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Role of Pellets and Capsules of *Acacia nilotica* and *Sapindus mukorossi* in Combination of Seed Bio-Priming with Microbial Antagonists in the Supression of Root Infecting Pathogenic Fungi and Promotion of Crop Plants

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Abstract

Present research investigate the effect of *Acacia nilotica* and *Sapindus mukorossi* pellets (pyrophyllite mixed with leaves powder @ 50:50 ratio), capsules (empty shells of capsules filled with leaves powder @ 0.5 g) alone or in combination with bio-priming of leguminous and non-leguminous seeds with microbial antagonists like *Trichoderma harzianum* and *Rhizoboium melilotii*. It was found that *A. nilotica, S. mukorossi* pellets and capsules in combination with biopriming of seeds with *T. harzianum* were most effective for the promotion of growth and reduction of root infecting fungal pathogens like *Macrophomina phaseolina, Rhizoctonia solani* and *Fusarium* sp on peanut, chickpea, okra and sunflower.

Keywords: Pellets; Capsules; Plant extracts; Microbial antagonists; Seed bio-priming

Introduction

Acacia nilotica (L.) Willd. *ex* Del. is also known as Gum Arabic tree, Babul, Egyptian thorn, or Prickly. *Acacia* is a multipurpose tree and play a great role in fixation of nitrogen [1]. Species of *Acacia* contains secondary metabolites including amines and alkaloids, cyanogenic glycosides, cyclitols, fatty acids and seed oils, fluoroacetate, gums, nonprotein amino acids, terpenes (including essential oils, diterpenes, phytosterol and triterpene genins and saponins), hydrolyzable tannins, flavonoids and condensed tannins [2]. *A. nilotica* also possess antimicrobial activity against some micro-organisms [3]. Mashram has observed the antimicrobial activity of *Acacia nilotica*, against micro-organisms like, *B. subtilis,* S. aureus and *E. coli in vitro* and found that the extracts of bark and leaf showed inhibition zone between 7.5-16 and 8-15.5 mm respectively and most effective against *E. coli.* and *B. subtilis.* recorded antifungal activity of methanolic extracts and aqueous extract of *A. nilotica* with percentage inhibition ranging from 34.27 ± 1.45 to 93.35 ± 1.99 [4]. Similarly *Sapindus mukorossi* (L.) is well known for its folk medicinal values. Recently many of the pharmacological actions of this plant have been explored which includes the antimicrobial cytoxoic molluscicidal insecticidal and fungicidal [5-9].The leaves are used in the baths to relieve joint pain and the roots are used in the treatment of gout and rheumatism. Since ancient times *S. mukorossi* has been used as a detergent for shawls and silks. The fruit of *S. mukorossi* was utilised by Indian jewellers for restoring the brightness of tarnished ornaments made of gold, silver and other precious metals [10]. Dawar et al. [11] studied that leaves, stem, bark and fruit powder of *Eucalyptus* sp., has ability to reduce the infection of root infecting fungi viz., *Fusarium sp., R.solani* and *M. phaseolina*.

Pelleting and encapsulation of plant parts gaining importance in recent times stated that pyrophyllite mixed *Avecenia marina* plant parts pellets (leaves and stem powder mixed separately with pyrophillite) had played a marked role in the elevation of growth parameters as well as in the reduction of soil borne root rot fungi like *F. oxysporum, M. phaseolina* and *R. solani* on cowpea and bringal plants [12]. Ghaffar, 1995 observed prominenet reduction in *M. phaseolina* infection on chickpea and mungbean plants when pellets of sodium alginate were mixed in the soil @ 1 and 10 pellets inside the plastic pots. Walker and Connick [13] used alginate type pellets in formulations of microbiological and chemical herbicides. According to, Tariq and Dawar [14] encapsulation of halophytic plant parts powder such as stem and leaves powder @ five capsules per pot prominently decreased the incidence of root-infecting pathogenic fungi on okra and mungbean plants.

