

## PERIODIC EFFECT OF COWPEA AND MUNG BEAN PELLETTED SEEDS WITH *AVICENNIA MARINA* (FORSSK.) VIERH PARTS POWDER AND THEIR CONTRIBUTION IN THE CONTROL OF ROOT KNOT NEMATODE

MARIUM TARIQ AND SHAHNAZ DAWAR\*

Department of Botany, University of Karachi, Karachi -75270, Pakistan.

\*Corresponding author e-mail: shahnaz\_dawar@yahoo.com

### Abstract

Cowpea and mung bean seeds pelleted with *Avicennia marina* (Forssk.) Vierh parts powder including leaves, stem, pneumatophore were investigated at different intervals of 0 and after 30, 60, 90, 180 and 360 days stored under 4 and 30°C. Both crops showed improved germination, plant length and plant weight at all storage period upto 360 days but maximum result was accounted upto 90 days interval. Temperature of 4°C was suitable for storage of pelleted seeds and caused reduction in root knot infection by reducing the number of galls and egg masses per root system in both crops upto 360 days. Leaves and stem powder used as pelletizing material gave better results followed by pneumatophore powder of *A. marina*. Pyrex (pyrophyllite) had a least effect on germination and growth parameters in contrast to other treatments.

### Introduction

The process of enclosing a seed with some material around and convert the seed from oddly shaped and light density to uniform, heavier and round as possible is regarded as seed pelleting. The materials surrounding the seed provides nutrition to young seedling and helps to maintain natural water holding potential (Krishnasamy, 2003). The advantage of smaller pelleted seeds was minimizing uptake of harmful chemicals from soil, thinning problem, gap filling, improving germination, establishment of seeds and the farmers plant these seeds in accurate and precise way (Bharathi *et al.*, 2003). Researchers used different botanicals like leaf powder of Arappu, Pongamia, Neem and chemicals including ZnSO<sub>4</sub>, Captan, Borax, Imidacloprid, KH<sub>2</sub>PO<sub>4</sub>, KNO<sub>3</sub>, K<sub>2</sub>SO<sub>4</sub> as a filler materials which suppresses soil borne diseases leading to enhancement in growth parameters of crops (Manjunath *et al.*, 2009; Kavitha *et al.*, 2009; Masuthi *et al.*, 2009; Gupta & Aneja, 2000).

Grey mangrove, *Avicennia marina* (Forssk.) Vierh, belongs to the family, Avicenniaceae, is located in Sindh and Balochistan provinces of Pakistan and covers an area of almost 600, 000 hectares (Mirza *et al.*, 1988). *A. marina* is classified as halophytic plant which can tolerate approximately 90 % of salt concentration compared to other mangrove species (Macnae, 1966; Burchett *et al.*, 1984, 1989). The chemicals present in mangroves act as antibiotic, antiviral, antibacterial and antifungal purposes (Miles *et al.*, 1999; Combs & Anderson, 1949; Jamale & Joshi, 1978; Ross *et al.*, 1980).

Several researches were hypothesized that some phenolic substances (toxic substances) might release from these mangrove into soil which suppressed the activities of phytonematodes and other mycoflora and improved the growth of crop plants (Mehdi *et al.*, 1999; Shaukat *et al.*, 2003; Tariq & Dawar, 2010, 2011). Phytonematodes particularly root knot nematodes (*Meloidogyne* spp.) are considered as most destructive pathogens whose primary symptoms is the formation of typical galls on the roots of host plant. Due to formation of galls, various other symptoms were noticed including changes in morphology and physiology of host plants, stunted growth, temporary

wilting, premature shedding of leaves, chlorosis and nutritional deficiencies (Agrios, 2004; Abad *et al.*, 2003; Karsen & Moens, 2006; Oka, 2010).

Information regarding *A. marina* in seed pelleting is lacking. Hence, the present study was designed to investigate the effect of cowpea (*Vigna unguiculata* L. Walp) and mung bean (*Vigna radiata* (L.) Wilczek) seeds pelleted with *A. marina* parts powder on growth parameters and infection of *Meloidogyne javanica* (Treub) Chitwood on roots.

### Materials and Methods

**Plant material:** *A. marina* parts (leaves, stem, pneumatophore) were collected from Sandspit, Karachi, Pakistan, washed with distilled water to remove dirt particles and dried under shade. The plant materials were powdered using an electric grinder and stored in an airtight container for further studies.

**Seed pelleting:** Seeds of cowpea and mung bean were surface sterilized using 1% sodium hypochlorite, thoroughly washed with MgSO<sub>4</sub> (0.1 M) solution and dried aseptically under laminar flow hood. Leaves, stem and pneumatophore powder of *A. marina* was separately mixed with pyrex (pyrophyllite) and pelleted the cowpea and mung bean seeds using sterilized gum Arabic solution (2%). To assess the effect of pyrex, seeds were pelleted with only pyrex mixed with sterilized distilled water. These pelleted seeds were air dried under laminar flow hood. The seeds were placed in air tight container and stored under 4°C (refrigerator) and 30 °C (incubator) for 360 days. At 0 day and after 30, 60, 90, 180 and 360 days interval growth parameters and root knot nematode were observed.

