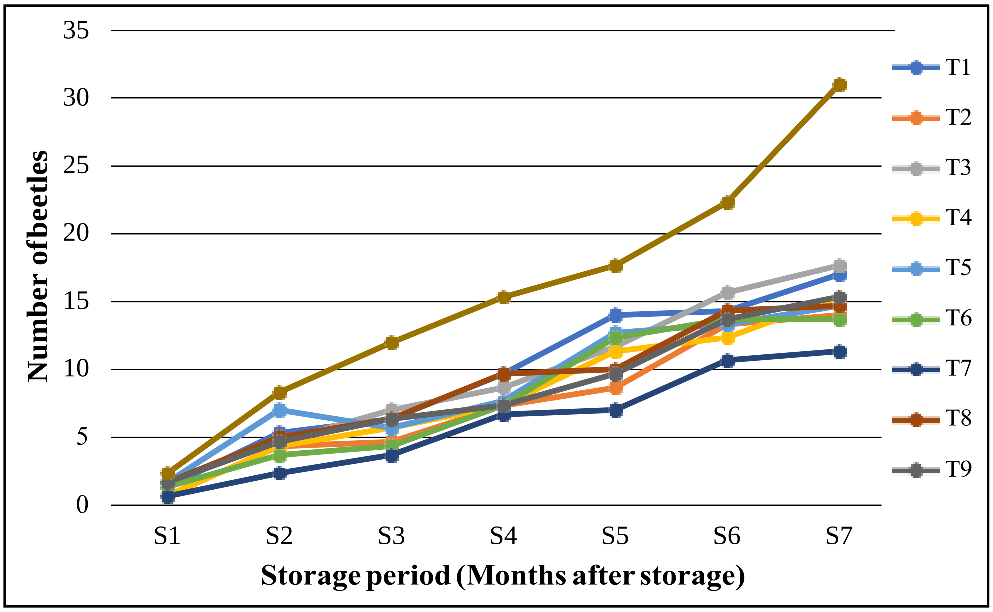
The interaction of seed treatment and storage period was found to exert significant influence on number of beetles in the seed sample.

Over the storage period, treating the seed with seed protectants was found to reduce the beetle infestation (Fig.10). The number of beetles in both the treated and untreated seeds were found to be on par with each other during the first three months after storage and after that significant differences were evident between treated and untreated seeds. Among the seed protectants used, beetle infestation was observed to be consistently low in treatments with diatomaceous earth, spinosad and neem seed kernel powder. However, no significant differences in beetle population were observed among the treated seeds during the storage period, implying the potential of treating seeds before storage in insect pest management.

Non-chemical methods are attractive since they neither leave chemical residues in the commodity nor do they cause resistance in insects. Diatomaceous earth has proved to be a promising alternative to the contact insecticides, against a wide range of storage pests (Subramanyam and Roesli, 2000; Stathers *et al.*, 2004). Botanicals and their derivatives appeared to be a safe alternative to chemical insecticides. Around 2400 plant species have been identified to possess potential pesticidical properties (Grainge and Ahmed, 1988). Neem products have been found effective in reducing the pest population. The neem materials, whether it is raw or enriched, affect the behaviour, survival and reproduction of stored product pests (Singh, 1993). Sharma (2011) found that wheat seeds treated with neem seed kernel extract was effective against *Tribolium castaneum*. Similar results were reported against *Sitophilus oryzae* (Mishra and Pandey, 2014; Priyanka *et. al.*, 2013), *Trogoderma granarium* (Odeyemi and Ashamo, 2005) and *Rhyzopertha dominica* (Khan and Marwat, 2003).

Sparkes *et al.* (2001) reported that insecticidal activity of spinosad is due to its ability to attack on nervous system of the insects and results in death of the insects. Yousefnezhad and Aasghar (2007) reported the effect of spinosad seed treatment on mortality of *T. castaneum* and *S. oryzae*. spinosad is also proven to be effective against *Rhyzopertha dominica* and *Plodia interpunctella* (Fang *et.al*., 2002; Nayak *et.al.*, 2005).

In spite of the advantage in seed protectant treatments, the beetle population increased over the storage period as observed in untreated seeds.



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T_{10}- Untreated control

Fig 10. Impact of interaction between treatment and storage period on number of beetles in rice

This increase irrespective of the effectiveness in seed treatment may be due to increase in seed moisture content, which resulted from prevailing high humidity in the atmosphere during the storage period. Kumari *et al.* (2017) found storing maize seeds in super bags prevented damage by pest infestation. They attributed this to the least variation in the moisture content in seeds thus stored. They also reported that jute bags were heavily infested with insects (45%) at the end of the storage period. The mortality rate of stored pests treated with diatomaceous earth decreased with increased relative humidity, due to reduced transpiration through the cuticle (Mewis and Ulrichus, 2001). High relative humidity above 60.00 per cent can prevent the drying action of diatomaceous earth. Reduced effectiveness of diatomaceous earth with an increase in relative humidity during the storage period has been reported (Wakil *et al.,* 2013).

This increase in storage pest population observed may be the reason for loss in seed quality attributes. Kalsa *et al.* (2019) reported that insect-infested wheat samples exhibited significantly lower mean germination (70.3%) than insect-free samples (80.5%). They also added that, damage to the seed could cause a significant reduction in seed capacity to produce a healthy seedling. The damage hastens the loss of nutrients during initial phases of seed germination and the seed fails to develop into normal seedlings. According to Likhayo *et al*. (2018), mould growth may result in serious quality changes and insect attack may reduce quality and cause weight losses, when grain with higher moisture (>14%) is stored under warm conditions. The seeds need to be stored by treating with seed protectants to reduce the stored grain pests. Storing them without treatments make seeds more susceptible for the attack of storage pests.

4.2.1.11 Number of infested seeds per 100 g of seed

Number of infested seeds per 100 g of seed varied significantly with treatments, storage period and their interaction.

4.2.1.11.1 Impact of seed treatment (T)

Irrespective of the storage period, seed treatments exerted significant influence on the number of insect infested seeds in the seed sample.

