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**BIOPHYSICAL CHARACTERIZATION OF MAGNETIC  
FIELD INDUCED ENHANCEMENT IN CHICKPEA  
(*Cicer arietinum* L.) SEEDS IN RELATION TO SOIL  
MOISTURE STRESS**

**NILIMESH MRIDHA**



**DIVISION OF AGRICULTURAL PHYSICS  
INDIAN AGRICULTURAL RESEARCH INSTITUTE  
NEW DELHI – 110 012 (INDIA)**

**2009**

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FIELD INDUCED ENHANCEMENT IN CHICKPEA  
(*Cicer arietinum* L.) SEEDS IN RELATION TO SOIL  
MOISTURE STRESS**

By  
**NILIMESH MRIDHA**

**A Thesis**

Submitted to the Faculty of the Post-Graduate School,  
Indian Agricultural Research Institute, New Delhi,  
in partial fulfilment of the requirements  
for the award of the degree of

**MASTER OF SCIENCE**  
**in**  
**AGRICULTURAL PHYSICS**  
**2009**

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		<b>Dr. C. Bharadwaj</b>	_____

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## CERTIFICATE

This is to certify that the thesis entitled **“Biophysical characterization of magnetic field induced enhancement in chickpea (*Cicer arietinum* L.) seeds in relation to soil moisture stress”** submitted to the Faculty of the Post-Graduate School, Indian Agricultural Research Institute, New Delhi in partial fulfillment of **Master of Science in Agricultural Physics**, embodies the results of *bona-fide* research work carried out by **Mr. Nilimesh Mridha, Roll No. 4651** under my guidance and supervision, and that no part of this thesis has been submitted for any other degree or diploma.

This assistance and help availed during the course of investigation as well as source of information have been duly acknowledged by him.

Date:  
Place: New Delhi

**Shantha Nagarajan**  
Chairman  
Advisory committee

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Date:

(Nilimesh Mridha)

Place: New Delhi

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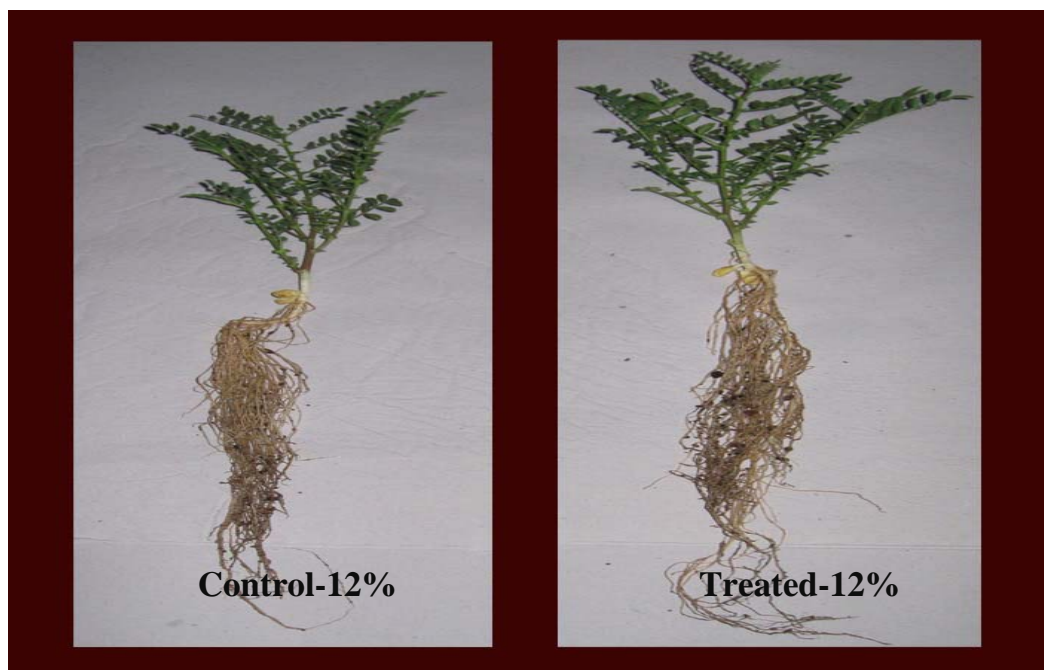
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**Plate 4.3.3a: Effect of magnetic field exposure on root development in Pusa 256 under 20% moisture level in sand culture**



**Plate 4.3.3b: Effect of magnetic field exposure on root development in Pusa 256 under 12% moisture level in sand culture**

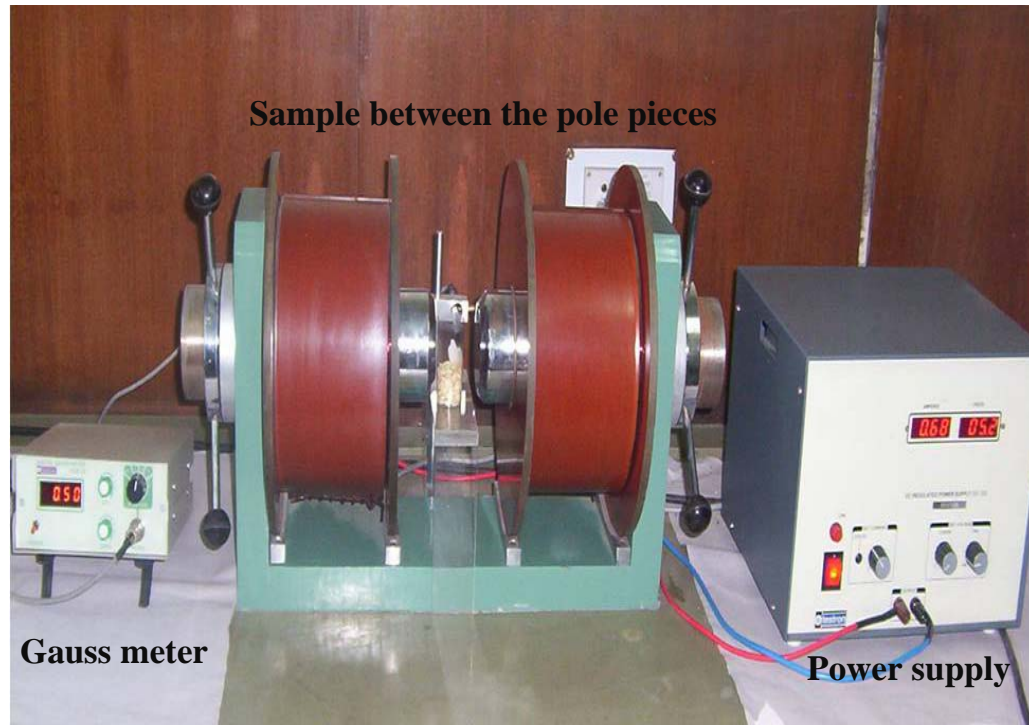


**Plate 4.3.1a: Effect of magnetic field exposure on root development in Pusa 1053 under 20% moisture level in sand culture**



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**Plate 3.3.2: Electromagnetic field generator**



**Plate 3.3.4. Root Scanner (LA 1600)**



**Plate 4.4.4c: Effect of magnetic field exposure on root development at flowering stage in Pusa 256 under -0.1 MPa soil moisture potential**



**Plate 4.4.4d: Effect of magnetic field exposure on root development at flowering stage in Pusa 256 under -0.2 MPa soil moisture potential**





**Plate 4.4.4a: Effect of magnetic field exposure on root development at flowering stage in Pusa 1053 under -0.1 MPa soil moisture potential**



**Plate 4.4.4b: Effect of magnetic field exposure on root development at flowering stage in Pusa 1053 under -0.2 MPa soil moisture potential**



**Table 4.2.3a: Relative water absorption and diffusivity of Pusa 1053 at soil water potential of –0.1 MPa (calculated using Crank’s equation)**

$t_g = 30$  h;  $M_g = 0.298$  g/seed

$t_g = 32$  h;  $M_g = 0.292$  g/seed

Time (h)	Treated			Control		
	$M_t/M_g$	$(Dt/a^2)^{1/2}$	$D \times 10^{-5}$ $\text{cm}^2 \text{min}^{-1}$	$M_t/M_g$	$(Dt/a^2)^{1/2}$	$D \times 10^{-5}$ $\text{cm}^2 \text{min}^{-1}$
2	0.237	0.07	0.624	0.215	0.065	0.538
4	0.359	0.12	0.917	0.36	0.12	0.917
10	0.680	0.26	1.722	0.61	0.23	1.348
16	0.869	0.39	2.422	0.84	0.37	2.18
20	0.938	0.485	2.996	0.915	0.44	2.466
24	0.974	0.545	3.15	0.918	0.445	2.10
28	0.989	0.655	3.90	0.952	0.500	2.275

**Table 4.2.3b: Relative water absorption and diffusivity of Pusa 1053 at soil water potential of –0.2 MPa**

$t_g = 58$  h;  $M_g = 0.271$  g/seed

$t_g = 60$  h;  $M_g = 0.268$  g/seed

Time (h)	Treated			Control		
	$M_t/M_g$	$(Dt/a^2)^{1/2}$	$D \times 10^{-5}$ $\text{cm}^2 \text{min}^{-1}$	$M_t/M_g$	$(Dt/a^2)^{1/2}$	$D \times 10^{-5}$ $\text{cm}^2 \text{min}^{-1}$
2	0.242	0.075	0.717	0.199	0.06	0.459
4	0.345	0.115	0.842	0.302	0.950	0.575
6	0.470	0.185	1.453	0.437	0.150	0.956
10	0.587	0.220	1.233	0.517	0.175	0.780
16	0.701	0.275	1.204	0.606	0.225	0.806
20	0.895	0.420	2.247	0.852	0.375	1.792
24	0.923	0.450	2.15	0.880	0.405	1.741
28	0.917	0.445	1.802	0.899	0.420	1.605
32	0.932	0.465	1.722	0.914	0.44	1.541
38	0.938	0.485	1.577	0.939	0.46	1.420
48	0.958	0.49	1.275	0.947	0.495	1.300
56	0.972	0.545	1.351	0.967	0.535	1.302

**Table 4.2.4a: Relative water absorption and diffusivity of Pusa 256 at soil water potential of –0.1 MPa**

**t<sub>g</sub> = 28 h; M<sub>g</sub> = 0.26 g/seed**

**t<sub>g</sub> = 30 h; M<sub>g</sub> = 0.255 g/seed**

Time (h)	Treated			Control		
	M <sub>t</sub> /M <sub>g</sub>	(Dt/a <sup>2</sup> ) <sup>1/2</sup>	Dx10 <sup>-5</sup> cm <sup>2</sup> min <sup>-1</sup>	M <sub>t</sub> /M <sub>g</sub>	(Dt/a <sup>2</sup> ) <sup>1/2</sup>	Dx10 <sup>-5</sup> cm <sup>2</sup> min <sup>-1</sup>
4	0.191	0.05	0.159	0.159	0.045	0.129
10	0.556	0.195	0.969	0.514	0.175	0.780
16	0.768	0.31	1.531	0.730	0.285	1.294
24	0.964	0.53	2.983	0.948	0.485	2.498
26	0.989	0.655	4.205	0.958	0.500	2.450
28				0.965	0.530	2.557