**Preparation of root knot nematode inoculums:** Infected roots of egg plant (*Solanum melongena* L.) with *M. javanica* (Treub) Chitwood were collected from University of Karachi garden and the root knot species were identified using perennial pattern (Taylor & Netscher, 1974). To extract eggs of *M. javanica* McClure *et al.*, (1973) technique using 1% Sodium hypochlorite solution was followed. For obtaining freshly hatched

juveniles  $J_2$ , suspension of eggs were poured on a cotton-wool filter and incubated at  $26 \pm 2^\circ\text{C}$ . Juveniles were collected within 72 h and used as inoculum for pot experiment.

**Experimental setup:** At 0 day of seed pelleting, two sets of pots were maintained for assessment of temperatures effect on cowpea and mung bean. Pots were filled with 300 g sandyloam soil (74% sand, 16% silt, 10% clay) having pH 8.1 and 40% moisture holding capacity (MHC) (Keen & Raczkowski, 1922). The treatments of pelleted seeds including pyrex only, leaves + pyrex, stem + pyrex, pneumatophore + pyrex and control (without pelleted seed) were sown in pots (5 seeds per pot). Concept of seed pelleting with combination of *A. marina* parts and pyrex was previously described by Tariq & Dawar (2011) in which these combination was used in preparation of pellets in controlling of root rot fungi. Each treatment was replicated three times and set in a complete randomized design on screen house bench. Ten days after germination of seeds, approximately 2000 freshly hatched *M. javanica* juveniles ( $J_2$ ) were introduced into holes made around roots of each seedling. After 50 days of inoculation of  $J_2$ , data were collected for germination, plant length (cm), plant weight (g), number of galls per root system and number of egg masses per root system. The above procedure with two sets of treatments was repeated at 30, 60, 90, 180 and 360 days of interval.

**Data analysis:** The entire data were analyzed using three-way ANOVA. The means were separated by using Fisher's least significant difference (LSD) at  $p < 0.05$ . For the homogeneity of variance, percentages in germination were arcsine transformed (Sokal & Rohlf, 1995).

## Results

**Effect on germination:** Seed pelleting with *A. marina* leaves, stem and pneumatophore powder on cowpea and mung bean seeds showed a great influence on germination when stored for 0 and after 30, 60, 90, 180 and 360 days at 4 and  $30^\circ\text{C}$ . Maximum germination was recorded when leaves pelleted seeds of cowpea and mung bean were stored for 60 days interval at 4 and  $30^\circ\text{C}$ . After 90 days interval, stem pelleted cowpea seeds showed increased germination upto 360 days while in mung bean, leaves pelleted seeds gave maximum germination after 360 days interval at storage of  $4^\circ\text{C}$ . Germination of both cowpea and mung bean seeds showed significant ( $P < 0.001$ ) increase after 30, 60 and 90 days interval at storage of  $4^\circ\text{C}$  which after 180 (76.9 and 81.1 respectively) and 360 days (54.9 and 72.2 respectively) start decreasing (Table 1).

**Effect on plant length:** A gradual increment in plant length of pelleted seeds were attained after 30 and 60 days interval (31.3 and 35.3 cm respectively) at  $4^\circ\text{C}$  which start decreasing after 90, 180 and 360 days (34.9, 29.4 and 25.8 cm respectively) at  $4^\circ\text{C}$  ( $p < 0.001$ ). In mung bean, maximum plant length was observed after 90 days (33.3 cm) interval which start decreasing after 180 and 360 days intervals (27.6 and 21.8 cm respectively) at  $4^\circ\text{C}$

( $p < 0.001$ ). Leaves pelleted seeds in cowpea and leaves, stem pelleted seeds in mung bean showed significant result ( $p < 0.001$ ) in enhancing plant length as compared to other treatments (Table 2).

**Effect on plant weight:** Pelletized mung bean and cowpea seeds with all treatments gave better result of plant weight after 30 and 60 days interval. However, maximum enhancement in weight was recorded after 90 days interval in mung bean and cowpea when pelleted with leaves and stem powder of *A. marina* (3.80 and 3.16 g respectively) at  $4^\circ\text{C}$  ( $p < 0.001$ ) (Table 3).

**Effect on root knot infection:** It was interesting to note that mung bean seeds pelleted with leaves powder of *A. marina* reduced the galls formation after 360 days interval when stored at 4 and  $30^\circ\text{C}$ . However, maximum reduction was attained after 180 days interval when mung bean seeds were pelleted with leaves powder of *A. marina* (20/root system) at 4 and  $30^\circ\text{C}$  compared to control (198, 211/root system respectively) ( $p < 0.001$ ). All treatments were also significantly efficient ( $p < 0.001$ ) in reducing gall formation on cowpea plants. Highest reduction in gall per root system were recorded when stem powder was pelleted on cowpea seeds (21/ root system) after 90 days interval followed by leaves pelleted seeds (31/root system) at  $4^\circ\text{C}$ . Pyrex pelleted seeds was not effective in controlling gall formation on both crops at 4 and  $30^\circ\text{C}$  (Table 4).