Apart from pelleting and encapsulation techniques, bio-priming of seeds with beneficial micro-organisms is also gaining importance in the control of many plant pathogens as another alternative to synthetic fungicides in present times. Bio-priming of seeds with antagonistic microbes are capable of colonizing the rhizosphere by potentially providing the advantages to the plant beyond the seedling emergence stage. Using antagonistic microorganisms was such an approach which was used increasingly on a commercial scale both in field and green house crops. Antagonistic micro-organisms like *Trichoderma* species are important biological control agents (BCAs) of many soil borne plant pathogens [15]. Different mechanisms have been used by *Trichoderma* in order to control plant pathogens which include competition for nutrients and habitat, mycoparasitism, release of antibiotics and fungal cell wall breaking enzymes [15-17]. Besides fungal antagonistic microbes, beneficial bacterial isolates have also been used for seed bio-priming. These beneficial bacterial micro-organisms contain antifungal and good plant growth promoting attributes. Bacterial strains in soil that have tremendous effects on plant growth and health are commonly known as plant growth promoting rhizobacteria (PGPR). PGPR enhance plant growth indirectly or directly with the biocontrol of pathogens, production of plant hormones or improvement of plant nutritional status [18]. Present research work was therefore carried

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out on role of pellets and capsules of *A. nilotica* and *S. mukorossi* in combination of seed bio-priming with microbial antagonists in the suppression of root infecting pathogenic fungi and promotion of crop plants.

Materials And Methods

Collection of plants and antagonistic agents

A. nilotica and *S. mukorossi* leaves parts were collected from Campus of University of Karachi, air dried separately and finely powdered in an electric grinder. Cultures of *Rhizobium meliloti* and *Trichoderma harzianum* were obtained from the Karachi University Culture Collection (KUCC).

Preparation of pellets and filling of capsules

A. nilotica and *S. mukorossi* pellets were prepared with the help of research method of Tariq and Dawar [12]. Similarly, *A. nilotica* and *S. mukorossi* pellets were prepared with the help of multiple pellet sampler of equal size and weight (1 g pellet containing 0.5 g pyrophyllite and 0.5 g leaves powder). These pellets were air dried under laminar air flow chamber. Leaves powder of *A. nilotica* and *S. mukorossi* were filled in empty capsules (0.5 g in each capsule).

Preparation of spore/cell suspension and bio-priming of seeds

Spore/cell suspension of *T. harzainum* and *R. melilotii* were prepared in distilled sterilized water and seeds were bio-primed in these suspensions separately for about 10 minutes and then these seeds were air dried for sowing.

Pots setup under screen house

Plastic pots containing 300 g of soil were placed under screen house in Department of Botany, University of Karachi under randomized block design. These experimental pots containing

(a) amendment of *A. nilotica* and *S. mukorossi* pellets in the soil separately in which non treated seeds were sown

(b) *T. harzianum* and *R. melilotii* primed seeds were sown in the soil separately in non-amended soil

(c) combined effect of both bioprimed seeds and pellets amended soil

(d) only pyrophyllite pellets were mixed in the soil which served as control no. 1

(e) 0.5 g of *A. nilotica* and *S. mukorossi* leaves powder were filled in empty capsules separately and mixed in the soil containing non treated seeds

(f) combined effect of both capsules and bioprimed seeds

(g) empty capsules mixed in the soil which regarded as control no. 2. These experimental pots were kept under screen house for about 30 days and for maintaining the moisture content watered regularly. After 30 days the plants were uprooted for the observation of growth parameters and their roots were washed in running tap water for estimation of roots colonization by root infecting fungi.

Statistical Analysis

Data obtained from the experiment were analysed with the help of ANOVA (analysis of variance) and LSD (least significant difference) test at P=0.05 and DMRT (Duncan's multiple range test) in order to compare treatment means, using STATISCA computer software.