**Table 4.2.4b: Relative water absorption and diffusivity of Pusa 256 at soil water potential of –0.2 MPa**

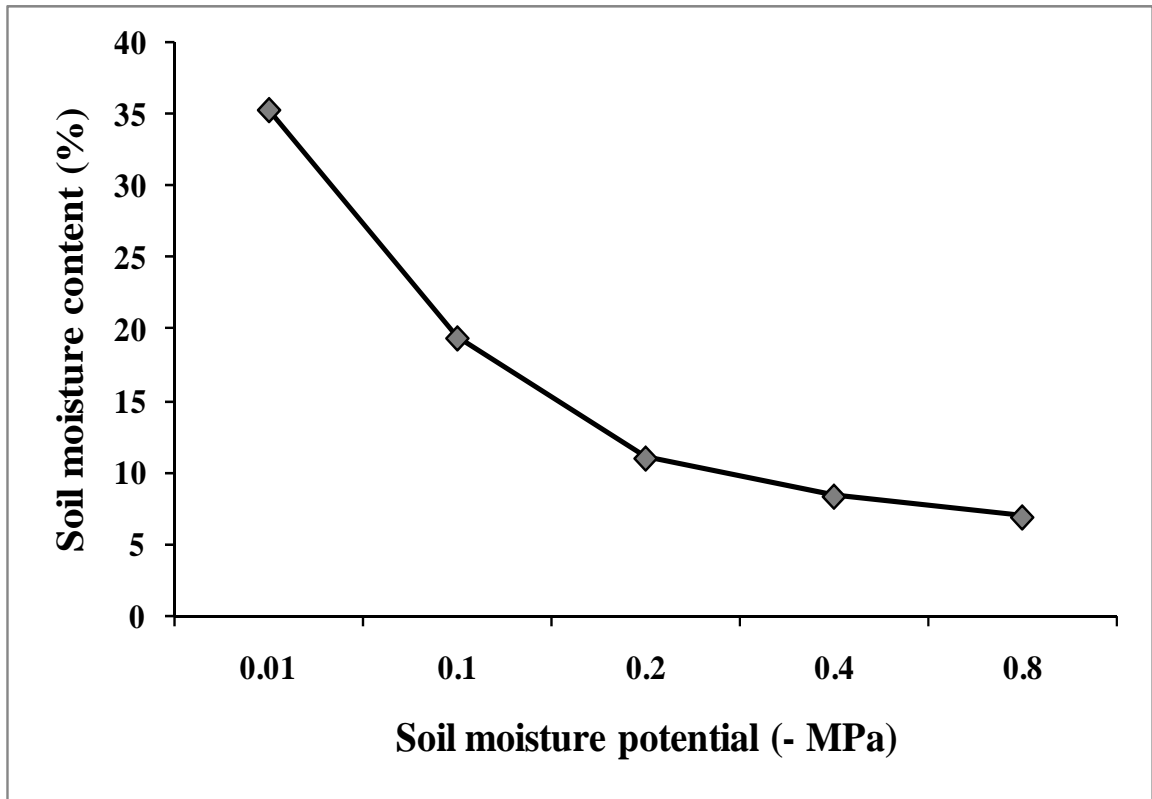
**t<sub>g</sub> = 56 h; M<sub>g</sub> = 0.265 g/seed**

**t<sub>g</sub> = 58 h; M<sub>g</sub> = 0.246 g/seed**

Time(h)	Treated			Control		
	M <sub>t</sub> /M <sub>g</sub>	(Dt/a <sup>2</sup> ) <sup>1/2</sup>	Dx10 <sup>-5</sup> cm <sup>2</sup> min <sup>-1</sup>	M <sub>t</sub> /M <sub>g</sub>	(Dt/a <sup>2</sup> ) <sup>1/2</sup>	Dx10 <sup>-5</sup> cm <sup>2</sup> min <sup>-1</sup>
4	0.197	0.055	0.193	0.196	0.055	0.193
10	0.347	0.115	0.337	0.300	0.095	0.230
16	0.478	0.155	0.383	0.396	0.135	0.290
24	0.649	0.245	0.637	0.581	0.215	0.491
28	0.684	0.265	0.639	0.671	0.252	0.578
34	0.750	0.300	0.675	0.736	0.290	0.630
44	0.793	0.330	0.631	0.772	0.315	0.575
52	0.880	0.405	0.804	0.866	0.385	0.726
56				0.967	0.535	1.302

**Table 4.5: Effect of magnetic treatment and soil moisture stress on yield components, RUE and WUE of two varieties of chickpea**

Variety	Soil moisture potential	Magnetic treatment	Biomass/pot (g)	Pod number/pot	Grain weight/pot (g)	RUE	WUE
Pusa 1053	-0.1 MPa	Control	17.85	18.6	5.344	0.933	1.257
		Treated	18.32	20.2	5.608	0.613	1.051
	-0.2 MPa	Control	4.62	5.2	1.425	0.185	0.485
		Treated	5.88	6.5	1.854	0.276	0.787
CD at 5% level			1.171	1.45	0.849		
Pusa 256	-0.1 MPa	Control	18.91	16.2	5.923	0.764	1.137
		Treated	28.08	35.6	13.954	1.461	1.984
	-0.2 MPa	Control	5.21	3.4	0.878	0.064	0.201
		Treated	7.27	6.6	2.376	0.084	0.271
CD at 5% level			1.347	5.577	1.030		



**Appendix-I: Relation between soil moisture potential (MPa) and soil water content (%)**

# INTRODUCTION

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Considering the ever-increasing global population and associated food crisis, modern agriculture is being more and more inclined towards productive growth of cultivated plants. Rainfed areas, which cover almost 70% of the total area under agriculture, would have a greater share in meeting the future food needs of the country, especially those of food legumes, which are major sources of protein for the Indian population. Chickpea (*Cicer arietinum* L.) is one of the world's most important but less-studied leguminous food crop with nearly 10 mha grown across Americas, the Mediterranean basin, East Africa, the middle east, Asia and Australia (FAOSTAT, 2004).

In India, chickpea occupies the first position among pulses, occupying about 35% of total cultivated area of pulses and contributing 45% towards total pulse production. At present global productivity of chickpea stands at 0.7 to 0.8 tonnes per hectare. The mean yield for the primary zone is  $0.84 \text{ t ha}^{-1}$ , which decreases to  $0.73 \text{ t ha}^{-1}$  in the tertiary zone. Most of the chickpea is grown during the post rainy season on stored soil moisture with one pre-sowing irrigation in northern and central India, where the rainfall during the rainy season is low to high. Hence, the crop must possess some tolerance to moisture stress as it has to mature under receding soil moisture conditions. When large number of Ethiopian land races were evaluated for drought tolerance, it was found that the tolerant genotypes produced more root weight, root volume and rooting depth compared to susceptible chickpea (Anbessa and Bejiga, 2004). Serraj *et al.* (2004) also suggested that deep and prolific root systems are associated with enhanced tolerance to drought in chickpea. Therefore, root traits are likely to be one of the most important components of drought tolerance in chickpea.

The uptake of water by seeds is an essential initial step towards germination. The movement of water from soil to seed during imbibition is governed by several factors, but particularly important is the water relations of the seed and of the soil. Therefore, a study of soil water and seed water diffusivities becomes relevant under moisture limiting conditions. Adequate water absorption from soil is the basic pre-requisite to initiate metabolic processes involved in the germination of seeds and its

subsequent growth into a plant. Under restricted water absorption, seeds may take longer time for germination and the seedling may lack uniformity. One of the most important parameters affecting seed germination is water diffusivity. Therefore, it is essential to study the water diffusivity under restricted moisture conditions to understand the behavior of drought tolerant genotypes (Ponkia *et al.*, 1991). So the study of seed water diffusivity, root characteristics as well as different physiological traits, plant water relations, water and energy use efficiencies under different soil moisture regimes give better understanding of survival behavior of the plant under stress conditions. Any treatment that has a stimulated effect on these characteristics can result in better growth and higher yield.

In the present day agriculture, the rational use of agricultural land is emphasized and greater importance is given to some physical methods of the pre-sowing treatment of seeds, which are commonly regarded as being friendlier to the environment. These physical treatments (energies) often modify the course of some physiological and biochemical processes in the seeds, which increases their vigour and contributes to the improved development of the plants. Magnetic field is especially worth our attention since its impact on the seeds can change the processes taking place in the seed and stimulate plant development. This technique has numerous practical applications in modern agriculture.

1. Increased germination rate and enhanced seedling growth.
2. Decrease seed rate per hectare by increasing the germination percentage.
3. Environment friendly: No environment effect.
4. In low viability seeds the biostimulation of magnetic field can ameliorate the deteriorating effects of storages. Hence costly seeds can be salvaged.
5. This treatment may provide an earlier ripening of the harvest.

Seeds are a resting system of organs of a future plant. What the plant will be and what results we will get depend upon the quality of the seed. Magnetic treatment of seeds is necessary while using the non-standard seeds, for the improvement of seed quality, their germination properties, and for the stimulation of growth during vegetative period.

Earlier experiments conducted in our laboratory clearly showed a significant increase in germination and field emergence characteristics of chickpea (var. Pusa 1053) when exposed to static magnetic field strengths of 100 mT and 150 mT for 1h

and 2h duration respectively (Vashisth and Nagarajan, 2008b). Also, importantly, a two fold increase in root length and root surface area was observed in one month old plants raised from magnetically exposed seeds.

Whether, this initial boost in vigour observed due to magnetic field treatment will sustain till the harvest and result in higher yield gains is not known. Also, the role of seed water diffusivity, root growth dynamics and the plant water relations and their influence on energy and water use efficiency under different soil moisture regimes need to be elucidated to understand the action of magnetic field on dry seeds.

Therefore, the present study is planned to characterize the effect of magnetic field on chickpea seeds with the following objectives:

1. To study the seed water status and seed water diffusivity under different soil water potentials in magnetically treated and un-treated seeds.
2. To evaluate the root growth dynamics and root characteristics in relation to soil water contents.
3. To study the biophysical, physiological parameters, energy and water use efficiency of the plants under different soil water potentials and their relationship with yield components.