The number of infested seeds were significantly lower in treated seeds. Among the treatments, treating seeds with diatomaceous earth and neem seed kernel powder proved to be more advantageous than the others. Kabir and Wulgo (2014) also reported lower insect infestation in cowpea seeds following seed treatment with diatomaceous earth and neem seed kernel powder. However, Antony (2016) had reported the effectiveness of insecticide spinosad in controlling bruchid infestation in cowpea and the efficacy of neem seed kernel powder and neem oil in insect control was also reported.

4.2.1.11.2 Impact of storage period (S)

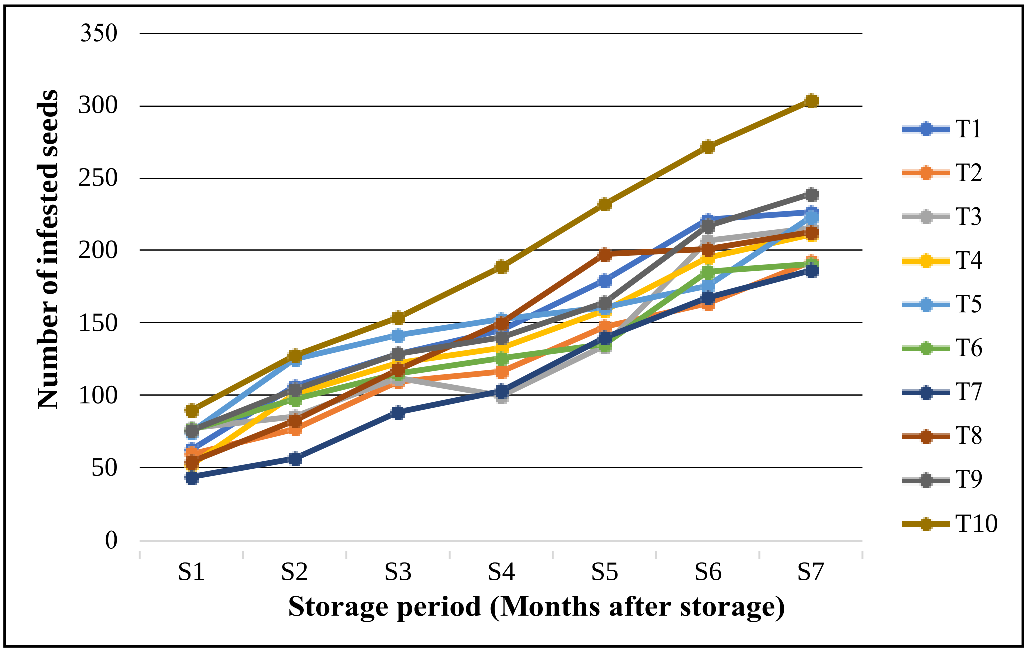
Irrespective of the seed treatment, storage period exerted significant influence on number of beetles in the seed sample.

A significant increase in the number of infested seeds was observed with the advancement in storage period in both treated and untreated seeds. This directly reflected the increase in pest population observed over the storage period which in turn was influenced by the gain in seed moisture content above the safe level. Manu *et al.* (2019) reported that the levels of insect damage to be positively correlated with seed moisture. An increase in seed moisture content resulted in an increase in the number of stored product insects. In addition, it was found that many agents that cause loss in seed are inter-related and insect activity leads to increased moisture content (Azzam *et al*., 2011)

4.2.1.11.3 Impact of interaction between Treatment (T) × Storage period (S)

The interaction between seed treatments and storage period registered significant impact on the number of infested seeds in the seed sample.

Throughout the storage period, the number of insect infested seeds was significantly high in untreated seeds than in the treated seeds, the damage due to insect infestation being on the rose as storage period increased (Fig.11).



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T_8- *Beaveria bassiana* @ 1×10^8 spores.ml^{-1}
T_9- *Bacillusthuringiensis*@1×10^8 spores.ml^{-1}
T_{10}- Untreated control

Fig 11. Impact of interaction between treatment and storage period on number of infested seeds in rice

As discussed earlier, an increase in seed moisture during storage has been attributed to increased insect damage observed during storage. Befikadu (2019) reported that, although grain moisture content recorded was below the maximum recommended (*i.e.* 13.5 %) for safe storage of maize grain, it was high enough to allow development of the insect pest species. Since in the present study, the seeds were stored in jute bags, it moisture pervious nature would have resulted in increased absorption of moisture, which is favourable for pest attack. The high porosity of gunny bag provided better aeration and resulted in an increase in the moisture content of the grain which in turn facilitated higher infestation (Ali *et al.,* 2009). Maximum insect infestation in paddy seeds was observed when they were stored in traditional storage containers, against the plastic bags and plastic drum (Hossain *et al.,* 2019). They attributed this to the persistence of high moisture content in the seed and the high level of oxygen availability in such porous packings, which favoured the growth of insects.

During the early period of storage, especially in the first two months, no significant difference in insect infestation was observed between untreated seeds and treated seeds, the exception being those treated with diatomaceous earth, neem leaf powder and *B. bassiana* in the first month, diatomaceous earth, neem seed kernel powder and *B. bassiana* in the second month. Thereafter, the insect infestation in several other treatments varied significantly from that observed in untreated seeds *i.e.,* the effect of seed protectants was more pronounced after two months of storage. Consistently throughout storage, seed treatment with diatomaceous earth was found to be the most effective in reducing insect damage. Seed treatment with botanicals neem seed kernel powder and sweet flag rhizome powder and spinosad were also found to be effective in reducing the insect damage. Sitaula *et al*. (2020) reported that the seeds treated with botanicals like sweet pepper, neem, sweet flag and turmeric recorded the minimum number of infested seeds as they possess insecticidal properties, while the maximum infestation was reported in untreated control during the storage of wheat seeds. Rajeswari and Srinivasan (2019) reported that seed treatment of rice with neem leaf, sweet flag rhizome, turmeric rhizome and rice husk recorded least per cent infestation than the control.

In a similar manner, spinosad was also effective. Hertlein *et al*. (2011) reported that spinosad successfully controls economically important beetle and moth pests associated with stored grain and is also effective against certain psocid species. Spinosad provides grain protection through control of adult or immature life stages of pest insects.