Results

There was significant ($p<0.001$) enhancement in growth parameters of peanut when *A. nilotica* pellets (pyrophyllite and leaves powder @ 50:50 ratio) in combination with bio-priming of seeds with *T. harzianum* spore suspension was used as compared to the control in which only pyrophyllite pellets was amended in the soil (Figure 1). Similarly combine effect of *A. nilotica* capsules (filled with 0.5 g leaves powder) and bio-priming of peanut seeds with *T. harzianum* spore suspension gave significant increase in root length, shoot length, root weight and shoot weight and prominent decrease was also noticed in the colonization *of Fusarium* sp ($p<0.001$), *R. solani* ($p<0.01$) and *M. phaseolina* ($p<0.5$) as compared to the control (empty shells of capsules) (Figure 1). In chickpea, root length, shoot length, root weight and shoot weight increased significantly ($p<0.001$) when *A. nilotica* pellets and capsules were amended in the soil and seeds were bio-primed with *T. harzianum*, whereas, *M. phaseolina* reduced significantly ($p<0.5$) in comparision with both the controls (Figure 2). Combine impact of *A. nilotica* (pellets) and *S. mukorossi* (capsules) along with *T. harzianum* bio-primed seeds significantly ($p<0.001$) elevated the growth parameters of okra and significant suppression in root infecting fungal pathogens like *R. solani* ($p<0.001$) and *Fusarium* sp ($p<0.001$)

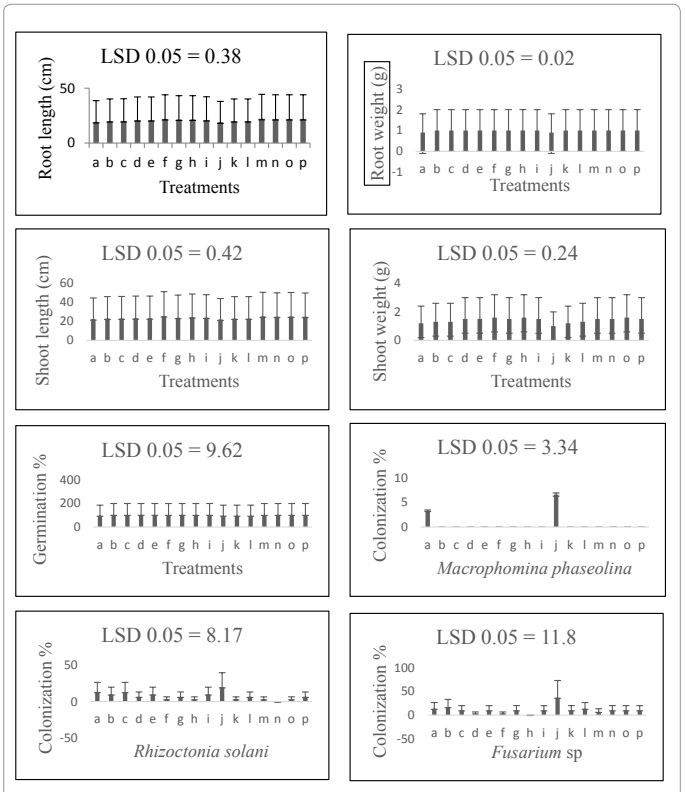

Figure 1: Role of pellets and capsules of *Acacia nilotica* and *Sapindus mukorossi* in combination of seed bio-priming with microbial antagonist in the suppression of root infecting pathogenic fungi and promotion of peanut plants. (a) Control (b) *A. nilotica* pellets (c) *S. mukorossi* pellets (d) Bio-priming with *T. harzainum (*e) Bio-priming with *R. melilotii* (f) Combined effect of *A. nilotica* pellets and bio-priming with *T. harzianum* (g) Combined effect of *A. nilotica* pellets and bio-priming with *R. melilotii* (h) Combined effect of *S. mukorossi* pellets and bio-priming with *T. harzianum* (i) Combined effect of *S. mukorossi* pellets and bio-priming with *R. melilotii* (j) Control (k) *A. nilotica* capsules (l) *S. mukorossi* capsules (m) Combined effect of *A. nilotica* capsules and bio-priming with *T. harzianum* (n) Combined effect of *A. nilotica* capsules and bio-priming with *R. melilotii* (o) Combined effect of *S. mukorossi* capsules and bio-priming with *T. harzianum* (p) Combined effect of *S. mukorossi* capsules and bio-priming with *R. melilotii*.