# REVIEW OF LITERATURE

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### 2.1. Seed Germination, Seedling growth and Yield

The effect of magnetic field on living system, particularly the effect on germination of seeds and growth of the plants has been the object of numerous researches. The first study in this field was conducted by Savostin (1930) who observed increases in the rate of elongation of wheat seedlings under magnetic conditions. Later, Murphy (1942) reported changes in seed germination due to magnetic field, Akoyunoglou (1964) reported that the activities of some enzymes were increased by exposure to magnetic field. Pittman (1965) reported that speed of germination and seedling growth of corn (*Zea mays* L.) and beans (*Phaseolus vulgaris* L.) were affected by pre-germination exposure of the dry seed to an introduced magnetic field. Duration of pre-germination exposure as well as temperature and seed orientation during germination affected total visible seedling growth. Pittman and Ormrod (1970) reported that seeds of winter wheat magnetically treated before germination respired more slowly, released less heat energy, and grew faster than untreated seed. They also reported that these seedlings absorbed more moisture and contained more reducing sugar than untreated seed. Bhatnagar and Deb (1977) conducted experiments on germination and early growth and reported that pre germination exposure of wheat seeds (Cv, Sonalika) increased the rate of germination, shoot length, maximum root length and total root length significantly. Bhatnagar and Deb (1978) reported improved seedling vigour due to pre-germination exposure of wheat seeds to magnetic fields of 50 to 300 mT and found higher respiratory quotient and alpha amylase activity as compared to control seeds. Pittman (1977) reported that pre-seeding magnetic treatment of barley (*Hordeum vulgare* L.) seed resulted in seed yield increases in 13 of 19 field tests. Similarly treatment of spring and winter wheat seed (*Triticum aestivum* L.) resulted in yield increases in 14 of 23 tests. Otas (*Avena sativa* L.) showed no yield response to magnetic treatment of the seed. Lebedev *et al.* (1977) reported that exposure to a magnetic field increased photochemical activities in a unit of chlorophyll molecule, resulting in an increase in the green pigment of wheat and bean. Gubbels *et al.* (1982) observed that seed lots of flax (*Linum*



*usitatissimum* L.), buckwheat (*Fagopyrum esculentum* Moench.), sunflower (*Helianthus annuus* L.) and field pea (*Pisum sativum* L.) exposed to magnetic field produced earlier and more vigorous seedlings in some seed lots and increased the yield of sunflower. Kavi (1983) observed that by subjecting the ragi (*Eleusine coracana* Gaertn.) seed to magnetic field, its internal potential energy changed and this could be used for higher yield by fixing the strength of magnetic field and exposure time of seeds suitably. Dayal and Singh (1986) reported that the treated 'Pusa Ruby' plants on an average produced 1.76 more branches/plant at early stages and 1.48 more branches/plant at later stages as compared with control. The increase was maximum in treatment of 125 mT for 15 min. 'Pusa Early Dwarf' produced more branches than the control only at early stage. The increase was noticed in magnetic field treatments of 50, 90 and 155 mT for 30 min. Kato (1988) observed that the rate of growth of primary roots of maize exposed to 500 mT was increased by about 25% over that of control. Saktheeswari and Subrahmanyam (1989) reported that there was an increase in the number of parenchymatous cells in the root and leaf of paddy, the root hairs were more in number and increased cell division. They also reported increase in the uptake of calcium ions, which may be responsible for proper root development and prevention of chlorosis of leaves and for enhancement of chlorophyll contents. Increase in the amount of chlorophyll a, b and total chlorophyll contents of the primary and secondary leaves, both after 10 and 14 days were observed when fresh paddy seeds exposed to pulsed magnetic field. Pietruszewski (1993) reported a positive effect of magnetic field on yield of wheat cultivars. Palov *et al.* (1994) noted that presowing magnetic treatment in cotton increased yield by 6.3% and fibre length by 9.4%. Kiranmai (1994) studied the mutagenic effect of magnetic field was in two varieties of *Helianthus annuus* L. exposed to 100, 200 and 300 mT for over 90 min. He found that among the three doses studied, 200 mT produced positive mutations in both varieties of sunflower. Alexander and Doijode (1995) found that onion and rice seeds exposed to a weak electromagnetic field for 12h showed significantly increased germination, shoot and root length of seedlings. Celestino *et al.* (2000) have reported enhanced sprouting rate, main shoot length, axillary shoot formation, fresh and dry weights of the emerged shoot of *Quercus suber* seedlings when exposed to chronic EM field. Martinez *et al.* (2000) reported the influence of a stationary magnetic field on the initial stages of barley plant

development. When germinating barley seeds were subjected to a magnetic field of 125 mT for different times (1, 10, 20 and 60 min, 24 h, and chronic exposure), increases in length and weight were observed. Maximum increases in the measured parameters were obtained when the time of exposure to magnetic field was long (24 h and chronic). Harichand *et al.* (2002) reported that field trial (100 gauss) with 40h exposure resulted in increase in plant height, seed weight per spike and yield of wheat. Anna (2002) observed that the magnetic field stimulated the shoot development of maize and led to the increase of the germinating energy, germination, fresh weight and shoot length. Podlesny *et al.* (2004, 2005) confirmed the positive effect of the magnetic treatment on the germination and emergence of both broad bean and pea cultivars. The magnetic stimulation of seeds favorably influenced the sprouting and emergence of seed. As a result of the application of this treatment plant emergence was more uniform and took place 2-3 days earlier than the emergence of plant in the control. The gain in seed yield resulting from the pre-sowing treatment of seeds with a magnetic field for both broad bean and pea was due to the higher number of pods per plant and the fewer plant losses in the unit area in the growing season. No significant differences were found in the course of most developmental phases of those plants grown from the treated and non-treated seeds. However, a few days acceleration was reported concerning the maturity of plants obtained from those seeds pre-treated magnetically in comparison to the control. Galland and Pazur, (2005) reported the unsystematic manner in which the research on magnetoreception in biology has been carried out in the past and explains presently accepted mechanisms of magnetoreception. Rajendra *et al.* (2005) have observed significant increase in mitotic index as well as  $^3\text{H}$ -thymidine incorporation into DNA in seeds of *Vicia faba* exposed to 100  $\mu\text{T}$  power frequency electromagnetic field. These are clear indications of enhancement of growth of germinated seedlings exposed to magnetic field. Florez *et al.* (2007) reported that exposure of maize seed to stationary magnetic field enhanced the germination and early growth of seedlings. The greatest increases were obtained for plants continuously exposed to 125 or 250mT. It has been established that the rate of the plant growths (chickpeas, beans and lentils) is enhanced by SMF that is intimately related to environmental temperature, when other environmental parameters (humidity, illumination, soil chemical state, etc) being kept under control (Akif, 2007).

## 2.2. Studies on rooting characteristics

Quantification of root growth is necessary to study water and nutrient dynamics in rhizosphere. It forms an important part of models of soil–plant interaction required for management guidance (Hanson *et al.*, 1999). Root characteristics such as mass, length, average diameter, surface area and volume were used to assess the quality of roots and functional size of the root system. Total root mass being easier to measure than root length and surface area, was used frequently to compare root systems (Murphy and Smucker, 1995). However, later on it was seen that root mass alone could not describe many root functions involved in studying plant-soil relationships such as uptake of water and nutrient by roots from soil. It was finer roots with larger length density and surface area which contributed to more water and nutrient uptake from surface as well subsurface soil than the thicker roots, which mainly contribute to the root mass and remained confined to upper surface layer only. Hence for studying soil-water-plant relationship it is better to use root length density rather than root weight density (Brewster and Tinker, 1970; Raper *et al.*, 1978; Fiscus, 1981 and Box and Ramseur, 1993).

Root growth systems are hierarchical and each lower order class of root members has lower root diameter, less mass per unit length. Thus much of plants root length is found in finer and smaller members of root systems. Fiscus (1981) reported that two third of total area of *Phaseolus vulgaris* L. root had an average diameter of 0.5mm. The use of clear plastic tubes, known as minirhizotrones along with micro video system installed in the field made effective nondestructive measurement of finer (more active and more fragile) root system possible (Merrill *et al.*, 2002).

Computer assisted electronic image analysis have made root analysis less time consuming and allowed more accurate and less subjective measurement of root characteristics than the human eye is capable of making (Merrill *et al.*, 2002). Electronic methods acquire images through video camera or optical scanner. Fine roots were underestimated when roots were measured using image analysis as they could not be successfully detected due to their small diameter and near transparency (Burke and LeBlanc, 1988; Pan and Bolton, 1991; Murphy and Smucker, 1995). Such roots account for a substantial proportion of total root length in a number of species (Merrill *et al.*, 2002).

Improvement of lighting source and technical development in scanning technology and development of image analysis software made root length measurement easy and accurate. One such image analysis software developed by RHIZO (Regent instruments, Quebec) was tested for its accuracy in measuring root length for samples in which root overlap and also for measuring the distribution of length in different diameter classes (Bauhus and Messier, 1999).

### **2.3. Studies on diffusivity**

#### **2.3.1. Water in seeds**

Water is the major constituent of cells in all-living organisms, since most biochemical reactions proceed in aqueous solutions. The dynamic state of water affects many physiological phenomena. Water is also an important substrate in many reactions (Buitnik *et al.*, 1998). Its removal can lead to reduced activities due to low substrate concentrations. The most important function is the role of water as the solvent for many biochemical reactions. Loss of water as solvent will reduce the diffusion rate of solute substrates to an active site (Leopold and Vertucci, 1989). Water also affects the intermolecular motions of proteins that are essential for catalytic activities. In plants, water is present in all tissues including the seed propagules. In order to germinate, seeds imbibe water from the surrounding environment (Chai *et al.*, 1998).

#### **2.3.2. Biophysical characterization of water in seed**

Seed water status refers to the measurement of state of water in relation to seed and it is used in a relative sense (Vertucci and Roos, 1990, 1993; Vertucci *et al.*, 1994). Water status can be described either by measuring the moisture levels of tissue water content or by measuring the energy status of cell water. Moisture levels of seeds are the important determinant of seed longevity (reviewed by Cromarty *et al.*, 1985). Moisture levels in seeds can be described in several ways: moisture content (mc), water activity ( $a_w$ ) and chemical potential ( $\mu_w$ ). While related, these parameters have very different thermodynamic implications.

### Water / Moisture content

This simply is a measure of the concentration of water in the seed. Tissue water content is generally expressed as a percentage of fresh weight or dry weight of the tissue. Often this quantity is expressed as the amount of water per unit fresh weight of the tissue (Vertucci, 1994).

### Water activity

This specifies the relative purity of water, measures how many times as effective as water is at promoting aqueous reaction at a given mc compared with a standard reference state (usually, pure liquid water). Water activity describes the tendency for a chemical reaction involving water at a given temperature; it is always  $\leq 1$  (Ellis *et al.*, 1989; Roberts and Ellis, 1989; Ellis *et al.*, 1990; Vertucci and Ross, 1990).

$$a_w = (p_o \gamma_o / p \gamma) \cong RH/100 \quad \text{as } RH = p_o/p \times 100 \text{ and } \gamma_o \approx 1$$

where  $p_o$  is the water vapour pressure,  $p$  is the total pressure of the system and  $\gamma$  is the fugacity coefficient. Thus,  $RH/100$  is the good approximation of water activity of the solution. Water activity of seeds is an intrinsic property, related to the composition, water content and temperature (Walters, 1998).