Among the seed protectant treatments, biocontrol agents were found to be less effective in reducing the number of infested seeds. In contrast, Sabbour (2011) reported that the amount of seed infestation with *S. oryzae* significantly decreased after *B. bassiana* and *M. anisopliae* seed treatment as compared to the control. The emerged adults were significantly decreased in the treated bags with entomopathogenic fungi *B. bassiana* and *M. anisopliae*. Sabbour *et al.* (2012) reported that bacteria *B. thuringiensis* significantly reduces the infestation by potato tuber moth during storage.

Nevertheless, all the mentioned seed treatments can be efficiently used for the control of insect infestation during the storage in rice.

4.2.1.12 Weight of damaged seeds (g) per 100 g of seed

Weight of damaged seeds per 100 g of seed was found to be significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.12.1 Impact of seed treatment (T)

Irrespective of the storage period, the seed treatments exerted significant influence on the weight of undamaged seeds in the seed sample.

A significant difference was observed between treated seeds and the untreated seeds with respect to weight of damaged seeds in seed sample during the entire storage duration. It was found that, the weight of damaged seeds was significantly high in the untreated seeds. Similarly, reduction in damaged seeds in treated seeds was reported in wheat and beans by Padin *et al*. (2002), and in maize seeds by Padmashri *et al*. (2017).

Diatomaceous earth followed by neem seed kernel and spinosad were found to be most effective treatment in reducing the insect infestation *i.e*, in lowering the weight of damaged seeds. Although all the treatments were effective in lowering the damage, treating seeds with manja koova rhizome powder, *B. bassiana* and *B. thuringiensis* were found to be less effective compared to the superior treatments specified above.

Jean *et al*. (2015) reported that the diatomaceous earth formulation FossilShield significantly reduced grain damage from *S. zeamais* infestation, with the treated samples recording a smaller number of damaged grains than the controls. The samples treated with this dust showed no grain damage, and correspondingly no weight was lost. However, unlike in the present study, several reports on the effectiveness of *B. bassiana* and *B. thuringiensis* in control of storage pests has been reported (Oluwafemi *et al.*, 2009; Sabbour and Solieman, 2014; Mallik and Raisat, 2014; Bello *et al.*, 2017).

4.2.1.12.2 Impact of storage period (S)

Irrespective of the seed treatments, the storage period exerted significant influence on the weight of undamaged seeds in the seed sample.

The present study revealed a significant and progressive increase in weight of damaged seeds with an increase in the storage period. The damaged seed weight during storage due to insect infestation increased 2.46 folds, as compared to the estimate before treatment. The surge in the weight of damaged seeds over the storage period was due to the increase in the number of beetles infesting the seed. Kim and Kossou (2003) stated that grain weight loss has positive correlation with increase in insect population.

Ahmad *et al.* (2017) reported that as the storage period increased, there was increase in the kernel damage of wheat grains. The weight loss in grains recorded was 20.00 per cent, 8.00 per cent and 2.50 per cent by *S. oryzae*, *T. granarium* and *T. castaneum* respectively.

4.2.1.12.3 Impact of interaction between Treatment (T) × Storage period (S)

The interaction of seed treatment and storage period exerted significant influence on the weight of damaged seeds in the seed sample.

Lower weight of damaged seeds was recorded in the treated seeds, compared to the untreated seeds (Fig.12). But even in the treated seeds, a surge in weight of damaged seeds was observed with respect to storage period. This is because of the increased number of insects and an increase in moisture content of seeds. Increased pests can damage a greater number of seeds. Nawrot *et al*. (2012) found that the degree of damage is directly related to the insect infestation rate. As the population increased, debris and damaged kernels also increased. In the present study, the elevated temperature and relative humidity may have contributed indirectly to increasing the insect damage over the storage period through enhancing the rate of moisture imbibition above the safe level for storage. The conducive environment created for insect multiplication and resulted in an increase in damaged seeds noticed in both the treated and untreated seeds.

Subedi *et al*. (2009) reported that the per cent grain damage (18.75%) due to weevil infestation was higher under room temperature (25 ± 3ºC) in case of polished rice compared to controlled condition. Compton *et al.* (1998) reported that *S. zeamais* caused maximum per cent grain damage (85 - 93%) at 60 days after release in maize under room temperature condition. This was attributed to the susceptibility of the hosts and conducive climatic conditions (28°C and 65% RH).

All the seed treatments were effective in reducing the damage caused by the storage pests during the storage. Among the seed protectant treatments, diatomaceous earth, neem seed kernel powder and spinosad had registered comparatively lower weight of damaged seeds. Considering the initial estimate, the increase in damaged seeds at the end of storage (S_7) was lower in seeds treated with diatomaceous earth (1.06 folds), neem seed kernel powder (1.33 folds) and spinosad (1.41 folds), compared to untreated control (8.93 folds).

Pacheco *et al.* (2015) reported that diatomaceous earth seed treatment in beans resulted in highest mortality of bruchids than in control. It showed 1.18 per cent and 1.38 per cent repellency during seventh and eighth month after storage. In the present study, the botanical seed treatment also has similar effects in reducing the weight loss caused by the storage pests. Pramanik *et al.* (2009) reported that the leaf, stem bark, stem wood and roots of *Glycosmis pentaphylla* were having strong repellency against *T. castaneum*. The highest repellency (60.99%) was recorded in neem leaf powder. Gadewar *et al.* (2017) reported the effectiveness of seed treatment of rice with sweet flag rhizome powder against *S. oryzae*. The treated seeds recorded minimum weight loss (0.40%) with maximum adult mortality (97.00%), when compared to control (18.00%-weight loss). Singh *et al*. (2017) reported that maximum inhibition of rice weevil (92.58%) occurred when seeds were treated with neem seed kernel powder. The treatment also showed minimum grain damage (21.16%), weight loss (1.15%) and adult emergence (16.17%), whereas, the untreated control registered maximum grain damage of 21.16 per cent, 13.83 per cent of weight loss and maximum adult emergence (157.58 adults). They concluded that seeds can be stored with minimum loss by seed treatment with neem seed kernel powder. Similar results were reported by Jhala *et al.* (2018) against rice moth. They concluded that the effectiveness of botanicals is due to their anti-feedant and repellency action.

Padin *et al.* (2002) reported that seed treatment with *Beauveria bassiana* resulted in significant reduction in storage pest infestation. The weight loss due to insect feeding reduced by 81.50 per cent when compared to the untreated seeds.

