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SEED PRIMING WITH EXTRACTS OF *ACACIA NILOTICA* (L.) Willd. *ex* Delile AND *SAPINDUS MUKOROSSI* (L.) PLANT PARTS IN THE CONTROL OF ROOT ROT FUNGI AND GROWTH OF PLANTS

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Abstract

Seed priming with plant extracts and chemicals has been used as an important growth enhancement tool in crop plants. In this research, an attempt was made to understand the mechanism of various seed priming treatments on greenhouse-grown okra (*Abelmoschus esculentus* (L.) Moench.), sunflower (*Helianthus annuus* L.), peanut (*Arachis hypogaea* L.) and chickpea (*Cicer arietinum* L.) for the control of root infecting fungi like *Rhizoctonia solani* (Kühn), *Fusarium* spp. and *Macrophomina phaseolina* (Tassi) Goid by plant parts extracts (stem, leaves and seeds) of *Acacia nilotica* (L.) Willd. *ex* Delile and *Sapindus mukorossi* (L) at different time intervals (5, 10, 20, 40 minutes). Results showed significant suppression of root rot fungi and significantly enhanced the growth parameters like shoot length, root length, shoot weight and root weight. Seed-priming with *A. nilotica* and *S. mukorossi* leaves extract for 10 minutes time interval was found to be effective for the control of root rot fungi and growth of all tested leguminous and non-leguminous plants.

Key words: Seed priming, Plant extarct, Leguminous and non leguminous crops, Root rot fungi, Plant growth.

Introduction

Priming is one of the most important developments to help rapid and uniform germination and emergence of seeds and to increase seed tolerance to adverse environmental conditions (Harris *et al.*, 1999). Seed priming not only improves the speed and uniformity of germination (Khan *et al.,* 2008; Khalil *et al.,* 2010) but also stimulates various biochemical changes in the seed, which are vital in breaking dormancy, the mobilisation or hydrolysis of seed reserves, enzyme activation, and the emergence of embryonic tissues (Asgedom & Becker, 2001; Çatav *et al*., 2012). According to El-Mougy and Abdel-Kader (2008) primed faba bean seeds showed a highly significant effect causing complete reduction of root rot incidence at both pre and post-emergence stages of plant growth compared with the control treatment. Seed treatment control the fungi residing on the surface of seed or inside the seed and are affective against pathogen that reside in the soil and cause seed rot, damping off and root rots (Martha *et al*., 2003).

According to Garrett (1970) root infecting fungi constitute a substantial and omnipresent threat to the welfare of all crops. Root rot caused by *Aphanomyces euteiches, R. solani, Fusarium* spp*., Sclerotium rolfsii* are the most destructive soil-borne diseases of pea, chickpea, lentil, faba bean and lupine (Abou-Zeid *et al*., 1997; Abdel-Kader *et al*., 2002; Infantin *et al*., 2006). *Macrophomina phaseolina* (Tassi) Goid is reported to produce charcoal rot, seedling blight, root rot, stem rot, pod rot on more than 500 species of plants (Dhingra & Sinclair, 1978). Several *Rhizoctonia* spp cause root rot, but the most common is *Rhizoctonia solani* which causes wilting and death in several hundred genera of plants. *Rhizoctonia* root rot is favoured by relatively high temperatures and intermediate moisture (Steve, 2001). An average yield loss of 2.2 ha in pea was observed due to root rot diseases caused by *Fusarium solani* and *F. oxysporum* with complete loss in many cases (Tu, 1987).

Extracts obtained from some crop and tree residues have been reported to play roles in crop growth and yield (Chung & Miller, 1995; El Atta & Bashir, 1999; Ahmed & Nimer, 2002; Farooq *et al*., 2008). Babu *et al.* (2008) investigated plant extracts and their compounds for control of fungal pathogens and found that plant extract significantly inhibited the radial growth of isolated fungus. The formulation of plant extract can be successfully devised as fungicides using a simple process with minimum instrumentation and few chemical agents. These formulations may be considered suitable for seed and foliar treatment. Present work was therefore carried out on effect of seed priming with *A. nilotica* and *S. mukorossi* plant part extracts in the control of root rot fungi and growth of okra, sunflower, chickpea and peanut.