### Chemical potential of water ( $\mu_w$ )

Chemical potential is viewed as the potential for chemical or physical change (Vertucci and Roos, 1993). The chemical concerned is water and so its chemical potential is referred as the chemical potential of water. Chemical potential of water is the components of free energy ( $G$ ) of a system.

$$\begin{aligned} \mu_w (\text{seeds}) &= \mu_w^o + RT \ln (a_w) \\ &\cong \mu_w^o + RT \ln (RH/100) \end{aligned}$$

$\mu_w^o$  is the chemical potential of the standard i.e. pure liquid water

### Water potential

The parameters "water potential" ( $\Psi_w$ ) is basic to the study of water relations in plants. Water potential is actually a measure of pressure, which can be derived from the chemical potential (energy) (Walters *et al.*, 1997). By convention,  $\Psi_w$  is calculated

from  $\mu_w$  by dividing by molar volume of water ( $v$ ) at 20°C and 1 atm (18.048 ml/mole) and by setting the value of  $\mu_w^0 / v = 0$ . Thus, the water potential can be calculated as

$$\psi_w = RT \ln (p\gamma/p\gamma_0) / v$$

In general, water activity and the chemical potential of water are believed to be the relevant parameters to evaluate the role of water in deteriorative reactions (Ellis *et al.*, 1989, Roberts and Ellis, 1989; Ellis *et al.*, 1990; Vertucci and Roos, 1990; 1993; Vertucci *et al.*, 1994; Walters, 1998).

### 2.3.3. Physical status of water in seed

Discrete changes in metabolic activities with moisture content to be associated with discrete changes in the physical status of water in seeds (Vertucci and Farrant, 1995). At least, five types of water in seeds are distinguished from the calorimetric and the motional properties (Myers *et al.*, 1992).

- (i) At moisture level below 8% (-150 MPa) water association is in ionic sites.
- (ii) When moisture content is between 8-25% (-150 to -11 MPa), water is in association with hydrophilic sites and catabolic activity starts.
- (iii) At moisture levels between 24-45%, the catabolic activities continue unabated and processes utilizing the high-energy intermediates are impaired.
- (iv) At about 45% (-3 MPa) protein synthesis ceases and repair processes become inoperative.
- (v) When the moisture level are below -1.5 MPa (>70%), tissues no longer grow or expand.

### 2.3.4. Imbibition kinetics: diffusivity

#### Uptake of water from soil

The uptake of water by the seeds is an essential, initial step toward germination. The total amount of water taken up during imbibition is generally quite

small and may not exceed two or three times the dry weight of the seed. For subsequent seedling growth, which involves the establishment of the root and shoots systems, a larger and more sustained supply of water is required.

Several factors that govern the movement of water from soil to the seed, but particularly important are the water relations of the seed and of the soil. Water potential ( $\Psi$ ) is an expression of the energy status of water, net diffusion of water occurs down an energy gradient from high to low potential (i.e., from pure water to water containing solutes). Pure water has the highest potential, and by convention, it is assigned a zero value. Other potentials, therefore have positive (i.e.,  $>0$ ) or, negative (i.e.,  $<0$ ). The water potential of the cell in a seed can be expressed as follows:

$$\Psi_{\text{cell}} = \Psi_{\pi} + \Psi_{\text{m}} + \Psi_{\text{p}}$$

Where,

$\Psi_{\text{cell}}$  = water potential of the cell

$\Psi_{\pi}$  = osmotic potential

$\Psi_{\text{m}}$  = matric potential

$\Psi_{\text{p}}$  = pressure potential

This means that cell water potential is affected by three components:

- 1)  $\Psi_{\pi}$  - The osmotic potential, the concentration of dissolved solutes in the cell determines the osmotic potential- the greater their concentration, the lower is the osmotic potential and hence the greater the energy gradient along which water will flow. Thus, the concentration of solutes in the solutes in the cell influences water uptake.
- 2)  $\Psi_{\text{m}}$  - The matric potential, this is contributed by the hydration of matrices (eg., cell walls, starch, protein bodies) and their abilities to bind water.
- 3)  $\Psi_{\text{p}}$ -The pressure potential, which occurs because as water enters a cell the internal pressure builds up which exerts a force on the cell wall. Values for  $\Psi_{\pi}$  and  $\Psi_{\text{m}}$  are negative since they have a lower potential than pure water, and  $\Psi_{\text{p}}$  is a positive and hence opposite force. The sum of the three terms, the water

potential, is a negative number, except in fully turgid condition where it approaches zero.

The difference in water potential between seed and soil is one of the factors that determines availability and rate of flow of water to the seed. Initially, the difference in  $\Psi$  between the dry seed and moist soil is very large because of the high  $\Psi_m$  of the dry coats, cell walls, and storage materials. But as the seed moisture content increases during imbibitions and the matrices become hydrated, the water potential of the seed increases and that of the surrounding decreases as water is withdrawn. Hence, the rate of water transfer from soil to seed decreases with time, more quickly in soil of low water holding capacity. Continued availabilities of water to the seed depend on the rate at which water moves through the soil i.e., the hydraulic conductivity of the soil.

### **Water uptake by seeds**

Under optimal conditions of uptake of water by seeds is triphasic

Phase 1 : The water potential of a mature dry seed is much lower than that of the surrounding moist substrate and can exceed - 100 MPa because of its high  $\Psi_m$ . Water uptake in phase I, or imbibition, is largely a consequence of these matric forces, and water uptake occurs regardless of whether the seed is dormant or non-dormant, viable or nonviable. A wetting front is formed as water permeates the seeds, and there is an abrupt boundary of water content between wetted cells and those to be wetted. Moreover, the average water content of the wetted area increases as a function of time. This initial pattern of water uptake is thus marked by three characteristics.

1. A sharp front separating wet and dry portion of the seed
2. Continued swelling as water reaches new regions and
3. An increase in water content of the wetted area

Phase 2 - this is a lag phase of water uptake when the  $\Psi_m$  no longer play a significant role, and the  $\Psi$  of the seed is largely a balance between  $\Psi_{\Pi}$  and  $\Psi_p$ . In this phase, the value of  $\Psi$  for many seeds probably does not exceed -1 to -1.5 MPa. During this phase major metabolic activities take place in preparation for radicle emergence from non-dormant seeds; dormant seeds are also metabolically active at this time.



Phase 3 - Although dormant seeds may achieve phase 2, only germinating seeds enter this third phase, which is concurrent with radicle elongation. The increase in water uptake is initially related to the changes that cells of the radicle under go as they extend, marking the completion of germination. Then water uptake is influenced by decrease in  $\Psi_{\Pi}$  resulting from production of low molecular weight, osmotically active substances resulting from the post germinative hydrolysis of stored reserves.

Water may be directed preferentially toward the radicle of the maize embryos during early imbibition. There are structural modifications to the coat adjacent to the radicle, which facilitate this. Water then diffuses to the embryos. Hydration of endosperm is slower because water has to penetrate the surrounding pericarp, which is not structurally modified to permit its rapid uptake.

### **Factors affecting imbibition**

Vertucci and Leopold (1983) reported that water uptake rates were slowed by low temperature and low initial moisture content of the tissue. The role of water viscosity in the temperature effects on imbibition was examined, and a linear relation between imbibition rate and the reciprocal of viscosity was found only for seeds of very high initial moisture content. The data are interpreted as indicating a first component of water entry which is a wetting reaction influenced by the surface tension of the water, and or second component which resembles water flow through a porous matrix and is influenced by the water viscosity.

The seeds responded differently to reduced  $\psi_m$  (matric potential), so did the different germination parameters. Plumule emergence is generally more sensitive to  $\psi_m$  than radicle emergence. The response of plumule elongation to moisture tension seems to be the most critical of all. The threshold value for plumule elongation was  $\psi_m = -7$  bar in wheat,  $-13$  bar in barley and  $-10$  bar in sorghum (El-Sharkawi and Springuel, 1977).

Water uptake patterns and germination rates of chickpea (*Cicer arietinum* L.) and vetch (*Vicia faba* L.) as affected by constant and changing external water potential, were studied experimentally by Hadas (1976). The initial water uptake rate was found to decrease as the external water potential decreased, due to reduced diffusivity to water of the seed coats.

Many workers have studied the seed water uptake and diffusivities of germinating gram, cotton, soybean, corn and cowpea and groundnut (Philips, 1968; Chatterjee *et al.*, 1981; Chatterjee *et al.*, 1982; Ponkia *et al.*, 1991). They reported differences both at species and cultivars levels. Application of diffusion models, unlike empirical relationships has enhanced our capacity to realistically describe the physical processes of water absorption by germinating seed (Collis-George and Melville, 1975; Chatterjee *et al.*, 1981).

#### **2.4. Studies on photosynthesis, WUE, RUE, osmotic potential**

Osmotic adjustment is considered as an important physiological mechanism of drought adaptation in many plants (Subbarao *et al.*, 2000) and particularly in chickpea cultivars at lower osmotic potential (Chopra *et al.*, 1995, Leoport *et al.*, 1998). Various studies has shown that in chickpea subjected to terminal drought that leaf photosynthesis is markedly decreased as the soil dries and the leaf water potential decreases.

Chickpea yield, intercepted radiation and the number of pods per plant decreased linearly as the maximum potential soil moisture deficit ( $D_{p_{max}}$ ) increased. The study has shown that using actual evapotranspiration and water-use efficiency, the biomass yield and seed yield of Kabuli chickpeas can be accurately predicted in Canterbury. Soil water shortage has been identified as a major constraint to increasing chickpea production. Drought was quantified using the concept of maximum potential soil moisture deficit ( $D_{p_{max}}$ ) calculated from climate data. Drought responses of yield, phenology, radiation use efficiency and yield components were determined, and were highly correlated with  $D_{p_{max}}$ . The maximum potential soil moisture deficit increased from about 62 mm (irrigated throughout) to about 358 mm (dryland plots). There was a significant correlation ( $P < 0.001$ ) between water use and biomass yield ( $R^2 = 0.80$ ) and water use and seed yield ( $R^2 = 0.75$ ). (Anwar *et al.*, 2003).

Singh and Bhushan (1980) found that addition of P to chickpea (*Cicer arietinum* L.) increased yield, water use, and WUE. The increase in WUE was from 8.5 to 12.2 kg ha<sup>-1</sup> mm<sup>-1</sup> at 0 and 100 kg P ha<sup>-1</sup> respectively. This gain was due to a greater depletion of soil water with fertilizer and a yield increase. Zhang *et al.*, (2000) examined water use and water-use efficiency of chickpea and lentil from 3 experiments over 12 seasons, 1986–87 to 1997–98, in northern Syria. The strongest

determinant of grain yield of chickpea and lentil and their water use under rainfed conditions is rainfall and its distribution. Both the average water use efficiency and potential transpiration efficiency for lentil and chickpea were lower than those for cereals. Lower water use efficiency was associated with low seed yield in this study. Chickpea may adapt to drought stress by maximizing its water uptake through continuous root growth up to seed-filling (Brown *et al.*, 1989) and by maintaining substantial water uptake until the fraction of extractable moisture in the root profile falls to 0.4 (Keatinge and Cooper, 1983; Siddique and Sedgley, 1987). Tanner and Sinclair (1983) suggested that semiarid region may have the most potential for improvement in WUE because the water vapor gradient between plants and the atmosphere is small and evaporation rates may be reduced.