Maximum reduction in egg masses per root system were attained after 90 days interval when cowpea seeds pelleted with stem powder stored at  $4^\circ\text{C}$  followed by leaves powder ( $p < 0.001$ ). Mung bean seeds pelleted with leaves powder showed maximum reduction of egg masses per root system after 180 days interval at  $4^\circ\text{C}$ . Pneumatophore pelleted seeds also showed reduction in egg masses per root system but its effect was comparatively less than leaves and stem powder. However, only pyrex pelleted seeds had a little effect on reduction on egg masses compared to control. Temperature of  $4^\circ\text{C}$  gave more pronounced result in contrast to  $30^\circ\text{C}$  on both crops ( $p < 0.001, 0.01$ ) (Table 5).

## Discussion

Present results demonstrated that cowpea and mung bean seeds pelleted with all parts of *A. marina* showed germination upto 360 days interval when stored at 30 and  $4^\circ\text{C}$  temperatures. However, 90 days storage interval gave maximum plant length and weight in seeds pelletized with leaves and stem powder compared to non palletized seeds which showed decline in growth parameters after 60 days interval. It was thought that a pelletized seed was heavier in weight and easily comes in contact with soil and process of imbibition occurs. Our results can be compared with other studies on seeds of tomato and cowpea when pelletized with leaves powder of Neem or arappu were observed to be better in maintaining seed viability and vigour stored for ten months (Maraddi, 2002; Nargis & Thiagarajan, 1991). In addition, highest germination throughout the storage period and improved quality were recorded when soybean seeds were treated with 2 g bavistin (Sullivan, 1979).

Table 1. Effect of seed pelleting with *A. marina* parts powder on germination at 0 and after 30, 60, 90, 180 and 360 days of storage under 4 and 30°C.

Treatments	Days																							
	0				30				60				90				180				360			
	Temperatures (°C)																							
	Cowpea																							
Control	43.0 ± 3.84	46.9 ± 3.84	51.1 ± 6.98	60.0 ± 15.36	55.3 ± 8.03	43.0 ± 3.84	38.8 ± 6.98	35.0 ± 4.22	38.8 ± 6.98	35.0 ± 4.22	35.0 ± 4.22	35.0 ± 3.84	30.7 ± 4.22	30.7 ± 4.22	30.7 ± 4.22	30.7 ± 6.98	21.9 ± 11.5							
Pyrex	42.7 ± 3.84	54.9 ± 4.22	43.0 ± 3.84	56.1 ± 16.92	51.1 ± 6.92	54.9 ± 4.22	43.0 ± 3.84	30.7 ± 4.22	43.0 ± 3.84	30.7 ± 4.22	30.7 ± 4.22	30.7 ± 6.98	43.0 ± 3.84	30.7 ± 4.22	30.7 ± 4.22	30.7 ± 6.98	21.9 ± 11.5							
Leaves	59.2 ± 1.88	90.0 ± 0.00	76.9 ± 13.07	90.0 ± 0.00	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	43.0 ± 3.84							
Stem	54.9 ± 4.22	72.2 ± 8.85	68.0 ± 11.55	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	51.1 ± 6.98							
Pneumatophore	51.1 ± 6.98	43.0 ± 3.84	51.1 ± 6.98	76.9 ± 13.07	72.2 ± 8.85	90.0 ± 0.00	81.1 ± 8.85	47.2 ± 8.06	81.1 ± 8.85	47.2 ± 8.06	43.0 ± 4.22	43.0 ± 4.22	38.8 ± 6.98	43.0 ± 4.22	43.0 ± 4.22	43.0 ± 4.22	38.8 ± 6.98							
LSD <sub>0.05</sub>	Days = 6.73 ***, Treatment = 6.14 ***, Temperature = 3.88 <sup>NS</sup>																							
	Mung bean																							
Control	43.0 ± 3.84	63.8 ± 13.0	54.9 ± 4.22	68.0 ± 11.5	59.2 ± 4.22	60.0 ± 15.3	51.1 ± 6.98	38.8 ± 6.98	46.9 ± 3.84	38.8 ± 6.98	43.0 ± 1.27	34.6 ± 8.06	30.7 ± 4.22	30.7 ± 4.22	30.7 ± 4.22	30.7 ± 4.22	30.7 ± 4.22							
Pyrex	46.9 ± 3.84	51.1 ± 6.98	46.9 ± 3.84	63.8 ± 13.0	68.0 ± 11.5	63.8 ± 13.0	54.9 ± 4.22	35.0 ± 4.22	35.0 ± 4.22	35.0 ± 4.22	35.0 ± 4.22	35.0 ± 4.22	35.0 ± 4.22	35.0 ± 4.22	35.0 ± 4.22	35.0 ± 4.22	35.0 ± 4.22							
Leaves	72.2 ± 4.22	90.0 ± 0.00	81.1 ± 8.85	90.0 ± 0.00	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	63.8 ± 13.07							
Stem	54.9 ± 13.0	81.1 ± 8.85	72.2 ± 8.85	81.1 ± 8.85	81.1 ± 8.85	90.0 ± 0.00	81.1 ± 8.85	72.2 ± 8.85	81.1 ± 8.85	72.2 ± 8.85	72.2 ± 8.85	72.2 ± 8.85	72.2 ± 8.85	72.2 ± 8.85	72.2 ± 8.85	72.2 ± 8.85	59.2 ± 4.22							
Pneumatophore	63.8 ± 4.08	66.4 ± 8.85	68.0 ± 11.5	81.1 ± 8.85	72.2 ± 8.85	72.2 ± 8.85	72.2 ± 8.85	54.9 ± 4.22	51.1 ± 6.98	54.9 ± 4.22	51.1 ± 6.98	54.9 ± 1.35	47.2 ± 8.06	47.2 ± 8.06	47.2 ± 8.06	47.2 ± 8.06	47.2 ± 8.06							
LSD <sub>0.05</sub>	Days = 7.43 ***, Treatment = 6.78 ***, Temperature = 4.29 **																							