Materials and Methods

Collection of material: Plant parts of *A. nilotica* and *S. mukorossi* were collected from Campus of University of Karachi, dried separately and ground in an electric grinder.

Extract preparation: Aqueous extracts of *A. nilotica* and *S. mukorossi* plants parts were prepared by soaking the stem, leaves, and seeds powder separately for whole night in distilled water (10 g powder and 90 ml distilled water). The suspension was filtered through Whatman's filter paper in order to get aqueous extracts for seed priming.

Soil used: Soil used was obtained from experimental plot of Department of Botany, University of Karachi. The sandy loam soil containing (sand, silt, clay, 70, 11 and 10%), pH ranged from 7.1-9.65 with moisture holding capacity (MHC) of 49% (Keen & Raczkowski, 1922), total nitrogen 0.077-0.099% (Mackenzie & Wallace, 1954), 3-7 sclerotia of *M. phaseolina* g^{-1} as found by wet sieving technique (Sheikh & Ghaffar, 1975), 5-20% of *R. solani* on sorghum seeds used as baits (Wilhelm, 1955) and *Fusarium* spp., 2000 cfu g^{-1} as assessed by soil dilution technique (Nash & Synder, 1962).

Seed priming: Peanut, chickpea, sunflower and okra seeds were primed with extracts of leaves, stem, and seeds of *A. nilotica* and *S. mukorossi* for 5, 10, 20 and 40 minutes and non-primed seeds were used as control. 5 seeds were sown in

8 cm diam., plastic pots and each containing 300g soil and watered regularly to maintained sufficient moisture required for the growth of plants. The pots were kept under screen house in randomized complete block design with three replicates per treatment at different time intervals. Pots containing un-treated seeds were also kept under screen house which served as control. Germination and growth parameters like shoot length, root length, shoot weight and root weight was observed. Colonization percentage was also recorded after 30 days of seed germination.

Determination of root infecting fungi: To determine the incidence of root rot fungi, one cm long root pieces of leguminous and non-leguminous plants after washing in running tap water were surface sterilized with 1% Ca $(OCl)_2$ and transferred on PDA (Potato dextrose agar) containing plates supplemented with Penicillin @ 200 mg and streptomycin @ 200 mg/litre (5 root pieces per plate). Petri dishes were incubated at room temperature for 5 to 7 days and colonization of roots by root infecting fungi was recorded after incubation period.

Statistical analysis: Data were subjected to analysis of variance (ANOVA) followed by the least significant difference (LSD) test at P = 0.05 and Duncan's multiple range test to compare treatment means, using statistica software according to Sokal & Rohlf (1995).

Results

There was significant enhancement of growth parameters of peanut, chickpea, sunflower and okra when seeds were primed with *A. nilotica* and *S. mukorossi* stem, leaves and seeds extracts at different time intervals (5, 10, 20 and 40 minutes). Significant increase in germination ($p<0.001$) was observed, when peanut, okra, chickpea and sunflower seeds were primed with *A. nilotica* and *S. mukorossi* stem, leaves and seeds extracts for 10 minutes time interval (Fig. 1-4) .In peanut, root length and root weight significantly increased ($p<0.001$) when seeds were primed with *A. nilotica* and *S. mukorossi* leaves extract whereas, shoot length and shoot weight significantly increased ($p<0.001$) when seeds were primed with *A. nilotica* and *S.mukorossi* leaves and seeds extracts for 10 minutes (Figs. 1 & 3). In okra, shoot length significantly increased ($p<0.001$) when seeds were treated with *A. nilotica* and *S. mukorossi* leaves and stem extracts for 10 minutes time interval (Figs. 1 & 3). In Chickpea, root length and shoot length were significantly increased ($p<0.001$) whereas, root weight and shoot weight were found to be significant ($p<0.01$) when seeds were treated with *A. nilotica* and *S. mukorossi* leaves extract for 10 minutes (Fig. 2 & 4). When sunflower seeds were primed with *A. nilotica* and *S. mukorossi* leaves extract for 10 minutes. Root length, shoot length, root weight and shoot weight were increased significantly ($p<0.001$) (Figs. 2 & 4). Priming of seeds with *A. nilotica* and *S. mukorossi* plant parts extracts showed significant suppression of root rot fungi like *Fusarium* spp., *R. soloni* and *M. phaseolina.* Highest reduction ($p<0.001$) in infection of *R. soloni* on peanut and *M. phaseolina* on okra was observed when seeds were primed with *A. nilotica* and *S. mukorossi* leaves and stem extracts for 10 minutes of time interval (Figs. 1 & 3) whereas, significant reduction ($p<0.001$) in *M. phaseolina* infection was found when leguminous and non-leguminous seeds were primed with seeds extract for 10 and 20 minutes time intervals (Figs. 1-4). It was found that, of the three plant parts extracts of *A. nilotica* and *S. mukorossi* used for seed priming, leaves extract and 10 minutes time interval was best for the enhancement of plant growth and reduction of root rot fungi.