# MATERIALS AND METHODS

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In order to achieve the objectives of the present study, experiments were carried out in the Indian Agricultural Research Institute, New Delhi. Keeping in view the basic objective of effect of magnetic field on root dynamics and growth and yield of both Desi and Kabuli type chickpea varieties under controlled moisture stress conditions, the experiments planned and carried out are given below under different headings.

### 3.1. SELECTION OF SEED

Breeder seed of chickpea (Kabuli var. Pusa 1053 and Desi var. Pusa 256) was obtained from Division of Genetics, Indian Agricultural Research Institute, New Delhi. Seeds without visible defects, insect damage and malformation were selected and stored in the desiccators having anhydrous calcium chloride.

### 3.2. SOIL USED

To study the first objective, different soil moisture regimes were created. For this, the surface soil (0-30 cm) was collected from IARI farm. The air-dried soil was sieved through 2mm size sieve and used as the experimental material. The physical and physico chemical properties of the soil are given below:-

Properties	Values
1. Mechanical Analysis	
a) Sand	62.5%
b) Silt	25.8 %
c) Clay	11.7%
2. Textural class.	Sandy loam
3. pH	7.9
4. Electrical Conductivity	0.51 dSm <sup>-1</sup>

### 3.3. INSTRUMENT USED

#### 3.3.1. Pressure plate apparatus

A pressure plate apparatus “NORGREN” from Soil Moisture Equipment Corp., Santa Barbara, CA, USA was used to obtain the soil moisture characteristic

curve which gives the information of soil moisture content at different matric potentials.

### 3.3.2. Electromagnetic field generator

An electromagnetic field generator “Testron EM-20” with variable magnetic field strength (50 to 500mT) was used to treat the chickpea seeds. A DC power supply (80V/ 10A) with continuously variable output current was used for the electromagnet. A digital gauss meter model DGM-30 operating on the principle of Hall effect monitored the field strength produced in the pole gap. The probe is made of Indium Arsenide crystal and is encapsulated to a non- magnetic sheet of 5mmX4mmX1mm and could measure 0-2 Tesla with full-scale range in increments of 5mT.

### 3.3.3. Rotronic Hygrometer / Water Activity Meter

**Definition:** Hygroscopic products may absorb water in different ways: sorption with formation of hydrate, binding with surface energy, diffusion of water molecules in the material structure, capillary condensation, formation of a solution, etc. Depending on the absorption process, water is bound to the product with more or less strength. Water content can include both an immobilized part (e.g. water of hydration) and an active part.

Water activity,  $a_w$  measures the vapor pressure generated by the moisture present in a hygroscopic product.

$A_w = p/p_s$  and % ERH =  $100 \times a_w$ , where

$p$  = Partial pressure of water vapor at the surface of the product

$p_s$  = Saturation pressure or partial pressure of water vapor above pure water at the product temperature

Water activity reflects the active part of moisture content or the part which, under normal circumstances, can be exchanged between the product and the environment.

**Applications:** The active part of moisture content and therefore water activity provide better information than the total moisture content regarding micro-biological, chemical and enzymatic stability of perishable products such as foods and seeds.

**Measurement:** The Rotronic HygroLab is a bench-top laboratory humidity temperature indicator that can be used with a wide variety of probes to meet specific application requirements. The standard method of measuring water activity consists in placing a sample of the product to be measured in a sealed container. The product sample slowly exchanges moisture with the air inside the sealed container until

equilibrium is reached. The equilibration process is monitored by measuring the humidity of the air above the product with a relative humidity sensor ( $\%RH=100XA_w$ ). Because temperature is an important factor when measuring water activity, the temperature of the air above the product is also monitored. By definition, water activity is equal to  $\%RH/100$  when equilibrium has been reached. At that time, the product no longer interchanges moisture with the surrounding air.

#### **3.3.4. Root Scanner**

Scanning and image analysis for root characteristics was carried out using Root Scanner (LA 1600) and the root morphology and Architecture measurements (total root length, root surface area, root thickness and root volume) were done by win RHIZO program from REGENT INSTRUMENTS Inc. Canada. The scanner of RHIZO system had two light sources, one below the scanner glass called flat bed or reflective and one above it (in the scanner cover) called transparency unit or TPU. For root morphological measurements TPU lighting system was used for scanning. The light rays from TPU passed through the sample and then to the camera sensor below the scanner glass. The image analysis system RHIZO acquires a direct digital (grayscale) image from the scanner and then created two other types of images for its analysis. One type of the image created was termed as 'Pixels classification image' in which all parts of the original image that fell below a user defined threshold of grayscale value were removed. The image had only two intensities, black for pixels belonging to roots and white for pixels belonging to the background. Another created image was termed as skeleton image, which consisted of a line, one pixel in diameter, which is superimposed over the earlier root image. RHIZO system measured the root length by scanning the length of the root skeleton. The colour of the skeleton indicated the diameter classification. Measurements of root morphological characteristics were based on Regent's non-statistical method (Arsenault *et al.*, 1995; Guay and Arseneault, 1996) with overlap compensation. The advantage of Regent's non-statistical method over Tennant's (1975) statistical method was that in addition to total root length density, root surface area and volume measurements, it further gave information of their distribution in various size classes based on their diameter.

#### **Length**

Total length can be measured with the following formula:

Length = (number of pixels in the skeleton)\*(pixel size)

### **Projected area**

Projected area is measured by counting the number of pixels belonging to the root in the pixel classification image. The count is then multiplied by the pixel area.

Pixel area= pixel width \* pixel height

Projected area= number of pixels \* pixel area

### **Average diameter, surface area and volume**

Average diameter was calculated with the following formula:

Average diameter = Projected area / Total length

This formula is based on the assumption that roots are round.

### **3.3.5. LI-6400 Photosynthetic system**

The LI-COR (LI-6400) utilizes gas exchange principles to measure the photosynthesis rates of plants. Net photosynthesis rates are expressed as rates of CO<sub>2</sub> uptake ( $\mu\text{mol CO}_2 \text{ m}^{-2}\text{s}^{-1}$ ). Gas exchange in the LI-COR measured in an open mode design. In the open mode an air flow is moved through a controlled atmosphere surrounding a plant leaf enclosed in an assimilation chamber. Thus, the CO<sub>2</sub> level of the air is maintained steady-state (Anonymous, 1996).

The hardware of the system is the console and the leaf chamber (the sensor). The leaf chamber has tightly sealed gaskets that do not interact with H<sub>2</sub>O or CO<sub>2</sub>, nor are deformed excessively by the leaf midribs. It also houses a PAR light sensor parallel to the leaf plane, a thermocouple, and a speed variable mixing fan. The sensor head encloses a leaf surface of up to 6 cm<sup>2</sup> and has integrated sensors for monitoring light, temperature, H<sub>2</sub>O and CO<sub>2</sub> levels. More importantly, these parameters can be precisely controlled to create the environmental condition desired. For example, the temperature of the chamber block is controlled by peltier cooler to any level within  $\pm 6^{\circ}\text{C}$  of the air temperature. Light can be adjusted at any level from 0 to more than 2000  $\mu\text{mol CO}_2 \text{ m}^{-2}\text{s}^{-1}$ , and CO<sub>2</sub> from 0 to more than 2000  $\mu\text{mol CO}_2 \text{ m}^{-2}\text{s}^{-1}$  as well. Artificial light is optionally supplied by a cold lamp (LED type, 670 nm ). The selected levels of CO<sub>2</sub> are supplied by a CO<sub>2</sub> injector system. Any combination of light and CO<sub>2</sub> level can be selected while the rest of the variable are held constant. Visible light external to the chamber can also be measured with an optional sensor. One distinctive aspect of the LI-6400 is that the CO<sub>2</sub> and H<sub>2</sub>O infrared analyzer are located in the sensor head instead of the console. The minimum distance between the

leaf atmosphere and the analyzer allows fast measurements of both CO<sub>2</sub> and H<sub>2</sub>O, which can thus be displayed in real time. The two infrared analyzers, whose readings are averaged, measure the absolute CO<sub>2</sub> and H<sub>2</sub>O concentrations at both the reference flow and the sample itself. CO<sub>2</sub> and H<sub>2</sub>O are discriminated from other IR absorbing gases by making reference measurements at 4.1 microns (CO<sub>2</sub>) and 2.4 microns. Thus, the analyzers avoid background noise.

### **3.4. EXPERIMENTS & METHODOLOGY**

#### **3.4.1. Standardization of magnetic field strength for maximum enhancement of germination characteristics in Desi type chickpea variety**

The seeds of desi chickpea var. Pusa 256 were exposed to the magnetic field of 50 to 250 mT in a sample holder, cylindrical in shape and made of non-magnetic thin transparent plastic sheet. Visibly sound, mature, healthy 100 seeds were held inside plastic container in a volume between the poles of the electromagnet having uniform magnetic field for various duration ranging from 1 to 3h. The required strength of the magnetic field was obtained by regulating the current in the coils of the electromagnet. Gauss meter was used to measure the strength of the magnetic field between the poles. The field strength and duration was standardized for maximum enhancement of germination and vigour in laboratory conditions. In case of Kabuli chickpea, magnetic field and duration for maximum enhancement in seedling characters standardized by Vashisth (2007) was used.

#### **3.4.2. Studies on seed water absorption**

##### **3.4.2.1. Moisture retention characteristics of the soil**

To maintain different levels of soil water potentials, it is necessary to calculate the moisture retention at these potentials. Therefore, the soil moisture characteristic curve was drawn using pressure plate membrane apparatus (NORGREN Soil Moisture Equipment Corporation, CA, USA) following the procedure of Richard (1948) and this curve is given in Appendix-I.

##### **3.4.2.2. Preparation of soil with different moisture potentials**

The moisture content of the air-dried sample was calculated on oven dry weight basis. Then by taking a known amount of soil, calculated amount of water was added to bring the moisture content to the appropriate value as given by the soil



moisture characteristic curve to get three different soil moisture regimes, namely, -0.1, -0.2 and -0.4 MPa potentials. The soil and water were mixed thoroughly and allowed to arrive at equilibrium by keeping it undisturbed for 2 days in airtight polythene bags. The three lots of soil so prepared were used to fill up plastic pots with a square top of 10 cm length and 12 cm height. The weight of soil required to fill the pots leaving top 1 cm with a bulk density of  $1.5 \text{ g cm}^{-3}$  was calculated from the volume of the pot. Separate samples in duplicate were taken for determination of soil moisture content of the prepared soils of three different potentials.