Values shown are means of three replicates ± SE. \*\* p&lt;0.01, \*\*\* p&lt;0.001, NS= Non-significant

Table 2. Effect of seed pelleting with *A. marina* parts powder on plant length (cm) at 0 and after 30, 60, 90, 180 and 360 days of storage under 4 and 30 °C.

Treatments	Days																							
	0				30				60				90				180				360			
	Temperatures (°C)																							
	Cowpea																							
Control	22.4 ± 1.12	24.0 ± 0.86	21.5 ± 1.21	21.2 ± 1.59	25.0 ± 1.16	21.9 ± 2.19	22.0 ± 1.41	19.1 ± 1.36	18.7 ± 0.68	18.8 ± 1.86	18.3 ± 1.29	18.8 ± 1.86	18.7 ± 0.68	18.8 ± 1.86	18.7 ± 0.68	18.8 ± 1.86	18.3 ± 1.29							
Pyrex	23.0 ± 0.82	23.9 ± 0.34	22.2 ± 1.41	22.6 ± 1.90	19.7 ± 1.55	25.2 ± 1.30	23.0 ± 1.76	19.2 ± 1.60	19.4 ± 1.05	19.4 ± 0.52	13.0 ± 6.55	19.4 ± 0.52	19.4 ± 1.05	19.4 ± 0.52	19.4 ± 1.05	19.4 ± 0.52	13.0 ± 6.55							
Leaves	26.7 ± 0.73	31.3 ± 1.55	30.9 ± 0.99	35.3 ± 1.58	33.8 ± 1.86	34.9 ± 2.17	32.9 ± 1.41	29.4 ± 1.09	27.9 ± 1.72	25.8 ± 1.27	25.2 ± 0.96	29.4 ± 1.09	27.9 ± 1.72	25.8 ± 1.27	25.8 ± 1.27	25.8 ± 1.27	25.2 ± 0.96							
Stem	25.0 ± 1.85	29.8 ± 0.75	29.5 ± 1.02	32.1 ± 1.14	30.3 ± 1.61	33.6 ± 1.64	31.3 ± 2.07	28.3 ± 1.51	26.8 ± 1.48	24.0 ± 1.20	23.0 ± 0.98	28.3 ± 1.51	26.8 ± 1.48	24.0 ± 1.20	24.0 ± 1.20	24.0 ± 1.20	23.0 ± 0.98							
Pneumatophore	24.2 ± 0.89	29.9 ± 0.88	26.9 ± 1.44	31.4 ± 1.48	29.0 ± 1.74	30.4 ± 0.80	27.7 ± 0.47	27.2 ± 1.45	26.3 ± 1.63	20.6 ± 1.41	19.7 ± 0.58	27.2 ± 1.45	26.3 ± 1.63	20.6 ± 1.41	20.6 ± 1.41	20.6 ± 1.41	19.7 ± 0.58							
LSD <sub>0.05</sub>	Days = 1.41 ***, Treatment = 1.28 ***, Temperature = 0.18 **																							
	Mung bean																							
Control	17.9 ± 0.82	20.3 ± 1.09	19.7 ± 0.89	23.9 ± 1.14	22.9 ± 1.34	23.2 ± 1.56	22.8 ± 0.84	18.6 ± 0.90	17.8 ± 0.82	15.7 ± 0.26	15.0 ± 0.92	18.6 ± 0.90	17.8 ± 0.82	15.7 ± 0.26	15.7 ± 0.26	15.7 ± 0.26	15.0 ± 0.92							
Pyrex	18.5 ± 1.18	20.4 ± 1.02	18.1 ± 0.92	21.8 ± 0.82	21.5 ± 1.12	18.1 ± 0.87	19.5 ± 0.43	19.8 ± 1.00	19.6 ± 0.46	13.4 ± 0.35	13.2 ± 0.76	19.8 ± 1.00	19.6 ± 0.46	13.4 ± 0.35	13.4 ± 0.35	13.4 ± 0.35	13.2 ± 0.76							
Leaves	24.9 ± 1.52	30.1 ± 0.92	27.4 ± 1.21	29.8 ± 0.83	28.6 ± 1.03	29.0 ± 1.78	29.5 ± 1.01	26.8 ± 0.79	25.1 ± 1.77	21.3 ± 0.50	20.7 ± 0.78	26.8 ± 0.79	25.1 ± 1.77	21.3 ± 0.50	21.3 ± 0.50	21.3 ± 0.50	20.7 ± 0.78							
Stem	23.9 ± 0.93	27.0 ± 1.30	26.4 ± 1.56	29.3 ± 0.57	28.2 ± 0.56	33.3 ± 0.87	29.1 ± 0.73	27.6 ± 0.93	26.4 ± 1.12	21.8 ± 0.43	22.0 ± 1.31	27.6 ± 0.93	26.4 ± 1.12	21.8 ± 0.43	21.8 ± 0.43	21.8 ± 0.43	22.0 ± 1.31							
Pneumatophore	23.3 ± 0.81	26.1 ± 0.92	24.5 ± 0.50	27.8 ± 0.79	26.0 ± 0.89	28.9 ± 0.89	28.0 ± 0.80	22.2 ± 1.31	24.6 ± 0.90	19.2 ± 0.18	17.7 ± 1.51	28.0 ± 0.80	22.2 ± 1.31	19.2 ± 0.18	19.2 ± 0.18	19.2 ± 0.18	17.7 ± 1.51							
LSD <sub>0.05</sub>	Days = 0.94 ***, Treatment = 0.86 ***, Temperature = 0.54*																							