Discussion

In the present study, seed priming with *A. nilotica* and *S. mukorossi* parts showed significant reduction in root rot infection of *Fusarium* spp.*, R. solani* and *M. phaseolina* in peanut, okra, sunflower and chickpea. *Acacia* species is one of the richest sources of bioactive flavonoids, alkaloids, phenolics, saponins, polysaccharides, tannins and terpenoids (Seigler, 2003). According to Sakhthivel *et al*. (2012), *A. nilotica* extracts could be used as natural anticancer agent for human health. The aqueous extracts of *A. nilotica* have inhibitory effect on carrageenan induced paw edema and yeast-induced pyrexia in rats. It also produced a significant increase in the hot plate reaction time in mice (Dafallah *et al*., 1996). By using powdered plant parts of *Prosopis juliflora* on mung bean and cowpea, significant reduction in the incidence of root rot fungi and enhancement of growth parameters was observed (Ikram & Dawar, 2013). *S. mukorossi* is an extremely valuable medicinal plant, distributed in tropical and sub-tropical regions of Asia. The major compounds isolated from *S. mukorossi* are triterpenoidal saponins of mainly three oleanane, dammarane and tirucullane types. Many of the pharmacological actions of this plant have been explored which includes the antimicrobial, cytotoxic, molluscicidal, insecticidal, piscicidal and fungicidal activities (Upadhyay & Singh, 2012). The crude extract of *S. mukorossi* exhibits a strong growth inhibition against the pathogenic yeast *Candida albicans*, which causes cutaneous candidiasis (Upadhyay & Singh, 2012). Present observation showed significant increase in germination and growth parameters of all tested plants with the use of plant extracts of *A. nilotica* and *S. mukorossi.*

Priming treatment is potentially prominent to induce profound changes in plant characteristics and to encourage more uniform seed germination and plants growth associated with fungi and bacteria coatings (Entesari *et al.,* 2013). *Avicennia marina* plant parts like leaves, stem and pneumatophore used as seed pelleting agents has a potential to increase growth and viability of cowpea and mung bean seeds and to suppress nematode population when stored at cold temperature (4□C) over a long period of time (Tariq & Dawar, 2012). In several cases, inoculation of seeds with biological agents in combination with priming has been reported to enhance and stabilize the efficacy of biological agents (Moeinzadeh *et al*., 2010; Bennett & Whipps, 2008). Priming of wheat seeds with 0.3% Zn significantly increased the mean shoot dry mass (Harris *et al*., 2008). Many reports on seed germination, mean germination time, seed vigor, root length, shoot length, primary establishment and seed emergence revealed the beneficial effects of seed priming (Farooq *et al*., 2006). Present results suggest that *A. nilotica* and *S. mukorossi* provide significant results in the control of root infecting fungi and to increase the growth and productivity of crop. Experiments would therefore carried out under field condition to control root rot fungi for obtaining the good quality of crop.