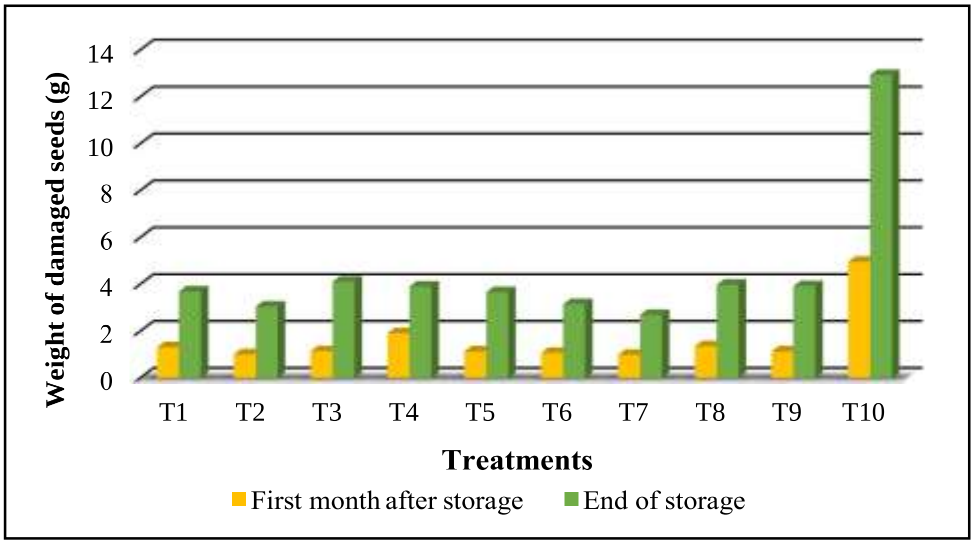
4.2.1.13 Weight of undamaged seeds (g) per 100 g of seed

Weight of undamaged seeds per 100 g of seed was significantly influenced by the seed treatment, the storage period and their interaction.

4.2.1.13.1 Impact of seed treatment (T)

Irrespective of the storage period, the seed treatments exerted significant influence on the weight of undamaged seeds in the seed sample.

A significant difference was observed between treated seeds and the untreated seeds with respect to the weight of undamaged seeds in seed sample during the entire storage duration. It is found that, the weight of undamaged seeds was significantly low in the untreated seeds.



T_1- Neem leaf powder @10 g/kg
T_2- Neem seed kernel powder @10 g/kg
T_3- Sweet flag rhizome powder @10 g/kg
T_4- Manja koova rhizome powder@10 g/kg
T_5- Panal leaf powder @ 10 g/kg
T_6- Spinosad @10 ppm
T_7- Diatomaceous earth @ 5 g/kg
T_8- *Beaveria bassiana* @ 1×10^8 spores.ml^{-1}
T_9- *Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1}
T_{10}- Untreated control

Fig 12. Weight of damaged seeds (g) at start (S_1) and end (S_7) of storage in rice

Diatomaceous earth followed by neem seed kernel and spinosad were found to be most effective treatment in reducing the insect infestation thereby a higher the weight of undamaged seeds was realised in these treatments compared to untreated control. Although all the treatments were effective in lowering the damage, treating seeds with manja koova rhizome powder, *B. bassiana* and *B. thuringiensis* was found to be less effective compared to the superior treatments specified above.

Ibrahim *et al*. (2012) reported that seed treatment of groundnut with diatomaceous earth and botanicals resulted in reduced seed damage. Lower weight loss and seed damage was recorded in these treatments when compared to the untreated seeds, due to their efficacy to cause mortality in *Callosobruchus subinnotatus.* Odeyemi and Ashamo (2005) reported that the per cent damage and weight loss caused by *T. castaneum* is reduced in the neem leaf (0.84%) and kernel (0.60%) treated seeds when compared to control (2.27%). Unlike in the present study, several reports on the efficacy of *B. bassiana* and *B. thuringiensis* in the control of storage pests have been reported (Cherry *et al*., 2004; Oluwafemi *et al.*, 2009; Sabbour and Solieman, 2014; Mallik and Raisat, 2014; Bello *et al.,* 2017). All the seed protectants were effective in reducing the weight loss during storage of rice.

4.2.1.13.2 Impact of storage period (S)

Irrespective of the seed treatments, the weight of undamaged seeds in the seed sample varied significantly and decreased progressively during the storage period.

The present study revealed a significant and progressive decrease in weight of undamaged seeds with an increase in the storage period. As compared to the estimate before treatment, the per cent decrease in undamaged seed weight during storage due to insect infestation amounted to 3.24 per cent. The decrease in weight of seeds can be attributed to increase in the number of storage pests and the difference between treatments. Keskin and Ozkaya (2015) reported that due to the increasing insect population during the storage period, kernel weight, test weight and fat content decreased in wheat seeds.

4.2.1.13.3 Impact of interaction between Treatment (T) × Storage period (S)

The interaction of seed treatment and storage period exerted significant influence on the weight of undamaged seeds in the seed sample.

Higher weight of undamaged seeds was recorded in the treated seeds, compared to the untreated seeds. The increase in the moisture content of seeds, allowed the development of a greater number of storage pests and increased pests damaged more number of seeds, leading to reduced weight in undamaged seeds. Reed *et al.* (2007) reported that grain moisture and temperature in the storage environment was the reason for infections by storage pests, moulds, development of kernel damage and changes in atmospheric gases within the grain masses. The rate of increase in new kernel damage was as high as 3.30 per cent per week. Similar results were reported in rice by Genkawa *et al*. (2008).

In the present study, as the seeds were stored in the jute bags, an increase in seed moisture content is inevitable. The jute bags are pervious to moisture and can be easily accessed by the insects as they have perforations. Howlader *et al.* (2004) reported that the highest insect population, seed damage and weight loss were recorded in gunny bags, when compared to metal structures and polythene or plastic bags during the storage of rice, due to high seed moisture content in case of seeds stored in gunny bags. Tang and Ngome (2015) found that the increase in damaged seeds was high and constant in jute bags and cloth bags. The high temperature and raised humidity levels are responsible for the growth and development of insects inside the stored rice.

However, it was found that, the weight of undamaged seeds declined during the storage period in both treated and untreated control.