Values shown are means of three replicates ± SE. \* p&lt;0.05, \*\* p&lt;0.01, \*\*\* p&lt;0.001

Table 3. Effect of seed pelleting with *A. marina* parts powder on plant length (g) at 0 and after 30, 60, 90, 180 and 360 days of storage under 4 and 30°C.

Treatments	Days																																																						
	30					60					90					180					360																																		
	Temperatures (°C)					Temperatures (°C)					Temperatures (°C)					Temperatures (°C)					Temperatures (°C)																																		
	<b>Cowpea</b>																																																						
Control	1.83 ± 0.35	2.13 ± 0.33	1.95 ± 0.44	2.04 ± 0.11	1.99 ± 0.13	1.98 ± 0.24	1.99 ± 0.30	1.96 ± 0.36	1.92 ± 0.41	1.67 ± 0.16	1.48 ± 0.30	1.80 ± 0.32	2.13 ± 0.30	1.91 ± 0.29	2.05 ± 0.19	2.01 ± 0.10	2.07 ± 0.17	1.94 ± 0.19	2.01 ± 0.28	1.89 ± 0.31	1.64 ± 0.22	1.25 ± 0.69	2.33 ± 0.20	2.91 ± 0.25	2.88 ± 0.33	3.13 ± 0.19	3.14 ± 0.24	3.16 ± 0.29	3.11 ± 0.22	2.54 ± 0.46	2.42 ± 0.40	1.91 ± 0.25	2.22 ± 0.49	2.05 ± 0.19	2.65 ± 0.44	2.68 ± 0.50	2.87 ± 0.36	2.80 ± 0.30	2.63 ± 0.39	2.53 ± 0.50	2.24 ± 0.32	1.91 ± 0.27	1.65 ± 0.32	1.96 ± 0.23	2.06 ± 1.85	1.94 ± 0.25	2.10 ± 0.20	2.16 ± 0.13	2.04 ± 0.23	2.03 ± 0.25	2.10 ± 0.25	1.70 ± 0.27	1.55 ± 0.24		
LSD <sub>0.05</sub>	Days = 0.27 ***, Treatment = 0.25 ***, Temperature = 0.15 <sup>ns</sup>																																																						
	<b>Mung bean</b>																																																						
Control	1.37 ± 0.41	2.04 ± 0.18	1.95 ± 0.13	2.47 ± 0.32	2.14 ± 0.20	2.32 ± 0.37	2.28 ± 0.40	1.49 ± 0.38	1.36 ± 0.43	1.52 ± 3.71	1.33 ± 0.32	1.63 ± 0.10	1.99 ± 0.11	1.83 ± 0.12	2.29 ± 0.23	2.20 ± 0.31	2.45 ± 0.23	2.29 ± 0.21	1.19 ± 0.32	1.13 ± 0.34	1.12 ± 7.81	0.98 ± 0.35	2.48 ± 0.50	2.99 ± 0.12	2.96 ± 0.18	3.19 ± 0.25	3.19 ± 0.19	3.12 ± 0.18	3.00 ± 0.18	2.93 ± 0.31	2.82 ± 0.33	2.53 ± 2.02	2.06 ± 0.43	2.40 ± 0.67	2.60 ± 0.57	2.58 ± 0.59	3.11 ± 0.23	3.06 ± 0.24	3.80 ± 0.11	3.14 ± 0.17	2.96 ± 0.42	2.90 ± 0.38	2.17 ± 2.40	2.08 ± 0.44	2.26 ± 0.22	2.32 ± 0.30	1.96 ± 0.56	3.14 ± 0.19	3.07 ± 0.21	2.92 ± 0.23	2.84 ± 0.25	2.06 ± 0.39	1.98 ± 0.22	1.98 ± 3.75	1.92 ± 0.17
LSD <sub>0.05</sub>	Days = 0.30 ***, Treatment = 0.27 ***, Temperature = 0.17 <sup>ns</sup>																																																						

Values shown are means of three replicates ± SE. \*\*\* p&lt;0.001, NS = Non-significant

Table 4. Effect of seed pelleting with *A. marina* parts powder on number of galls/root system at 0 and after 30, 60, 90, 180 and 360 days of storage under 4 and 30°C.