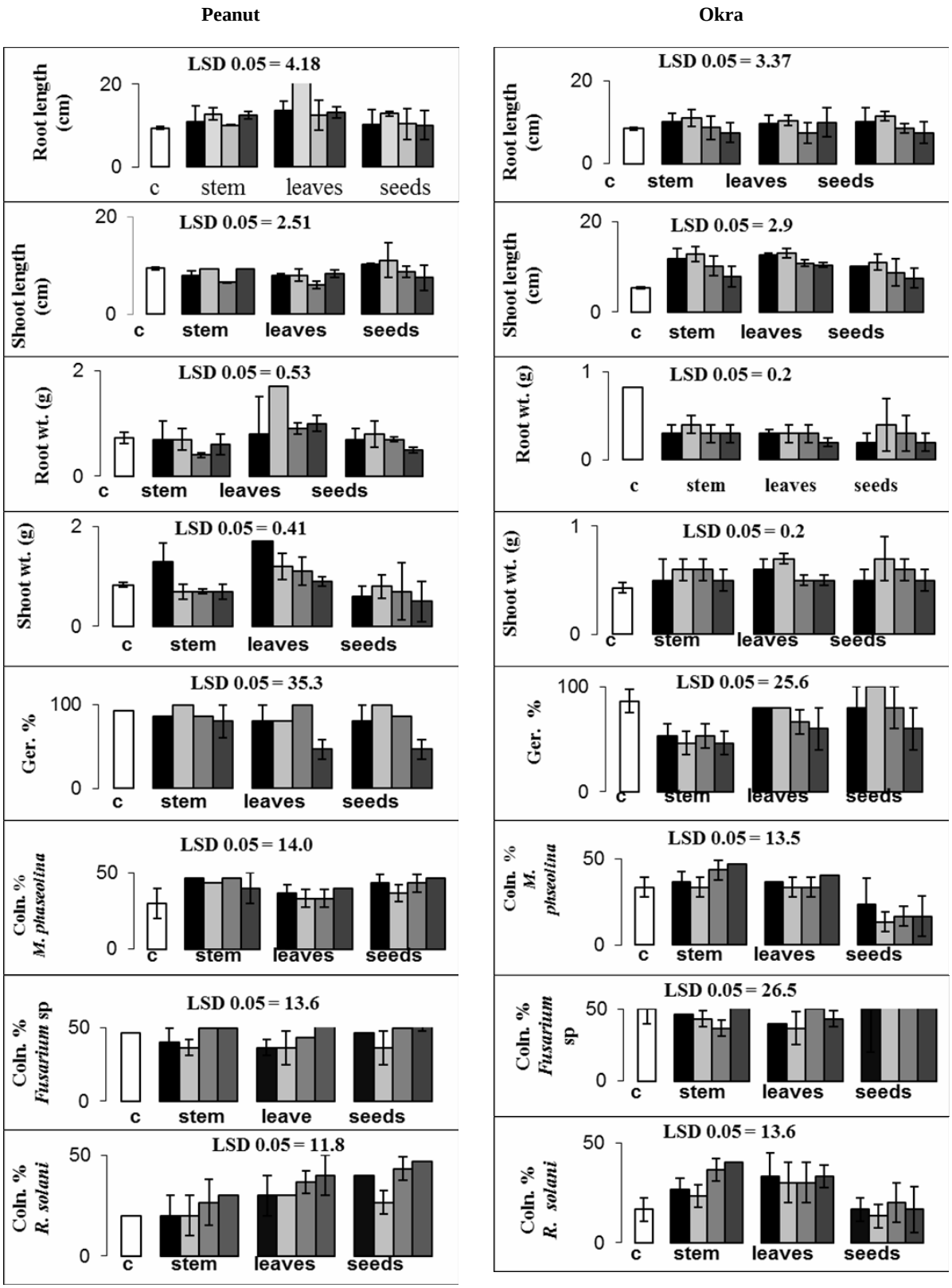

Fig. 1. Effect of *Acacia nilotica* extracts on growth parameters of peanut, okra and control of root rot fungi.
Time Intervals: Control 5 min 10 min 20 min 40 min