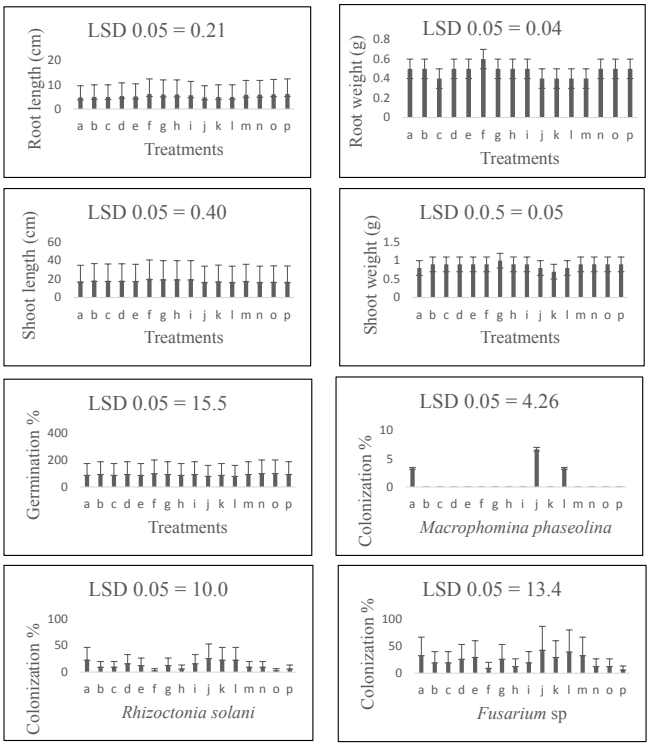

Figure 2: Role of pellets and capsules of *Acacia nilotica* and *Sapindus mukorossi* in combination of seed bio-priming with microbial antagonist in the suppression of root infecting pathogenic fungi and promotion of chickpea plants. (a) Control (b) *A. nilotica* pellets (c) *S. mukorossi* pellets (d) Bio-priming with *T. harzainum (*e) Bio-priming with *R. melilotii* (f) Combined effect of *A. nilotica* pellets and bio-priming with *T. harzianum* (g) Combined effect of *A. nilotica* pellets and bio-priming with *R. melilotii* (h) Combined effect of *S. mukorossi* pellets and bio-priming with *T. harzianum* (i) Combined effect of *S. mukorossi* pellets and bio-priming with *R. melilotii* (j) Control (k) *A. nilotica* capsules (l) *S. mukorossi* capsules (m) Combined effect of *A. nilotica* capsules and bio-priming with *T. harzianum* (n) Combined effect of *A. nilotica* capsules and bio-priming with *R. melilotii* (o) Combined effect of *S. mukorossi* capsules and bio-priming with *T. harzianum* (p) Combined effect of *S. mukorossi* capsules and bio-priming with *R. melilotii*.

was also observed (Figure 3). In sunflower, combined application of (*A. nilotica* pellets and biopriming of seeds with *T. harzianum*) and (encapsulation of *S. mukorossi* and *T. harzianum* primed seeds) gave significant ($p<0.001$) health and vigour to growth parameters such as root length, shoot length, root weight and shoot weight and significant decrease in root infecting fungi such as *Fusarium sp* ($p<0.001$), *R. solani* ($p<0.001$) and *M. phaseolina* ($p<0.5$) was also recorded (Figure 4). Of all the treatments and amendments, it was observed that pellets and capsules of *A. nilotica*, and *S. mukorossi* in combination with biopriming of seeds with *T. harzianum* spore suspension was found to be most effective for the promotion of growth and suppression of root infecting pathogenic fungi like *Macrophomina phaseolina, Rhizoctonia solani* and *Fusarium* sp on leguminous and non-leguminous crops.