Treatments	Days																																																					
	30					60					90					180					360																																	
	Temperatures (°C)					Temperatures (°C)					Temperatures (°C)					Temperatures (°C)					Temperatures (°C)																																	
	<b>Cowpea</b>																																																					
Control	68 ± 4.91	70 ± 2.08	77 ± 2.51	76 ± 2.08	87 ± 3.21	87 ± 2.08	88 ± 3.21	91 ± 3.84	99 ± 5.85	95 ± 2.64	100 ± 3.60	65 ± 3.60	58 ± 2.33	59 ± 1.15	69 ± 3.52	74 ± 2.90	83 ± 2.02	86 ± 2.30	87 ± 2.40	94 ± 3.52	99 ± 3.28	77 ± 39.2	53 ± 3.75	31 ± 1.73	40 ± 1.73	38 ± 3.21	51 ± 3.17	31 ± 1.20	40 ± 3.48	36 ± 3.21	38 ± 2.60	63 ± 3.48	66 ± 3.60	64 ± 2.60	45 ± 2.90	48 ± 2.64	27 ± 1.52	33 ± 2.30	30 ± 1.52	28 ± 2.60	30 ± 2.02	49 ± 2.08	66 ± 4.25	65 ± 2.08	50 ± 1.73	53 ± 7.68	28 ± 1.73	30 ± 2.30	40 ± 3.21	42 ± 3.75	38 ± 3.28	52 ± 4.72	64 ± 2.64	77 ± 3.21
LSD <sub>0.05</sub>	Days = 5.21 ***, Treatment = 4.75 ***, Temperature = 3.00 **																																																					
	<b>Mung bean</b>																																																					
Control	180 ± 19.5	159 ± 12.9	182 ± 10.0	162 ± 5.60	179 ± 10.8	169 ± 2.60	187 ± 15.8	198 ± 13.6	211 ± 16.1	215 ± 5.20	242 ± 6.00	163 ± 6.64	135 ± 5.23	157 ± 7.76	200 ± 10.2	123 ± 7.51	160 ± 8.71	168 ± 7.09	179 ± 8.51	196 ± 6.35	207 ± 5.68	66 ± 3.84	41 ± 2.64	47 ± 2.30	21 ± 2.33	28 ± 1.45	24 ± 1.15	24 ± 2.64	20 ± 2.02	20 ± 2.40	27 ± 1.76	28 ± 2.30	83 ± 3.75	50 ± 4.35	56 ± 3.05	28 ± 2.18	34 ± 3.17	25 ± 2.88	30 ± 2.72	25 ± 3.21	28 ± 3.21	25 ± 2.40	30 ± 1.76	99 ± 5.45	61 ± 6.38	63 ± 6.64	48 ± 2.40	59 ± 2.60	44 ± 3.17	50 ± 2.33	36 ± 3.46	41 ± 1.45	33 ± 2.88	36 ± 4.50
LSD <sub>0.05</sub>	Days = 6.42 ***, Treatment = 5.86 ***, Temperature = 3.70 **																																																					

Values shown are means of three replicates ± SE. \*\* p&lt;0.01, \*\*\* p&lt;0.001

Table 5. Effect of seed pelleting with *A. marina* parts powder on number of egg masses/root system at 0 and after 30, 60, 90, 180 and 360 days of storage under 4 and 30 °C.

Treatments	Days											
	0		30		60		90		180		360	
	4	30	4	30	4	30	4	30	4	30	4	30
	Temperatures (°C)											
	Cowpea						Mung bean					
Control	56 ± 4.91	59 ± 2.40	68 ± 2.96	63 ± 3.17	75 ± 3.28	73 ± 3.46	75 ± 3.52	77 ± 2.60	85 ± 3.21	84 ± 2.64	88 ± 2.96	
Pyrex	54 ± 4.25	45 ± 3.21	48 ± 1.85	55 ± 3.38	59 ± 1.52	70 ± 2.02	71 ± 1.73	73 ± 3.17	79 ± 4.04	86 ± 3.17	65 ± 33.3	
Leaves	39 ± 4.05	18 ± 1.20	27 ± 2.02	25 ± 2.88	37 ± 2.60	18 ± 1.45	25 ± 3.60	22 ± 3.78	25 ± 2.72	51 ± 3.92	54 ± 4.25	
Stem	52 ± 2.40	32 ± 2.40	36 ± 3.17	14 ± 1.76	18 ± 1.76	10 ± 1.45	17 ± 1.45	15 ± 2.33	18 ± 2.64	36 ± 2.33	51 ± 3.84	
Pneumatophore	52 ± 0.88	37 ± 1.85	39 ± 2.08	15 ± 2.02	19 ± 1.76	27 ± 2.08	28 ± 4.09	26 ± 3.28	39 ± 4.63	51 ± 3.46	64 ± 3.05	
LSD <sub>0.05</sub>	Days = 4.64 ***; Treatment = 4.24 ***; Temperature = 2.68 **											
Control	168 ± 17.9	149 ± 10.9	172 ± 11.0	154 ± 5.78	166 ± 10.3	158 ± 2.88	174 ± 14.4	189 ± 12.4	205 ± 17.3	199 ± 5.20	231 ± 6.35	
Pyrex	156 ± 6.74	127 ± 4.72	149 ± 7.81	152 ± 9.83	186 ± 11.0	110 ± 6.88	146 ± 8.71	160 ± 6.74	165 ± 7.63	184 ± 6.35	196 ± 6.56	
Leaves	54 ± 3.05	32 ± 2.08	39 ± 2.60	15 ± 1.45	20 ± 1.76	18 ± 1.45	17 ± 2.51	14 ± 2.08	15 ± 2.60	20 ± 1.76	23 ± 2.72	
Stem	75 ± 3.71	42 ± 3.52	47 ± 3.05	15 ± 2.18	25 ± 3.17	17 ± 2.60	24 ± 3.21	19 ± 3.17	23 ± 3.28	19 ± 2.40	25 ± 1.52	
Pneumatophore	91 ± 3.51	53 ± 5.78	56 ± 7.81	39 ± 2.72	52 ± 2.60	37 ± 3.75	44 ± 3.05	29 ± 4.61	35 ± 1.85	26 ± 2.88	30 ± 4.50	
LSD <sub>0.05</sub>	Days = 6.10 ***; Treatment = 5.57 ***; Temperature = 3.52 ***											