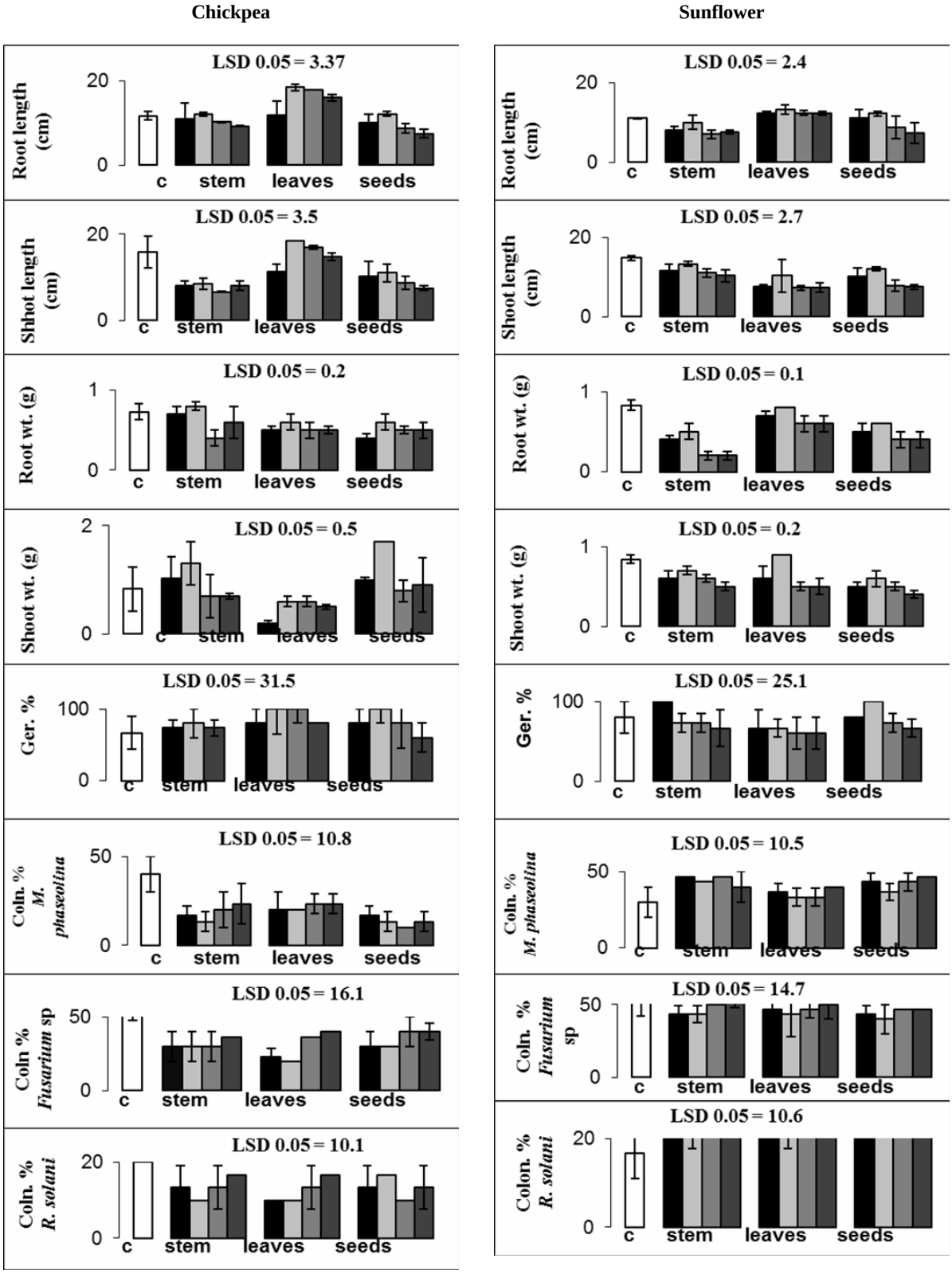

Fig. 2. Effect of *Acacia nilotica* extracts on growth parameters of chickpea, sunflower and control of root rot fungi.
Time Intervals: Control 5 min 10 min 20 min 40 min