Discussion

In the present research, encapsulation and pyrophyllite mixed pellets of *A. nilotica* and *S. mukorossi* alongwith *T. harzainum* primed seeds played a marked role in the enhancement of growth parameters as well as in the reduction of fungal pathogens like *M. phaseolina, R. solani* and *Fusarium sp* of leguminous and non-leguminous crops. Significant ($p<0.001$) enhancement in growth parameters of peanut was noticed when *A. nilotica* pellets (pyrophyllite and leaves powder @ 50:50 ratio) in combination with bio-priming of seeds with *T. harzianum* spore suspension was used. Tariq and Dawar [12] used formulations of *A. marina* plant parts pellets with pyrophyllite @ 50:50 ratio and observed that there was prominent increase in growth parameters of leguminous and non-leguminous plants and root infecting fungi like *M. phaseolina, R. solani* and *Fusarium* sp was also reduced significantly .Our results showed that in chickpea, root length, shoot length, root weight and shoot weight increased significantly with the combined effect of *A. nilotica* pellets, capsules and *T. harzianum* primed seeds whereas, *M. phaseolina* reduced significantly. Similarly, when mixed alginate pellets in the soil, there was significant suppression in the colonization of *M. phaseolina* on mung bean and chickpea plants [19]. According to our results, combined application of *A. nilotica* (pellets) and *S. mukorossi* (capsules) along with *T. harzianum* bio-primed seeds significantly elevated the growth parameters of okra and significant suppression in root infecting fungal pathogens like *R. solani* and *Fusarium sp* was also observed. Tariq and Dawar [20] investigate the effect of mangrove plant parts filled capsules and pellets and recorded that formulations of mangrove combined parts powder and pellets when amended in the soil found to releases compounds which are nematicidal in nature and reduced the activity of *Meloidogyne javanica* on okra and mungbean plants and thus increase plant growth and crop yield.

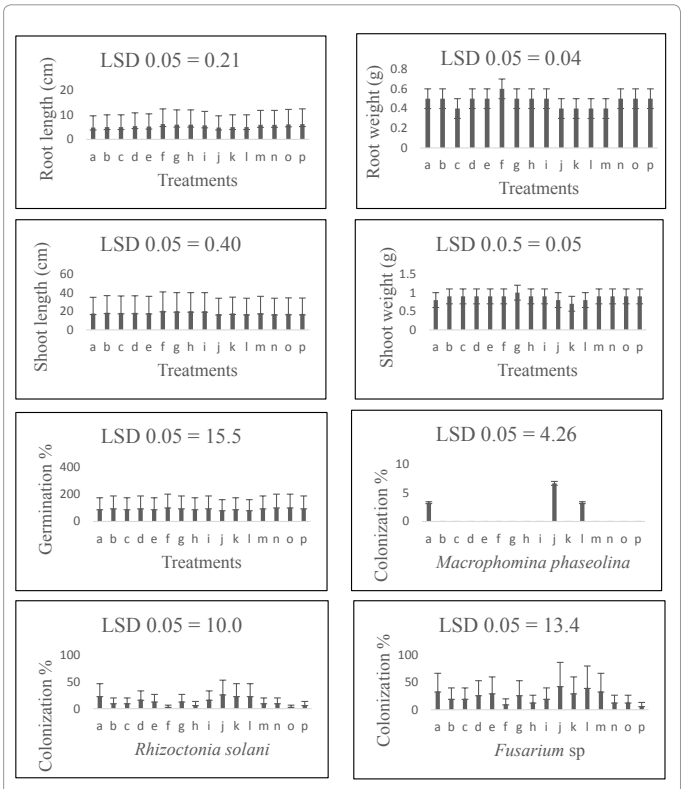

Figure 3: Role of pellets and capsules of *Acacia nilotica* and *Sapindus mukorossi* in combination of seed bio-priming with microbial antagonist in the suppression of root infecting pathogenic fungi and promotion of okra plants. (a) Control (b) *A. nilotica* pellets (c) *S. mukorossi* pellets (d) Bio-priming with *T. harzainum (*e) Bio-priming with *R. melilotii* (f) Combined effect of *A. nilotica* pellets and bio-priming with *T. harzianum* (g) Combined effect of *A. nilotica* pellets and bio-priming with *R. melilotii* (h) Combined effect of *S. mukorossi* pellets and bio-priming with *T. harzianum* (i) Combined effect of *S. mukorossi* pellets and bio-priming with *R. melilotii* (j) Control (k) *A. nilotica* capsules (l) *S. mukorossi* capsules (m) Combined effect of *A. nilotica* capsules and bio-priming with *T. harzianum* (n) Combined effect of *A. nilotica* capsules and bio-priming with *R. melilotii* (o) Combined effect of *S. mukorossi* capsules and bio-priming with *T. harzianum* (p) Combined effect of *S. mukorossi* capsules and bio-priming with *R. melilotii*.