All the seed treatments were effective in reducing the damage caused by the storage pests during the storage. Among the seed protectant treatments, diatomaceous earth, neem seed kernel powder and spinosad had registered comparatively higher weight of undamaged seeds. The highest per cent reduction of undamaged seeds compared to the initial estimate before treatment, amounted to 11.77 per cent in T_{10} (Untreated control). Among the treatments, the weight reduction in undamaged seeds varied between 1.42 per cent in T_7 (Diatomaceous earth @ 5 g/kg) and 2.84 per cent in T_3 (Sweet flag rhizome powder @ 10 g/kg). Reduction was also noticed in treatments T_2 (Neem seed kernel powder @ 10 g/kg: 1.76%), T_6 (Spinosad @ 10 ppm: 1.86%) and T_1 (Neem leaf powder @ 10 g/kg: 2.40%).

Befikadu (2019) reported that the grain weight loss reduction in stored maize treated with diatomaceous earth treatment reflects a reduction in insect infestation that could have caused grain weight loss by consuming grains. This is due to drying effect of DE on insect cuticle.

Mondal *et al.* (2018) reported that weight loss in chickpea seeds was found to be nil, when seeds were treated with spinosad. The untreated seeds were susceptible to bruchids infestation and recorded 4.33 per cent weight loss, with maximum per cent infestation (57.33%). They concluded that seed treatment with spinosad can reduce the damage caused by the storage pests. Similar results were reported by Vayias *et al*. (2009) and Hertlein *et al*. (2011) against the coleopteran stored-grain pests. Mohan *et al.* (1990) reported that maize treated with deoiled neem seed kernel powder at 0.1 per cent had no grain damage by *S. oryzae*. Neem leaf powder at 1.0 per cent (w/w) was found to be the most effective treatment against *S. oryzae*. Mishra and Pandey (2014) reported that the per cent grain damage (16.02%) and weight loss (13.13%) in wheat were least in neem kernel treated seeds. The untreated seeds were susceptible to weevil infestation and recorded high grain damage (29.60%) and weight loss (20.99%) at 90 days after storage.

Low seed quality observed in the untreated seeds may be attributed to the higher insect damage observed. Guenha *et al*. (2014) reported a significant 38.25 per cent drop in the germination potential in traditional storage, while under hermetic storage, that reduction remained within the acceptable values of 13.9-17.5 per cent. The decrease in germination potential is due to increase in the insect infestation (53.94 adults/kg of seed) and per cent loss (3.44%) in the traditional storage.

Storage of treated seeds is better in reducing the weight loss caused by the storage pests, over storing them untreated.

Ranking of seed treatments considering the seed quality parameters and insecticide efficacy

The treatments were ranked considering the seed quality parameters and insecticide efficacy as mentioned in section 3.6.1.2. The results are furnished in Appendices 3, 4 and 5.

Considering the impact on seed quality parameters like mean germination (%), length of root and shoot of seedling, seedling vigour indices, moisture content (%) and seed microflora (%), as well as weightage for seed longevity, seed treatment with microbial formulations were found most effective in reducing the seed deterioration. Treatment with *B. bassiana* (Rank 1), followed by *B. thuringiensis* (Rank 2) and neem seed kernel powder (Rank 3) were found most effective, while, treatments with neem leaf powder (Rank 8) and manja koova rhizome powder (Rank 9) as well as untreated control (Rank 7), were found disadvantageous.

Ranking of treatments on considering parameters like number of beetles, number of infested seeds, weight of damaged and undamaged seeds revealed that all the treatments were more effective in controlling the storage pests. Leaving seeds untreated (Rank 10) was found to be highly disadvantageous. Among the seed protectant treatments, the maximum insecticidal efficacy was exhibited by diatomaceous earth (Rank 1), followed by neem seed kernel powder (Rank 2) and spinosad (Rank 3), while seed protectant manja koova rhizome powder (Rank 9) was the least effective.

Since, seed longevity and maintenance of seed quality during storage is of paramount importance. A combined ranking of the treatments was done considering the total scores obtained by the treatment in the above rankings.

It was obvious that treatment with *B. bassaina* ranked the highest. It had extended the seed longevity by a period of one to two months over all other treatments as well as significantly superior to all other treatments in maintaining seed quality during storage. However, the insecticidal effect was low.

Seed treatment with neem seed kernel powder and diatomaceous earth ranked (Rank 2) next to best *B. bassiana*, while treatment spinosad ranked third. However, as treatment with both diatomaceous earth and spinosad had drastically reduced seed longevity compared to others, seed treatment with neem seed kernel powder would be the next best alternative to *B. bassiana.* The longevity of neem seed kernel powder treated seeds was one month more than the diatomaceous earth and spinosad treated seeds.

Mechanical, biocontrol and chemical approaches have been advocated to manage the storage pests in rice. Among these, seed treatment using protectants and fumigation remains most popular. Rampant use of insecticides often results in the development of pesticide resistance, hazardous effects on non-target organisms and is environmentally unsafe. In addition, the farming community tend to use the left-over seeds as food and feed. Under such circumstances, seed treatment with chemical pesticides is not advisable. The tendency to use botanicals and less toxic insecticides is on the rise.

In view of the above, the findings of the present study are highly valuable and in line with present emphasis on reducing the use of toxic chemicals in crop production. Use of entomopathogenic fungi *B. bassiana* @ 1×10^{8} spores.ml^{-1} and the botanical neem seed kernel powder @ 10 g/kg, were found as the best treatments in reducing the rate of seed deterioration during storage in rice. Of the two, treatment with neem seed kernel powder @ 10 g/kg accorded better control of stored pest infestation in rice seeds.

Summary

6. SUMMARY

Rice (*Oryza sativa* L.) is the major food crop consumed by over two-thirds of world population. Safe storage of seeds over a long period of time has always remained as a great challenge. Seed being a living entity, degradation in both quality and quantity, is inevitable during storage. Insect pests are one of the major biotic factors that degrade the seed during storage. Seed treatment with chemical pesticides or botanicals are options recommended to safe-guard the stored seeds from insect-pests. Chemical means of pest management is often not resorted to, as it is ecologically unsound. Considering the above, the present study 'Impact of seed protectants on seed longevity and storage pests in rice (*Oryza sativa* L.)' was carried out at Kerala Agricultural University (KAU), Vellanikkara during 2019-2020.