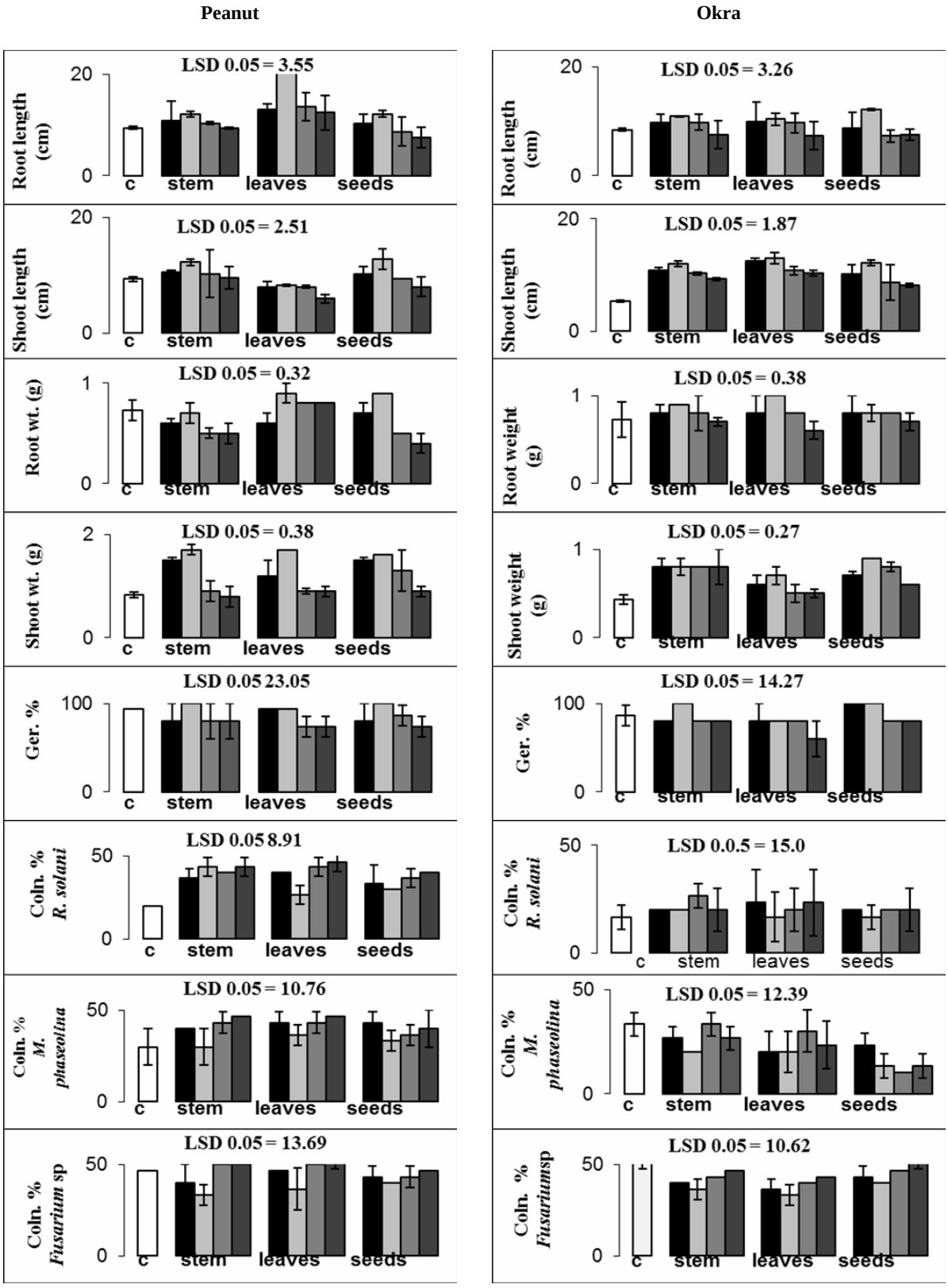

Fig. 3. Effect of *Sapindus mukorossi* extracts on growth parameters of peanut, okra and control of root rot fungi.
Time Intervals: Control 5 min 10 min 20 min 40 min