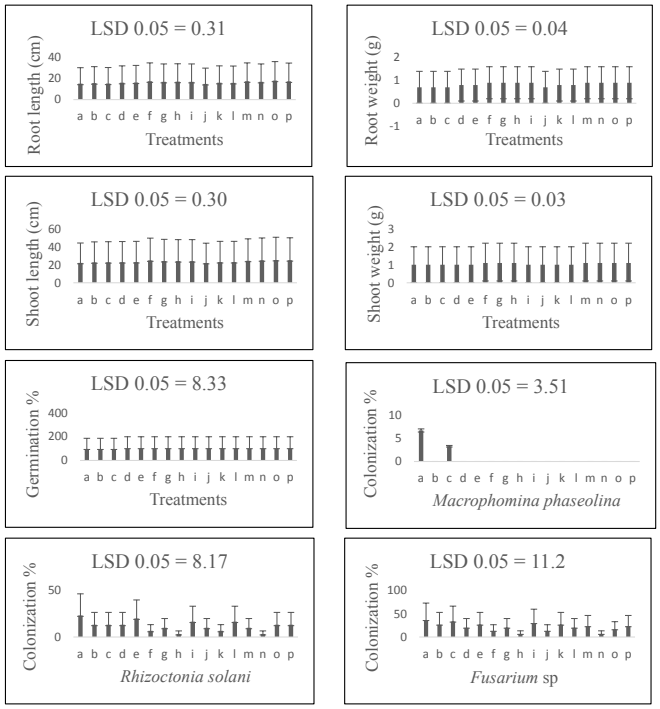

Figure 4: Role of pellets and capsules of *Acacia nilotica* and *Sapindus mukorossi* in combination of seed bio-priming with microbial antagonist in the suppression of root infecting pathogenic fungi and promotion of sunflower plants. (a) Control (b) *A. nilotica* pellets (c) *S. mukorossi* pellets (d) Bio-priming with *T. harzainum* (e) Bio-priming with *R. melilotii* (f) Combined effect of *A. nilotica* pellets and bio-priming with *T. harzianum* (g) Combined effect of *A. nilotica* pellets and bio-priming with *R. melilotii* (h) Combined effect of *S. mukorossi* pellets and bio-priming with *T. harzianum* (i) Combined effect of *S. mukorossi* pellets and bio-priming with *R. melilotii* (j) Control (k) *A. nilotica* capsules (l) *S. mukorossi* capsules (m) Combined effect of *A. nilotica* capsules and bio-priming with *T. harzianum* (n) Combined effect of *A. nilotica* capsules and bio-priming with *R. melilotii* (o) Combined effect of *S. mukorossi* capsules and bio-priming with *T. harzianum* (p) Combined effect of *S. mukorossi* capsules and bio-priming with *R. melilotii*.

Besides pelleting and encapsulation reports, several studies on bio-priming of seeds with beneficial micro-organisms have shown that seed bio-priming with microbial antagonists played a prominent role in the health and vigour of seedlings as well as in the reduction of many soil borne diseases. Rafi and Dawar [21] studied the effects of bio-priming on leguminous and non-leguminous crops at different time intervals and concluded that growth parameters markedly improved when seeds were bio-primed with microbial antagonists like *Trichoderma harzianum* and *Rhizobium meliloti* for 10 minutes whereas, root infecting fungal pathogens reduced significantly when seeds were bio-primed with *T. harzianum*, *Bacillus* sp and *R. meliloti* conidial/cell suspensions for 5, 10 and 20 minutes time interval .Present investigation found that in sunflower, combined application of (*A. nilotica* pellets and biopriming of seeds with *T. harzianum*) and (encapsulation of *S. mukorossi* and *T. harzianum* primed seeds) gave maximum growth parameters such as root length, shoot length, root weight and shoot weight and significant decrease in root infecting fungi was also recorded. Harman [22] observed that strain T-22 of *Trichoderma harzianum* causes plant to have more extensive root systems and not only suppressed the diseases but also improve the plant metabolism. Present research investigation clearly suggests that pelleting and capsulation of *A. nilotica*, and *S. mukorossi* in combination with biopriming of seeds with *T. harzianum* spore suspension was found to be most effective for the promotion of growth and suppression of root infecting pathogenic fungi like *Macrophomina phaseolina, Rhizoctonia solani* and *Fusarium* sp on all the tested plants.