Seed of red kernelled rice variety Jyothi was treated with various seed protectants, in order to assess the impact of seed protectants on seed quality, longevity and their efficacy against the storage pests in rice. The seed protectants used were powdered botanicals like neem leaf powder (T_1), neem seed kernel powder (T_2), sweet flag rhizome powder (T_3), Manja koova rhizome powder (T_4) and panal leaf powder (T_5) each @ 10 g/kg of seed, insecticide Spinosad @ 10 ppm/kg of seed (T_6), inert diatomaceous earth @ 5 g/kg of seed (T_7), entomopathogenic fungi *Beauveria bassiana* (T_8) and microbial agent *Bacillus thuringiensis* (T_9) each @ 1×10^8 spores.ml^{-1}. The untreated seeds served as control (T_{10}). Both the treated and untreated seeds were dried to less than 12 per cent moisture, before being packed in jute bags and stored under ambient conditions. The parameters on seed quality and insect pest incidence of the stored seeds were evaluated for a period of seven months. The findings of the study are summarized below:

I. Quality of seeds before storage

1. The seed lot used to initiate the study was of good quality and fit for storage studies and the quality parameters were well above the minimum standards advocated for rice crop. The insect infestation parameters like number of beetles (2.00) and infested seeds (48.00) were negligible and no seed microflora was detected.

2. Seed quality parameters were assessed immediately after treating the seeds. The quality parameters of the treated seeds varied among themselves.

3. Seeds treated with *B. bassiana* (T_8) registered the highest germination (%), length of shoot and root and seedling dry weight and seed vigour indices (VI-I and VI-II), while insect infestation was the least in diatomaceous earth (T_7) and neem seed kernel powder (T_2) treated seeds. The weight of damaged seed, the number of infested seeds and beetles in 100 g of seed sample were least in these treatments. The seed moisture content after treatment was brought below the level advocated for safe storage.

II. Quality of treated seeds during storage

A. Impact of seed protectants on seed quality and longevity of rice seeds during storage

Analysis of variance revealed the existence of significant difference in the impact of various treatments, storage period and their interaction on seed quality parameters like germination (%), vigour index I and II, moisture content (%) and seed microflora (%).

A(i). Impact of seed treatment (T)

1. Irrespective of the storage period, seed treatment exhibited significant influence on seed quality parameters.
2. Significant enhancement in germination (%), seedling length (cm), dry weight (g) and vigour was obtained in the treatment with *B. bassiana*. Treatment with *B. thuringiensis* and neem seed kernel powder had recorded higher seedling growth characters and seed vigour indices (I and II). The per cent microflora was observed to be significantly low in these treatments.
3. In addition to storing seeds untreated, treatment with neem leaf powder (T_1), sweet flag rhizome powder (T_3) and manja koova rhizome powder (T_4) were registered significant lower germination (%), vigour index I and II.

A(ii). Impact of storage period (S)

1. Irrespective of the seed treatment, the storage period exerted significant influence on the seed quality parameters.
2. A decline in seed quality was observed in both treated as well as untreated seeds. A significant decrease in germination (%), seedling length (cm), dry weight (g) and vigour indices (I and II) was evident with the increase in the storage duration, while the moisture content (%) and microflora (%) increased with the increase in the storage period.

A(iii). Impact of interaction between treatments (T) and storage period (S)

1. The seed quality was significantly influenced by the interaction between treatments and storage period.
2. Seeds treated with *B. bassiana* (T_8) exhibited significantly superior seed quality throughout the storage period as well as extended the seed longevity. Longevity of seeds treated with *B. bassiana* was for six months in storage *i.e.*, 8.5 months from harvest.
3. Seeds treated with protectants such as neem seed kernel powder (T_2), manja koova rhizome powder (T_4), panal leaf powder (T_5) and *B. thuringiensis* (T_9), as well as untreated seeds (T_{10}) retained germination above 80.00 per cent only for 5 months only (7.5 months from harvest).
4. Treatment with neem leaf powder (T_1), sweet flag rhizome powder (T_3) and spinosad (T_4) and diatomaceous earth (T7) was found detrimental to seed longevity and negatively impacted seed quality.

B. Impact of seed protectants on storage pests during storage of rice

Analysis of variance revealed the existence of significant difference in the impact of treatments, storage period and their interaction on the number of beetles and infested seeds, weight of insect damaged seeds and undamaged seeds, over the storage period.

B(i) Impact of treatment (T)

1. Irrespective of the storage period, the seed treatments exerted significant influence on number of beetles and infested seeds, weight of damaged (g) and undamaged seeds (g).
2. All the seed protectant treatments exhibited significant higher efficiency in reducing the insect population and their infestation during storage compared to the untreated control.
3. Among the seed protectant treatments, treating seeds with diatomaceous earth (T_7), neem seed kernel powder and spinosad resulted in significant low beetle population and damage due to insect infestation.

B(ii) Impact of storage period (S)

1. Irrespective of the seed treatments, the number of beetles, number of infested seeds, weight of damaged (g) and undamaged (g) seeds in the seed sample varied significantly with storage period.
2. The storage duration exerted strong influence on the amount of infestation. A progressive increase in the beetles and quantity of infested seeds with a concomitant decline in the weight of undamaged seeds (g) was observed.
3. Compared to initial estimates before treatment, by the end of the storage (S_7), the weight of damaged seed during storage due to insect infestation increased 2.46 fold and the per cent decrease in undamaged seed weight amounted to 3.24 per cent.

B(iii). Impact of interaction between treatments (T) and storage period (S)

1. The interaction of seed treatment and storage period exerted significant influence on the number of beetles, number of infested seeds, weight of damaged (g) and undamaged (g) seeds in the seed sample.
2. Storing seeds untreated had resulted in significant high insect infestation by seeds and associated seed damage.
3. Among the seed protectants used, treating seeds with diatomaceous earth (T_7), neem seed kernel powder (T_2) and spinosad (T_6) conferred relatively higher protection, although no significant differences were observed among the treated seeds during

the storage period. Consistently, seed treatment with diatomaceous earth (T_7) was found to be the most effective in reducing insect infested seeds, throughout the storage.