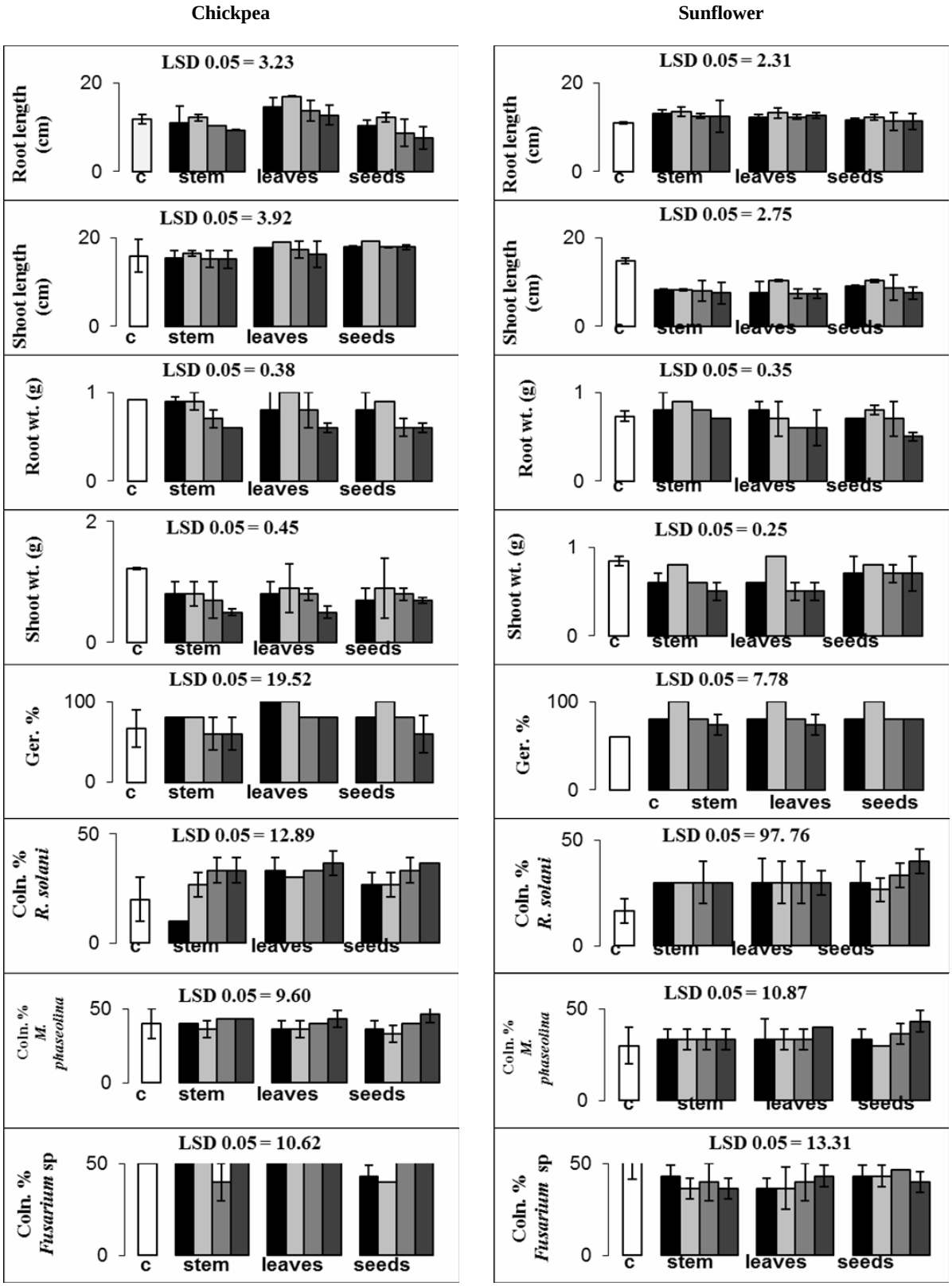

Fig. 4. Effect of *Sapindus mukorossi* extracts on growth parameters of chickpea , sunflower and control of root rot fungi.
Time Intervals: Control 5 min 10 min 20 min 40 min

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