Values shown are means of three replicates ± SE. \*\*\* p<0.001; \*\* p<0.01

In the present study, seeds of cowpea and mung bean pelleted with leaves and stem powder showed good germination and viability when stored at low temperature (4°C). It was assumed that low temperature decreased the activities of pathogen and metabolism which led to lesser deterioration of seeds. Roberts (1972) observed that pathogens were suppressed in storage condition due to protective layer of insecticide and fungicides which results in enhancement of seed vigor and viability. The chemicals or botanicals coated around seeds act as a protective layer which might protect the seeds been eaten up by birds and animals.

Present study demonstrated that all the parts of *A. marina* showed satisfactory result but leaves and stem observed to be more active in enhancement of growth and reduction of gall formation by *M. javanica*. The leaves powder of *A. marina* which pelleted mung bean and cowpea seeds contains chlorophyll molecules which come in contact with amino acid and humic acid present in soil rhizosphere helps in growth and development of seeds which results in higher seed yield and quality (Balaji, 1990). The root knot nematodes distributed worldwide attack on various crops. They damaged roots of crop plant and produced giant cells due to which reduction in supply of water and nutrition from soil occurs and finally lead to the death of plant. Due to treatment applied, the plant cell wall becomes harder physically caused inhibition in nematodes infection (Oka *et al.*, 1999). Different bioactive compounds present in mangrove plants which contains steroids, triterpenes, saponins, flavonoids, alkaloids and tannins are responsible for antibiotic, antiviral, antibacterial and antifungal activities (Bandaranayake, 1998).

Our result clearly summarized that *A. marina* parts like leaves, stem and pneumatophore used as seed pelleting agents has a potential to suppress root knot nematode population. Furthermore, growth and viability of seeds also increased over a long period of time when stored at cold temperature (4°C).

## Acknowledgements

Authors thanks to Higher Education Commission (HEC) for providing financial support through funding the research program.

## References

- Abad, P., B. Favery, M. Rosso and P. Castagnone-Sereno. 2003. Root knot nematode parasitism and host response: molecular basis of a sophisticated interaction. *Molecular and Plant Pathology*, 4: 217-224.
- Agrios, G.N. 2004. *Plant Pathology*. 4<sup>th</sup> ed. Academic Press, San Diego, California, USA, p. 635.
- Balaji, D.S. 1990. *Studies on the seed and soil relationship to certain crops; paddy, greengram, soybean, redgram, sunflower, groundnut and cotton*. MSc Thesis. Tamil Nadu Agric Univ, Coimbatore, India.
- Bandaranayake, W.M. 1998. Traditional and medicinal uses of mangroves. *Mangroves and salt marshes*. 2: 133-148.
- Bharathi, A., P. Nateshan, K. Vanangamudi, P. S. Sherin, M. Ramya and P. Thangavelu. 2003. *Conceptual and utility differences among seed enhancement technologies viz., seed pelleting, seed coating and seed coloring*. ICAR short course on seed hardening and pelleting technologies for rainfed/garden land ecosystems. Tamil Nadu Agricultural University, Coimbatore, p. 131.