#### **3.4.2.3. Filling up of pots and placement of seeds**

Initially 10 seeds of each cultivar were weighed separately and kept in paper envelopes. Each pot was filled with weighed amount of the soil of the required moisture content layer by layer leaving about 3 cm at the top. Then, the pre-weighed seeds were placed in a geometric pattern and pressed gently for proper seed soil contact. The remaining soil was used to cover the seeds uniformly and compressed to maintain the same depth of 1 cm from top and the pots were closed tightly to avoid moisture loss and kept at a constant temperature of  $25^{\circ}\text{C}$  in an incubator. There were three replications for each cultivar, soil moisture regime and time duration.

#### **3.4.2.4. Measurement of seed water uptake**

The time was noted after placing the seeds in each pot. After varying period of time starting from 4h, the seeds were removed and weighed on the balance after quickly removing the adhering soil particles. The difference between this weight and the initial air-dry weight gave the water absorbed by 10 seeds in that particular period. Thus the amount of water absorbed by single seed  $M(t)$  was determined at various times ( $t$ ). This was continued till the seeds germinated and the radicle of about 2-4 mm was produced. The total amount of water absorbed by the seed  $M(g)$  at the time of germination ( $t_g$ ) was noted. After determination of the weight, the seeds were quickly transferred to a water activity meter for measurement of seed water activity  $a_w$ . The average radius of the seeds was determined by finding out sphere equivalent volume of the seed of each cultivar separately. The volume was obtained by measuring the water displaced by 10 seeds using a specific gravity bottle.

### 3.4.3. Determination of seed water diffusivity

In considering the absorption of water by a seed during germination, spherical geometry was assumed where water moves in a radial direction only. The error introduced due to the seeds being not exact spheres was shown to be very small by Philips (1968).

Seed water diffusivity was computed by following the procedure outlined by Philips (1968) and following the equation:

$$\frac{M(t)}{M(t_g)} = \frac{\pi^2 / 6 - \sum_{m=1}^{\infty} (1/m)^2 \exp(-Dm^2\pi^2 t / a^2)}{\pi^2 / 6 - \sum_{m=1}^{\infty} (1/m)^2 \exp(-Dm^2\pi^2 t_g / a^2)} \quad (1)$$

Where D is the seed water diffusivity ( $\text{cm}^2 \text{min}^{-1}$ ) and 'a' is the sphere equivalent radius of the seed in cm.

The diffusivity values (D) cannot be evaluated explicitly from equation (1) and are evaluated by a graphical solution of the above equation (1). For this purpose the values of  $M(t)/M(t_g)$  along with experimentally determined t,  $t_g$ , and a values were used. The plot of  $M(t)/M(t_g)$  ie., relative water uptake as a function of time t for the germinating seeds at different soil water potentials were obtained. Graphical solution of the equation (1) attained by a computer programme (IBM 1620) by an earlier student of the division (Chatterjee, 1976) was used. A plot of  $M(t)/M(t_g)$  as a function of  $D\pi^2 t/a^2$  was obtained for several values of  $t_g/t$ .

Equation (1) cannot be used for calculating diffusivity values D for all conditions, if the  $M(t)/M(t_g)$  ratio exceeds the intercept shown in (Appendix II) for the particular  $t_g/t$  ratio under consideration. This generally happens for large values of  $t_g/t$  and consequently for small values of  $M(t)/M(t_g)$ . Under such situations, the simplified equation of Crank (1956) was used for calculation of D values.

$$\frac{M(t)}{M(t_g)} = 6 \left( \frac{Dt}{a^2} \right)^{1/2} \left\{ 0.5645 + 2 \sum_{n=1}^{\infty} \text{ierfc} \left( \frac{na}{\sqrt{Dt}} \right) \right\} - \frac{3Dt}{a^2} \quad (2)$$

The graphical representation of this equation is given in Appendix-II.

Where  $\text{ierfc}$  = integral of complementary error function. In our experiment, diffusivity constant values were calculated using Crank's equation as in most cases,  $M(t)/M(t_g)$  ratio exceeded the intercept of Philips equation.

#### **3.4.4. Seed water activity ( $a_w$ )**

In the above experiment, at different intervals of seed imbibitions, immediately after taking the fresh weight of the seeds, they were placed in the cavity of the Water activity meter (Rotronics, Switzerland) and reading was noted after the seeds attained equilibrium. All measurements were taken at a constant temperature of 25°C by circulating water from a water bath maintained at 25°C around the cavity.

#### **3.4.5. Studies on root dynamics and root growth**

Magnetically exposed and unexposed seeds of desi and kabuli were grown in sand and peat medium maintained at 20% and 12% moisture content on dry weight basis. Two moisture levels of 20% and 12% were selected for this study representing -0.1 MPa and -0.2 MPa soil moisture potentials based on soil moisture characteristic curve.

##### **3.4.5.1. Preparation of medium for root and shoot studies**

Air dried river sand and sterilized peat was mixed thoroughly in the ratio of 1:1 and the initial moisture content of the mixture was determined gravimetrically. The amount of water to be added to 10 kg soil + peat mixture to create 12 and 20% moisture content was calculated and added and was kept sealed in a polythene bag overnight for moisture equilibrium and used for filling the pots. Plastic pots of 17 cm dia. and 17.5 cm height were filled with 2 kg mixture of soil and peat (containing macro and micro plant nutrients including substantial quantities of K) at 12 or 20% moisture and weighed individually. Pre-sprouted magnetically treated and untreated seeds of both species of chickpea were planted one each in these pots and kept in the green house in a completely randomized fashion for various numbers of days. The pots were weighed on alternate days and brought back to initial weight by adding water from a hand sprayer. No additional nutrient were applied. There were four replications per treatment.

**Sampling:** Periodic samplings will be done by washing the roots in running water at 10, 20, 30 & 40 days interval.

**Parameters measured:** Total root length, Length longest root, Root surface area, Root diameter, Root distribution and Root and shoot dry weights.

#### 3.4.5.2. Rooting Characteristics

The root samples were washed carefully by gentle stream of water for complete separation of root from soils. Then the roots were air-dried so as to make the root samples ready for scanning. Scanning and image analysis for root characteristics was carried out using Root Scanner (LA 1600) and the root morphology and architecture measurements (total root length, root surface area, root thickness and root volume) were done by win RHIZO program from REGENT INSTRUMENTS Inc. Canada. There were four replications per treatment.

#### 3.4.6. Pot culture Experiment

From soil characteristic curve, the amount of water required to maintain a soil moisture potential of -0.1 MPa and -0.2 MPa in a given amount of soil with known initial moisture was calculated. Thus prepared soil with two potentials were filled in 12" earthen pots and individually weighed after adding 0.5g DAP/pot. Magnetically exposed and unexposed control seeds of both genotypes were treated with bavistin 2 g/kg of seed and were sown @ 5 per pot. Two plants were retained per pot after thinning. They were arranged in the green house in completely randomized fashion. The pots were weighed twice a week and the loss in weight from the initial value was made up by adding measured amount of water.

At flowering and at podding stages, the following parameters were measured. There were three replications for each growth stage per treatment.

##### 3.4.6.1. Leaf photosynthesis, stomatal conductance & transpiration

Net photosynthesis (Pn) was measured *in vivo* on the second mature leaf from the top using LI-6400 photosynthetic system (LICOR, USA) by giving constant light of 1000  $\mu\text{mol m}^{-2}\text{s}^{-1}$ . Air temperature during measurement was 26°C and ambient CO<sub>2</sub> was 380 ppm. In all four measurements were taken per pot and three pots per treatment was used. At the end of light period, gas exchange parameters, namely, stomatal conductance ( $g_s$ ) and transpiration (E) were recorded. Photosynthetic Water Use Efficiency was calculated as Photosynthesis ( $\mu\text{mol m}^{-2}\text{s}^{-1}$ )/Transpiration ( $\text{mmol m}^{-2}\text{s}^{-1}$ ) and expressed as  $\mu\text{mol}/\text{mmol}$ .

### 3.4.6.2. Total chlorophyll content

Fresh leaf samples from different treatments were cut and 100 mg of it was weighed accurately in an analytical balance and chlorophyll was extracted by a non macerated method equilibrating it with 10 ml DMSO (Dimethyl Sulphoxide) in a capped vial and keeping in an oven at 65°C for about 3 hours (Hiscox and Israelstam, 1979). The decanted solution was used to estimate the absorbance at 645 and 663 nm wavelength using Spectronic-20 Spectrophotometer. The total chlorophyll content was calculated using the formula given by Arnon (1949).

$$\text{Total Chlorophyll content (mg g}^{-1} \text{ of fresh wt.)} = \frac{[20.2 * A_{645} + 8.02 * A_{663}] * V}{1000 * W}$$

Where

$A_{645}$  = Absorbance at 645 nm

$A_{663}$  = Absorbance at 663 nm

V = final volume of chlorophyll extract in DMSO

W = Weight of plant sample

### 3.4.6.3. Plant water relations

**Leaf water potential ( $\psi$ ):** Second leaf from top of the plants were sampled with intact petiol between 10-11 AM and brought to the laboratory in moist polythene bags. Leaf water potential ( $\psi$ ) was measured using a pressure chamber (s-pms Instruments, New Delhi) following the method of Scholander *et al.* (1964). Each value is the mean of six plants from three pots. Prior to this measurement, the wall of the chamber was lined with moist filter paper and the petiol of an excised leaf was sealed into the chamber such that its cut basal end projected to the outside (atmospheric pressure) and the axial or leaf blade surface remain inside where they were subjected to controlled gas pressure. When the sample was excised, xylem sap pressure potential was increased to atmospheric pressure and sap withdraws from the cut surface in response to its movement from xylem into surrounding cells. When the increasing gas pressure was applied on the blade inside the chamber, cell sap water potential correspondingly increased forcing water back into the xylem. When surface

of the xylem sap just returned to the cut surface, the applied pressure equals that of the sap in the xylem vessels. This balancing pressure which is equal to the leaf water potential was recorded.

**Osmotic potential ( $\pi$ ):** Immediately after the measurement of  $\psi$ , the leaves were killed by dipping in liquid nitrogen and stored at  $-20^{\circ}\text{C}$  for osmotic potential ( $\pi$ ) determination. These leaves were thawed and their sap was extracted and loaded into the pre-calibrated osmometer (5130 B, Wescor Inc. UT., USA) for  $\pi$  measurement. Osmotic potential measurements inferred from measurements of equilibrium, relative humidity (water vapor pressure, assumed to be a direct function of sample water potential) in a small, sealed chamber containing the sample and reference. This Psychrometer employs the principle of Peltier cooling to condense periodically the reference solution (pure, free water) from the chamber atmosphere on to the measuring junction (Spanner, 1951). These types consist only of a wetted thermocouple junction, the surface of which is assumed to be completely wetted by the condensed reference solution (although the wetting is often incomplete because of microscopic surface irregularities on the junction). In addition to the measuring circuit, this psychrometer also requires provision of peltier cooling circuit. The psychrometer was calibrated with a series of solutions of known osmotic potentials.