Evaluation of treatments based on their impact on seed quality and insecticidal efficacy

1. Although, the insecticidal effect was low, treatment with *B. bassiana* was considered best among the treatments as it had extended the seed longevity the maximum *i.e.,* by a period of one to two month over all other treatments as well as found significantly superior to all other treatments in maintaining seed quality during storage.

2. Seed treatment with neem seed kernel powder was the next best alternative to *B. bassiana.* The longevity of neem seed kernel powder treated seeds was one month more than that observed in the diatomaceous earth and spinosad treated seeds. These treatments had exhibited considerable high efficiency in control of insect population and associated seed damage.

Future line of work:

1. Impact of the seed protectants found superior in the present study, on seed quality and insect damage control in seeds stored under controlled environmental conditions may be studied.
2. Being a high humid and high temperature prone area, the study of the seed protectants on storability and quality of seeds packed in moisture impervious containers may be assessed.
3. The physiological, biochemical and molecular basis for superior performance of seeds on seed treatment need to be deduced.
4. The economic analysis of the qualitative and quantitative loss in stored seed and the advantage conferred by the seed protectant treatments during storage need to be assessed.

Plate 2: Detection of seed microflora

Blotter paper method

Agar plate method

Seed infection by microflora

Aspergillus niger

Aspergillus flavus

Plate 3: Rice seed infestation by storage pests

Red flour beetle

Tribolium castaneum

Lesser grain borer

Rhyzopertha dominica

Rice weevil
Sitophilus oryzae

Insect damaged seeds

Insect infestation

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Appendices

Appendix 1: Fold change in the weight of damaged seeds during storage in rice

Treatments	Initial weight (g) (before treatment)	Final weight (g) (at the end of storage)	Fold change increase in weight
T_1	1.30	3.67	1.82
T_2	1.30	3.04	1.33
T_3	1.30	4.10	2.10
T_4	1.30	3.89	1.99
T_5	1.30	3.65	1.80
T_6	1.30	3.14	1.41
T_7	1.30	2.68	1.06
T_8	1.30	3.96	2.04
T_9	1.30	3.91	2.00
T_{10}	1.30	12.92	8.93

Appendix 2: Per cent reduction in the weight of undamaged seeds during storage in rice

Treatments	Initial weight (g) (before treatment)	Final weight (g) (at the end of storage)	Per cent reduction in weight
T_1	98.70	96.33	2.40
T_2	98.70	96.96	1.76
T_3	98.70	95.9	2.84
T_4	98.70	96.11	2.62
T_5	98.70	96.35	2.38
T_6	98.70	96.86	1.86
T_7	98.70	97.3	1.42
T_8	98.70	96.03	2.70
T_9	98.70	96.09	2.64
T_{10}	98.70	87.08	11.77

Appendix 3: Weighted ranking of treatments based on seed quality parameters

Treatment	Common name	Germination	Longevity	Root length	Shoot length	Dry weight	VI-I	VI-II	Moisture	Microflora	Total Score	Rank
T_1	Neem leaf powder	4.00	3.00	6.00	7.00	0.75	7.00	2.50	2.25	3.25	35.75	8
T_2	Neem seed kernel powder	1.75	2.00	4.00	4.00	1.00	3.75	1.50	1.25	1.00	20.25	3
T_3	Sweet flag rhizome powder	2.75	3.00	6.00	6.00	1.75	6.00	3.75	1.25	3.25	33.75	8
T_4	Manja koova rhizome powder	2.75	2.00	7.00	8.00	3.00	6.75	5.00	4.00	5.00	43.50	9
T_5	Panal leaf powder	1.75	2.00	4.00	5.00	1.75	3.75	1.25	1.25	3.25	24.00	4
T_6	Spinosad	4.00	3.00	2.00	3.00	1.00	2.75	1.50	1.00	2.00	20.25	5
T_7	Diatomaceous earth	2.00	3.00	3.00	4.00	0.75	3.00	1.75	1.00	2.25	20.75	6
T_8	*Beauveria bassiana*	1.00	1.00	1.00	1.00	1.00	1.00	1.00	3.25	2.00	12.25	1
T_9	*Bacillus thuringiensis*	2.00	2.00	2.00	2.00	0.75	2.00	2.00	4.00	3.25	20.00	2
T_{10}	Untreated (control)	1.75	2.00	5.00	5.00	1.75	5.00	2.50	4.00	6.00	33.00	7

Appendix 4: Weighted ranking of treatments based on insecticidal efficacy

Treatment	Common name	Number of beetles	Number of infested seeds	Weight of damaged seeds	Weight of undamaged seeds	Total	Ranking
T_1	Neem leaf powder	1.25	3.50	3.25	2.75	10.75	5
T_2	Neem seed kernel powder	1.25	1.25	1.25	0.75	4.50	2
T_3	Sweet flag rhizome powder	2.00	2.25	3.25	2.75	10.25	4
T_4	Manja koova rhizome powder	2.00	6.00	4.00	4.00	16.00	9
T_5	Panal leaf powder	2.00	6.25	3.25	2.75	14.25	8
T_6	Spinosad	1.25	2.25	2.25	1.75	7.50	3
T_7	Diatomaceous earth	1.00	1.00	1.00	1.00	4.00	1
T_8	*Beauveria bassiana*	2.00	3.50	3.25	2.75	11.50	6
T_9	*Bacillus thuringiensis*	1.25	6.00	3.25	2.75	13.25	7
T_{10}	Untreated (control)	3.00	7.00	5.00	5.00	20.00	10

Appendix 5: Weighted combined ranking of treatments based on seed quality parameters and insecticidal efficacy

Treatment	**Common name**	**Seed quality parameters- (Total score: A)**	**Insecticidal efficacy (Total score: B)**	**Grand total (A + B)**	**Rank**
T_1	Neem leaf powder	35.75	10.75	46.50	7
T_2	Neem seed kernel powder	20.25	4.50	24.75	2
T_3	Sweet flag rhizome powder	33.75	10.25	44.00	6
T_4	Manja koova rhizome powder	43.50	16.00	59.50	9
T_5	Panal leaf powder	24.00	14.25	38.25	5
T_6	Spinosad	20.25	7.50	27.75	3
T_7	Diatomaceous earth	20.75	4.00	24.75	2
T_8	*Beauveria bassiana*	12.25	11.50	23.75	1
T_9	*Bacillus thuringiensis*	20.00	13.25	33.25	4
T_{10}	Untreated (control)	33.00	20.00	53.00	8

IMPACT OF SEED PROTECTANTS ON SEED LONGEVITY AND STORAGE PESTS IN RICE (*Oryza sativa* L.)