- Burchett, M.D., C.D. Field and A. Pulkownik. 1984. Salinity growth and root respiration in the grey mangrove *Avicennia marina*. *Physiology Plantarum*, 60: 113-118.
- Burchett, M.D., C.J. Clarke, C.D. Field and A. Pulkownik. 1989. Growth and respiration in two mangrove species at a range of salinities. *Physiology Plantarum*, 75: 299-303.
- Combs, C.A. and H. Anderson. 1949. Use of mangrove bark. *Australian Leather Trade Rev.*, 43: 270-274.
- Gupta, A. and K.R. Aneja. 2000. Field efficacy of seed dressing chemicals on seedling emergence, seed yield and quality in soybean. *Seed Res.*, 28: 54-58.
- Jamale, B.B. and G.V. Joshi. 1978. Effect on age of mineral constituents, polyphenoloxides in mangrove leaves. *J. Exp. Biol.*, 16: 117-120.
- Karassen, G. and M. Moens. 2006. Root-knot nematodes. In: *Plant Nematology*. (Ed.) R.N. Perry and M. Moens. CABI Publishing, Wallingford, UK, pp. 59-90.
- Kavitha, M., V. K. Deshpande, B.S. Vyakaranahal, J.S. Awakkanavar, Y. Hegde and J.C. Mathad. 2009. Seed pelleting with organic and inorganic inputs for vigour and viability in chilli seeds. *Karnataka J. Agric. Sci.*, 22: 296-300.
- Keen, B.A. and H. Raczkowski. 1922. Clay contents and certain physical properties of soil. *J. Agric. Sci.*, 11: 441-449.
- Krishnasamy, V. 2003. *Seed pelleting-principles and practices*. ICAR short course on seed hardening and pelleting technologies for rainfed/garden land ecosystems. Tamil Nadu Agricultural University, Coimbatore, p. 96.
- Macnae, W. 1966. Mangroves in eastern and southern Australia. *Australian journal of Botany*, 14: 67-104.
- Manjunath, S.N., V.K. Deshpande, O. Sridevi, D.S. Uppar, H.B. Babalad and M.S.L. Rao. 2009. Influence of seed pelleting on crop growth, seed yield and quality of paprika chilli (*Capsicum annuum* L.). *Karnataka J. Agric. Sci.*, 22: 762-764.
- Maraddi, B.M. 2002. Influence of growth retardants on seed yield and quality and seeds treatments on storability of cowpea cv. C.152. MSc (Agi) Thesis, Univ Agric Sci, Dharwad, India.
- Masuthi, D., B.S. Vyakaranahal and V.K. Deshpande. 2009. Influence of pelleting with micronutrients and botanical on growth, seed yield and quality of vegetable cowpea. *Karnataka J. Agric. Sci.*, 22: 898-900.
- McClure, M.A., T.H. Kruk and I. Misaghi. 1973. A method for obtaining quantities of clean *Meloidogyne* eggs. *Journal of Nematology*, 5: 230.
- Mehdi, F.S., I.A. Siddiqui, S. Erum and R. Ali. 1999. Effect of *Avicennia marina* and *Paecilomyces lilacinus* on root rot-knot diseases of tomato. *Pakistan Journal of Biological Sciences*, 2: 1462-1466.
- Miles, D.H., U. Kokpol, V. Chittawong, S. Tip-Pyang, K. Tunsuwan and C. Nguyen. 1999. *Mangrove forest-The importance of conservation as a bioresource for ecosystem diversity and utilization as a source of chemical constituents with potential medicinal and agricultural value*. IUPAC. (<http://www.iupac.org/symposia/proceedings/phuket97/miles.html>).
- Mirza, M.I., M.Z. Hasan, S. Akhtar, J. Ali and M.A. Sanjrani. 1988. Remote sensing survey of mangrove forest along the coast of Balochistan. In: *Marine science of the Arabian sea*. (Ed.) M.F. Thompson and N.M. Tirmizi. AIBS, Washington, DC, pp. 339-348.
- Nargis, S. and C.P. Thiagarajan. 1991. Storage studies with pelleted seeds of tomato cv PKM-1. *South Indian Hort.*, 45: 181-183.
- Oka, Y., Y. Cohen and Y. Spiegel. 1999. Local and systemic induced resistance to the root-knot nematode in tomato by DL- $\beta$ -amino-n-butyric acid. *Phytopathology*, 89: 1138-1143.
- Oka, Y. 2010. Mechanisms of nematode suppression by organic soil amendments – a review. *Applied Soil Ecology*, 44: 101-115.
- Roberts, E.H. 1972. *Cytological, genetical and metabolic changes in seed viability associated with loss of viability of seeds*. (Ed.) E.H. Roberts. Chapman and Hall Ltd, London, pp. 253-306.
- Ross, S.A., S.E. Megalla, D.W. Bisby and A.H. Awad. 1980. Studies for determining some antibiotic substance in some Egyptian plants. Screening of some antimicrobial activity. *Fitoterapia*, 51: 303-308.
- Shaukat, S.S., I.A. Siddiqui and F.S. Mehdi. 2003. *Avicennia marina* (mangrove) soil amendment changes the fungal community in the rhizosphere and root tissue of mung bean and contributes to control of root-knot nematodes. *Phytopathol Mediterr*, 42: 135-140.
- Sokal, R.R. and F.J. Rohlf. 1995. *Biometry: The principles and practices of statistics in biological research*. Freeman, San Francisco, USA, p. 887.
- Sullivan, G.A. 1979. Soybean seed treatment and storage study 1978. *Newsletter AOSA*, 53: 40-41.
- Tariq, M and S. Dawar. 2010. Impact of biocontrol bacteria with *Rhizophora nucronata* plant parts in suppression of *Meloidogyne javanica* (Treb.) Chitwood on crop plants. *Archives of phytopathology and Plant Protection*, 43(8): 754-760.
- Tariq, M and S. Dawar. 2011. Formulation of *Avicennia marina* pellets and its application in controlling root diseases in leguminous and non leguminous plants. *Pak. J. Bot.*, 43: 1411-1415.
- Taylor, D.P. and C. Netscher. 1974. An improved technique for preparing perennial pattern of *Meloidogyne* spp. *Nematologica*, 20: 268.

(Received for publication 31 January 2012)