**Turgour potential (P):** This was calculated as the difference between leaf water potential and osmotic potential.

#### 3.4.6.4. Shoot, root dry weight and leaf area/ pot

After cutting the shoot part from the base, the pots were inverted and soil washed with a stream of water to recover the roots. Leaves were separated from the stem and their total area was measured using leaf area meter (LICOR-100). Then they were dried along with stem part in a hot air oven at  $80^{\circ}\text{C}$  to get the dry weight of the shoot. Correction was made for the leaves used for water potential measurement. Roots were dried after measuring the root characteristics in Root Scanner and added to the shoot weight to get total biomass per pot. Number of nodules was measured per replicate and its dry weight added to total biomass.

**Root characteristics:** This was measured as described earlier using Root Scanner.

### **3.4.6.5. Harvest and yield components**

Five replicates per treatment were harvested at physiological maturity and the plant parts were separated. Number of pods and seed weight per pot and total biomass per pot was recorded.

### **3.4.6.6. Water use efficiency (WUE)**

As described earlier, pots were watered twice a week with measured quantity of water to maintain two soil moisture tensions and these values were added for each replicate. After subtracting the remaining moisture content of the soil in the pot, the total water used by the plants in each replicate was computed until flowering, podding and maturity. Water use efficiency ( $\text{g DM kg}^{-1} \text{ water}$ ) was computed as the slope of the linear regression of daily cumulative above dry matter (g) verses cumulative water used (kg).

### **3.4.6.7. Radiation Characteristics (RUE)**

Both incoming and outgoing Photosynthetically Active Radiation (PAR) values were measured at three heights viz. top, middle (50 per cent canopy height) and bottom of chickpea plants in pots from one month after sowing till physiological maturity using point quantum sensor (LICOR-3000). To get reflected radiation from top, middle and bottom ground, the sensor was held in inverse position. The above measurements were taken at weekly intervals on clear days between 1130 and 1200 hours IST when disturbances due to leaf shading and leaf curling and solar angle were minimum. These data were further used to derive radiation use efficiency.

### **Absorbed Photosynthetically Active Radiation (APAR)**

APAR by the whole canopy = {Incident radiation on the top of the canopy – reflected radiation by the top of the canopy – incident radiation at the bottom (transmitted radiation) + reflected from the ground}

The cumulative daily values were computed for the period until flowering, podding and maturity. Radiation Use efficiency ( $\text{gMJ}^{-1}$ ) was computed as the slope of the linear regression of daily cumulative above dry matter ( $\text{gm}^{-2}$ ) verses cumulative daily absorbed photosynthetically active radiation ( $\text{MJ m}^{-2}$ ).

**Statistical Analysis**

The data was analyzed using the software INDOSTAT. For all the experiments, completely randomized design was followed. For the first experiment on standardization of magnetic field for maximum enhancement in germination characteristics, two factor analysis of variance was performed keeping magnetic field as main factor and duration as sub-factor. The significant level of difference of all measured traits among magnetic fields, duration of exposure and their interaction were calculated. For all other studies, soil moisture potentials/levels were considered as sub-factor and the least significance difference (LSD) among treatments for each trait was calculated.



# RESULTS

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### 4.1. Standardization of magnetic field strength and duration for maximum enhancement in germination of desi chickpea (Pusa 256)

The result of the experiments conducted to identify the optimum magnetic field strength of exposure and duration of exposure are given in Fig.4.1a-f. Germination percentage increased significantly over the unexposed control irrespective of magnetic field and duration (Fig.4.1a). Among the different combination of the field strength and duration of exposure, a 100 mT (1h) gave the maximum germination percentage (7% more than control). Speed of germination also improved over the control in all treatments and was maximum for 50 mT (3h), 100 mT (1h) and 150 mT (1h) (Fig.4.1b). Shoot length of seedlings did not show much variation among the treatments but were higher than the control value (Fig.4.1c). In root length of seedlings, even though most the treatments showed improvement over the control, three of them did not show any increase over the control (Fig.4.1d). Seedling dry weight which gives a measure of vigour of seedlings was highest for 100 mT (1h) treatment (Fig.4.1f).

The overall assessment of the results showed that the exposure of the seeds of chickpea var. Pusa 256 to 100 mT (1h) was the best among different combinations of magnetic field and exposure time and therefore this treatment was used in the subsequent experiments. In case of kabuli chickpea var. Pusa 1053, the same magnetic field exposure of 100 mT (1h) which was found to be the best by an earlier worker (Ananta Vashisth, 2007) was used in the subsequent experiments.

### 4.2. Seed water absorption, seed water activity and seed water diffusivity

The moisture content corresponding to -0.1 MPa, -0.2 MPa and -0.4 MPa soil water potentials calculated from the soil moisture characteristics curve were 19.42%, 11.06% and 8.4% respectively (Appendix I). These moisture contents were maintained throughout the experimentation. Soil samples were taken periodically and the moisture content of the prepared soils were checked gravimetrically and found to be within the error range of 1-3%.

#### **4.2.1. Kinetics of seed water absorption in relation to soil moisture potential as affected by magnetic field exposure**

The temporal changes in seed water absorption expressed on mg water per seed were studied at 25°C using soil with different moisture regimes. The seed absorption of the two genotypes when placed in soils maintained at -0.1, -0.2 and -0.4 MPa soil water potential are given in Fig.4.2.1(a-c) for Pusa 1053 and in Fig.4.2.1(d-f) for Pusa 256. The observations were recorded till the radical protrusion took place. In all the soil moisture regimes, there was an increase in seed water content with respect to time though the rates of absorption varied with genotypes and soil water potential. As expected, seed water absorption increased with increase in soil water potential. But in both varieties, seed water uptake was consistently higher for magnetically treated seeds as compared to untreated controls. The percent increase in seed water absorption over the unexposed control was higher initially which reduced as time progressed for all three soil water potentials studied and in both the varieties.

#### **4.2.2. Kinetics of seed water activity in relation to soil moisture potential**

The temporal changes in seed water activity which reflects the active part of moisture content were measured at 25°C using Rotronic Hygrometer / Water Activity Meter at different moisture regimes. The seed water activity of the two genotypes when placed in soils maintained at -0.1, -0.2 and -0.4 MPa soil water potentials are given in Fig.4.2.2(a-c) for Pusa 1053 and in Fig.4.2.2(d-f) for Pusa 256. In case of seed water activity, there was no specific trend with respect to imbibitions time in Pusa 1053 at all soil water potentials and also at -0.4 MPa potential in case of Pusa 256, even though magnetically exposed seeds showed higher values in general over unexposed control. Water activity in Pusa 256 increased continuously in both treated and untreated seeds with imbibitions time at -0.1 and -0.2 MPa potentials and the magnetically treated seeds showed greater values than the untreated seeds.

#### **4.2.3. Seed water diffusivity in relation to soil water potential**

The calculation of seed water diffusivity either by Phillips method or by Crank's equation requires the time of germination ( $t_g$ ) and moisture content at germination ( $M_g$ ). In the present experiment, at -0.4 MPa soil water potential, both

varieties did not germinate even after 10 days. Therefore, the soil water diffusivity at -0.4 MPa potential could not be calculated.

The radical protrusion was earlier in magnetically treated seeds over the control. At -0.1 and -0.2 MPa soil water potentials, the relative moisture contents were too small and the Philips equation could not be used for the calculation of diffusivity. Hence the Crank's equation which was a simpler version of Philip's equation was used to calculate seed water diffusivities at -0.1 and -0.2 MPa potentials. The relative values of seed water absorption ( $M_t/M_g$ ) at different times of imbibitions were used to get the respective values of  $(Dt/a^2)^{1/2}$  from the graph (Appendix II). Substituting the value of sphere equivalent radius ( $a = 0.391$  cm), the diffusivity ( $D$ ) at different imbibition time ( $t$ ) was calculated. Table 4.2.3a&b and Table 4.2.4a&b give the relative water absorption and diffusivity calculation for the two varieties at -0.1 and -0.2 MPa soil water potentials respectively. The respective  $t_g$  (germination time) and  $M_g$  (seed water at germination) are also given in the tables. Change in seed water diffusivities with time at -0.1 MPa and -0.2 MPa potentials are given in Fig.4.2.3a&b for Pusa 1053 and in Fig.4.2.4a&b for Pusa 256. The results showed significant increase consistently in seed water diffusivity over the unexposed control at -0.1 MPa soil water potential in both varieties of chickpea. At -0.1 MPa soil water potential, the percent increase in seed water diffusivity over the control in Pusa 1053 varied from 15.9% (2h) to 71.61% (30h) and in Pusa 256, it varied from 23.45% (4h) to 71.61% (28h). At -0.2 MPa potential, there was no specific trend in Pusa 1053 whereas in Pusa 256, there was consistent increase for treated seeds over control and the percent increase in seed water diffusivity over the control varied from 46.53% (4h) to 10.66% (56h).

### 4.3. Root growth dynamics and root characteristics

Magnetically exposed and unexposed seeds of desi chickpea and kabuli chickpea were grown in sand and peat mixture in plastic pots maintained at 20% and 12% moisture content on dry weight basis. The pots were kept inside the green house and the mean maximum and minimum temperatures during the experimentation period were 22.8°C and 9°C respectively. Periodic samplings were done by washing the roots in running water.

#### 4.3.1. Dynamics of root characteristics of Pusa 1053 under different soil moisture content

Root characteristics like total root length, root volume and root surface area were studied by Win RHIZO Program and other parameters like longest root length, root weight and shoot dry weight were measured in the laboratory for magnetically treated and untreated control. Results obtained from periodic samplings for root characters are presented in Fig.4.3.1(a-c) for 20% soil moisture and in Fig.4.3.1(d-f) for 12% soil moisture. Average root diameter in all cases including varieties and magnetic treatment did not show any appreciable variation and hence omitted from presenting the data.

**Total Root length:** It is clearly seen that there was little increase with time until 20 DAS and then a sharp significant increase for the subsequent two samplings for all the above mentioned parameters for plants from treated seeds over the unexposed control in both 20% and 12% soil moisture content(SMC). These increases were 116% and 154% over control at 30 and 40 DAS at 20% SMC and were 126% and 93% respectively at 12% moisture content. At 20 percent soil moisture content percent increase over the control varied from 20.6% (10 DAS) to 154% (40 DAS) whereas at 12% SMC varied from 35% (20 DAS) to 126% (30 DAS).