References

1. Bargal K, Bargali SS (2009) Acacia nilotica: a multipurpose leguminous plant. Nature and Science 7: 11-19.
2. Seigler DS (2003) Phytochemistry of Acacia-sensu lato. Biochemical Systematics and Ecology 31: 845-873.
3. Mashram N (2009) Antimicrobial activity of methanol extracts of medicinal plants against bacterial species. International Research Journal I: 147-150.
4. Mahesh B, Satish S (2008) Antimicrobial activity of some important medicinal plant against plant and human pathogens. World Journal of Agricultural Sciences 4: 839-843.
5. Ibrahim M, Khan AA, Tiwari SK, Habeeb MA, Khaja MN (2006) Antimicrobial activity of Sapindus mukorossi and Rheum modi extract against Helicobacter pylori: invitro and invivo studies. World J Gasteroenterol 12: 7136-7142.
6. Sengupta D, Pal R, Dwivedi AK, Singh S (2004) Characterisation of saponin in Sapindus mukorossi saponin and quantitative determination of sapindoside. BJ Sci Ind Res 63: 181-186.
7. Huang HC, Liao SC, Chang, Kuo YH, Wu YC (2003) Molluscicidal saponins from sapindus mukorossi, inhibitory agents of golden apple snails pomaceae canaliculata. J Aric Food Chem 51: 4916-4919.
8. Geyter ED, Lambert E, Geelan D, Smagee G (2007) First results on the insecticidal action of saponins. Commun. Agri Appl Biol Sci 72: 645-648.
9. Tanaka O, Tamura Y, Masuda H, Mizutani K (1996) Applications of saponins in food and cosmetics:saponins of Mohova yucca and Sapindus mukorossi Gaertn, In: Saponins used in food and agriculture. Plenum Press, NewYork pp. 1-11.
10. Singh N, Kaur A, Yadav K (2010) A reliable invitro protocol for rapid mass propagation of Sapindus mukorossi. Nature and Science 8: 41-47.
11. Dawar S, Younus SM, Tariq M, Zaki MJ (2007) Use of Euclyptus in the control of root infecting fungi on mung bean and chick pea. Pak J Bot 39: 975-979.
12. Tariq M, Dawar S (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.
13. Walker HL, Connick WJ (1983) Sodium alginate for production and formulation of mycoherbicides. Weed Sci 31: 333-338.
14. Tariq M, Dawar S (2013) Encapsulation of halophytic plant parts powder in suppression of root rot diseases. Archieves Phytopathol and Plant Protection 46: 322-325.
15. Benitez T, Rincon AM, Limon M, Carmen M, Codon A (2004) Bioncontrol mechanisms of Trichoderma strains. International Microbiology 7: 249-260.
16. Kubicek CP, Mach RL, Peterbauer CK, Lorito M (2001) Trichoderma: From genes to biocontrol. Journal of Plant Pathology 83: 11-23.
17. Harman GE, Howell R, Charles R, Viterbo R, Chet A, et al. (2004) Trichoderma species: opportunistic, avirulent plant symbionts. Nature Reviews Microbiology 2(1): 43-56.
18. Glick BR (1995) The enhancement of plant growth by free living bacteria. Can J Microbial., 41: 109-117.
19. Ghaffar A (1995) Mass production of biocontrol agents for field application and plant disease control. PSF Project. Department of Botany, University of Karachi, Karachi-75270, Pakistan. p. 86.
20. Tariq M, Dawar S (2015) Mangrove formulations for the management of Meloidogyne javanica (Treub) chitwood under field conditions. Pak J Bot 47: 347-352.
21. Rafi H, Dawar S (2014) Effects of bio-priming of leguminous and non-leguminous crop seeds in the management of root rot fungi and growth of crop plants. Int J Biol Biotech 11: 375-382.
22. Harman GE (2000) Myths and Dogmas of biocontrol. Changes in perceptions derived from research on Trichoderma harzianum T-22. Plant Dis 84: 377-393.