By

HARSHAPRADA K

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ABSTRACT OF THE THESIS

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COLLEGE OF HORTICULTURE,

VELLANIKKARA, THRISSUR - 680 656

KERALA, INDIA

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ABSTRACT

In coastal states like Kerala, owing to sub-tropical conditions, maintenance of the quality of the seed during storage has always been a challenge, mostly because the conditions are highly conducive for the growth and proliferation of storage pests. Use of insecticides for management of the stored grains is most common, but, the chemical means of pest management are not advisable in rice as the seeds are often used as food or feed at the end of the season or on losing viability. Considering the above, a study to assess the impact of seed protectants on seed quality, longevity and their efficacy against the storage pests in rice was conducted in the College of Horticulture, Vellanikkara, Thrissur, during 2019 - 2020.

The experiment was conducted with red kernelled rice variety following a completely randomized design with three replications and 10 treatments (T_1 to T_{10}). Before treating the seeds, the seed lot was analysed for the seed quality parameters and it was proved to be of good quality and fit for storage studies. Seed of rice variety Jyothi was treated separately with seed protectants *viz.*, Neem leaf powder @ 10 g/kg (T_1), Neem seed kernel powder @ 10 g/kg (T_2), Sweet flag rhizome powder @ 10 g/kg (T_3), Manja koova rhizome powder @10 g/kg, Panal leaf powder @ 10 g/kg, Spinosad @ 10 ppm, Diatomaceous earth (T_7), *Beauveria bassiana* @ 1×10^8 spores.ml^{-1} (T_8), *Bacillus thuringiensis* @ 1×10^8 spores.ml^{-1} (T_9). Untreated seeds (T_{10}) served as control. Both treated and untreated seeds were dried to ≤12 per cent moisture content and packed in jute bags. The seed quality parameters like germination, seedling vigour indices, seed moisture content and insect infestation (number of beetles and larvae, number of infested seeds, weight of damaged and undamaged seeds) were recorded at the start and subsequently, at monthly intervals for a period of seven months. Seed microflora was recorded at the start and end of the storage.

Following treatment with seed protectants, variation in the quality parameters of seed observed, even before the onset of the storage period. In a few instances, the parameters remained unchanged after treatment, while in most treatments, a marginal increase in seed quality was observed. Seeds treated with *B. bassiana* @ 1×10^8

spores.ml^{-1} (T_8) had registered the highest germination (%), seed vigour indices (VI-I and VI-II), length of shoot and root and seedling dry weight, while seed infestation was the least in diatomaceous earth and neem seed kernel powder. The weight of damaged seed, the number of infested seed, beetles in 100 g of seed sample were the least in these treatments. However, a decline in quality was observed in untreated seeds (T_{10}).

Results revealed the existence of significant difference in the impact of various seed protectants on seed quality and insect infestation related parameters during the storage period. In both treated and untreated seeds, irrespective of the treatment, germination, seedling growth (dry weight, length of root and shoot), vigour index- I (VI-I), vigour index- II (VI-II), weight of undamaged seeds, decreased significantly over the storage period. However, the seed moisture content, seed microflora and parameters like weight of insect damaged seeds, number of beetles, larva and number of infested seeds, in 100 g of seed samples, increased with increase in storage period. In both treated and untreated seeds, irrespective of the treatment, germination and vigour index- I (VI-I) and vigour index- II (VI-II), decreased significantly and progressively over the storage period. However, there was a significant increase in seed moisture content and seed microflora, with the increase in storage period.

Seed treatment with *B. bassiana* resulted in significant enhancement in germination and seed vigour indices (VI-I and VI-II). The treatment also helped to extend seed longevity by one month (S_6) in comparison with seeds treated with seed protectants such as neem seed kernel powder, manja koova rhizome powder, panal leaf powder and *B. thuringiensis,* as well as untreated seeds. However, in comparison to seeds treated with *B. bassiana,* treatment with neem leaf powder, sweet flag rhizome powder, Spinosad and diatomaceous earth, reduced seed longevity by two months. Considering the date of harvest, the longevity of *B. bassiana* treated seed was to 81/2 months, while that of untreated seeds was 71/2 months. In addition, considering the impact on seed longevity and seed quality during storage, treatment with neem leaf powder (T_1), sweet flag rhizome powder (T_3) and spinosad (T_4) and diatomaceous earth (T_7) was found disadvantageous

The efficacy of seed protectants against storage pests was also evaluated at monthly intervals. A significant surge in the insect infestation was observed during the storage period in both treated and untreated control. The estimates of weight and number of insect damaged seeds and number of beetles were the least and significantly low in the treated seeds. Untreated seeds registered the highest insect infestation. Hence, treating seeds with seed protectants was found advantageous over storing them untreated.

Among the seed protectants used, treating seeds with diatomaceous earth (T_7), neem seed kernel powder (T_2) and spinosad (T_6) conferred relatively higher protection, although no significant differences were observed among the treated seeds during the storage period. However, in spite of its insecticidal efficacy, seed treatment with diatomaceous earth and spinosad had a negative impact on seed longevity and quality. The longevity of neem seed kernel powder treated seeds was one month more than that observed in the diatomaceous earth and spinosad treated seeds.

Although, the insecticidal effect was low, treatment with *B. bassaina* was considered the best among the treatments as it had extended the seed longevity the maximum *i.e.,* by a period of one to two month over all other treatments as well as found significantly superior to all other treatments in maintaining seed quality during storage. Seed treatment with neem seed kernel powder was the next best alternative to *B. bassiana.* These treatments had exhibited considerable high efficiency in control of insect population and associated seed damage.

In considerations of the above, the use of entomopathogenic fungi *Beaveria bassiana* @ 1×10^8 spores.ml^{-1} and the botanical neem seed kernel powder @ 10 g/kg, were found as the best treatments in reducing the rate of seed deterioration during storage in rice. Of the two, treatment with neem seed kernel powder @ 10 g/kg accorded better control of stored pest infestation in rice seeds.