**Root volume:** Similar trend was observed in case of root volume. Significant increase was observed only for 30 DAS and 40 DAS samplings and these increases over the control were 204% and 137% respectively at 20% SMC and were 147% and 166% respectively at 12% SMC. For 20% SMC, percent increase over the control varied from 14.7% (10 DAS) to 204% (30 DAS) and for 12 percent soil moisture content it was from 34.5% (20 DAS) to 166 % (40 DAS).

**Root surface area:** Significant increases were observed over control in plants from magnetically treated seeds in 30 DAS and 40 DAS samplings in both soil moisture levels. The increases over control were 149% and 131% respectively at 20% SMC and were 114% and 120% respectively at 12% SMC. The percent increase over the control varied from 13.6 (10 DAS) to 149 (20 DAS) at 20% SMC as compared to 36 (20 DAS) to 120 (40 DAS) at 12% SMC.

#### 4.3.2. Dynamics of root and shoot growth of Pusa 1053 under different soil moisture content

The growth parameters like longest root length, root and shoot dry weights at different samplings for Pusa 1053 at two different soil moisture regimes are given in Fig.4.3.2(a-f) for magnetically treated and untreated seeds.

**Longest root length:** In case of longest root length significant increase over untreated controls were obtained at 30 DAS and 40 DAS samplings in treated plants when the soil moisture was 20% or 12%. The increases were 48% and 53% respectively at 20% soil moisture as compared to 40.5% percent and 45.5% percent respectively at 12% soil moisture content. Percent increase over the control varied from 11.4 (10 DAS) to 53 (40 DAS) for 20% soil moisture content and 13 (10 DAS) to 43 (40 DAS) for 12% soil moisture content.

**Root and shoot dry weight:** Root dry weight of magnetically treated plants showed significant increase over control from 20 DAS onwards under both soil moisture levels. Percent increases over control were 55 to 140% at 20% soil moisture and were 37% to 48% at 12% soil moisture content. Significant increases in shoot dry weight were observed at 30 and 40 DAS at both soil moisture contents. The increase over control for 30 & 40 DAS samplings were 47.8% and 66.4% respectively at 20% moisture and were 28 and 44% at 12% moisture content.

#### 4.3.3. Dynamics of root characteristics of Pusa 256 under different soil moisture content

Root characteristics like total root length, root volume and root surface area measured at different time intervals for Pusa 256 are presented in Fig.4.3.3(a-c) for 20% soil moisture and in Fig.4.3.3(d-f) for 12% soil moisture content.

**Total root length:** Significant increase over control was observed both for 30 and 40 DAS samplings at both soil moisture levels in treated plants. Increases were 49% and 39% respectively for 20% SMC and 12.8% and 6.9% respectively for 12% SMC. In 20% SMC percentage increase over control varied from 23% (10 DAS) to 49% (30 DAS), whereas for 12% SMC, it varied from 6.9% (40 DAS) to 47.6% (10 DAS)

**Root volume:** Significant increase of 44.9% over control was observed only at 40 DAS sampling for 20% SMC, whereas there was consistent increase from 9 to 40% at all samplings in case of 12% SMC.

**Root surface area:** Similar trend was observed as root volume as significant increase of 44.2% over was observed only at 40 DAS sampling for 20% SMC. For 12% SMC significant increase (73.5%) was observed in 10 DAS and the percent increase over control decreased with each sampling with an increase of only 5.5% at 40 DAS.

#### **4.3.4. Dynamics of root and shoot growth of Pusa 256 under different soil moisture content**

The growth parameters like longest root length, root and shoot dry weights at different samplings for Pusa 256 at two different soil moisture regimes are given in Fig.4.3.4(a-f) for magnetically treated and untreated seeds.

**Longest root length:** This parameter increased sharply until 30 DAS in treated and control plants and then declined in both 20 and 12% SMC. In case of 20% SMC significant increase (43.5% and 39% respectively) were observed at 30 and 40 DAS whereas increases of only 28.3% (10 DAS) and 15.3% (30 DAS) were recorded for 12% SMC.

**Root and shoot dry weight:** Increase in root dry weight of treated plants over control was significant at most samplings in both soil moisture contents. Increases varied from 14.5% to 41% in 20% SMC and varied from 14% to 63% in 12% SMC. Though shoot weight increased significantly at 20% SMC, the corresponding increase at 12% SMC was only marginal.

The result of the root dynamics study showed clearly that in treated plants, there were consistent increase in all parameters over unexposed control. But the improvement was greater at 20% SMC than at 12% SMC in most cases. Even though both varieties showed similar trend, the absolute values and percent increase over control were varying. The values for total root length, root volume, root surface area and root weight were consistently higher and the values for longest root length, shoot weight were lower in case of Pusa 256 compared to Pusa 1053 at both 20% and 12%

soil moisture levels. But percentage increases over control were consistently lower in Pusa 256 in comparison to Pusa 1053.

#### **4.4. Photosynthesis, water relations and growth parameters**

Pot culture experiment was conducted in the green house protected from rain with a polyacrylic sheet cover with 90% radiation penetration. Mean maximum and minimum temperatures during this growth period were 26.81°C and 11.5°C respectively. Flowering and podding were early in Pusa 1053 by 2-5 days and was earlier by 3-4 days in -0.2 MPa stressed plants of both varieties in both varieties compared to unstressed plants (-0.1 MPa). But all matured around the same time due to increased in temperature during second week of March.

##### **4.4.1. Leaf photosynthesis parameters**

Photosynthesis and related parameters like stomatal conductance and transpiration rate measured on second matured leaf from top at flowering and podding stages of Pusa 1053 plants are given in Fig.4.4.1(a-c). Similar measurements made on the plants of chickpea variety Pusa 256 is given in Fig.4.4.1(d-f). In both varieties, irrespective of magnetic treatment, there was significant reduction in photosynthesis at podding stage compared to flowering stage and reduction due to soil moisture stress. At flowering stage, though both varieties showed a marginal increase of about 6% due to magnetic treatment at -0.1 MPa potential, at -0.2 MPa potential, only Pusa 1053 showed an significant increase over the control plants (38.6%). At podding stage, except for an increase of 57% over control in treated plants of Pusa 256 at -0.1 MPa potential, all other treatments did not show any improvement over the control. Leaf stomatal conductance followed nearly the same trend as photosynthesis with significant increase in treated plants at flowering stage in stressed plants. Leaf transpiration rate did not show any advantage for treated plants over controls in most cases. But at podding, there was significant increase in Pusa 256 at -0.1 MPa and a decrease at -0.2 MPa for treated plants over the untreated controls.

#### **4.4.2. Leaf chlorophyll, leaf area and specific leaf weight**

Chlorophyll content of the second mature leaf on which photosynthetic measurements were taken, leaf area/pot and specific leaf weight for both varieties are given in Fig.4.4.2(a-c) & Fig.4.4.2(d-f). In Pusa 1053, in both growth stages and in both soil moisture levels, the treated plants had more chlorophyll than the untreated controls. In Pusa 256, in stressed plants of both stages, there was decline in treated plants. Leaf area/pot declined drastically due to soil moisture stress in both treated and untreated plants of both varieties. However, treated plants of both varieties maintained greater leaf area as compared to untreated control plants. Specific leaf weight of treated plants of Pusa 1053 showed marginal (1.3%) to significant (24%) increase over the control. In Pusa 256, except for a 8.4% increase at -0.1 MPa in podding stage, the treated plants had lesser values than the control plants.

#### **4.4.3. Leaf water relations**

Leaf water status as given by total leaf water potential, osmotic potential and turgour potential at flowering and podding stages measured on the second mature leaf are given in Fig.4.4.3(a-c) for Pusa 1053 and in Fig.4.4.3(d-f) for Pusa 256 respectively. In both varieties, there was decrease in total water potential with respect to soil moisture stress and with maturity. There was concomitant decrease in osmotic potential which resulted in positive turgour in all treatments. Leaf water potential of treated plants were in general higher than the untreated plants in both varieties. Similarly, turgour potential of treated plants was greater for treated plants than control which was more significant under stress conditions.

#### **4.4.4. Root characteristics**

Total root length, root volume and root surface area of the treated and control plants grown under two moisture regimes are given in Fig.4.4.4(a-c). & Fig.4.4.4(d-f). for Pusa 1053 and Pusa 256 varieties. All root parameters drastically decreased under stress conditions in both varieties. But, in treated plants of both varieties, all of them showed increase to different levels over untreated controls. In Pusa 1053, total root length of treated plants were greater by 19% in -0.1 MPa and were 54 % and 15% in -0.2 MPa soil moisture potential. In Pusa 256, the increase over control at -0.1 MPa



was very high both at flowering (71.5%) and at podding (101%) stages. Root volume increased marginally (from 9.1 to 38%) in treated plants of Pusa 1053 under all moisture levels. Only in case of Pusa 256, at podding stage, highest increase of 80 % was recorded at -0.1 MPa. Similar trend was observed for root surface area and the increase over control varied between 14 % and 46%. The highest increment of 76 % was recorded in Pusa 256 at podding stage under -0.1 MPa potential.

#### **4.4.5. Shoot and root dry weight and nodule number**

In both varieties, there was significant decrease in shoot and root dry weight in stressed plants of both treated and untreated plants (Fig.4.4.5.a-f). However, treated plants of Pusa 1053 maintained 9.6 to 25.3% increase in shoot weight and 24 to 40% increase in root weight over the untreated controls. Pusa 256 also maintained an increase of 12 to 44% in shoot weight and 12 to 54% in root weight over the controls. But, in this variety, at podding, there was a decline of 8.4% in root dry weight in treated plants as compared to control at -0.2 MPa potential. Under stress conditions, no nodules were produced in both treated and untreated plants. At -0.1 MPa potential, an increase of 11 to 22 % in Pusa 1053 and an increase of 31 to 34% in Pusa 256 in number of nodules was recorded.

#### **4.5. Biomass, pod number, grain weight/ pot, RUE and WUE**

At physiological maturity, biomass, pod number and grain weight/pot of the harvested plants of both varieties and treatments are presented in Table 4.5. Also presented in the table, are the calculated values of Radiation use efficiency (RUE) and Water use efficiency (WUE) of different treatments. From the table it is clear that irrespective of varieties and magnetic treatment, there was drastic reduction in biomass and grain yield in stressed plants. In Pusa 1053, due to magnetic treatment, there was only a marginal increase in yield components in unstressed plants. But under stress conditions, the improvement in magnetically treated plants over untreated control was significant and varied between 25 to 30%. In Pusa 256, magnetically treated plants showed significant increase in yield parameters at both soil water potentials. The improvement in grain weight was 67% at -0.1 MPa and 32% at -0.2 MPa soil water potential. This increase was due to significant enhancement in biomass and pod number per pot. In Pusa 1053, both RUE and WUE improved

substantially in treated plants over control (49 to 62%) at both soil water potentials. However, in Pusa 256, under -0.1 MPa potential, both RUE and WUE of treated plants decreased by 20 and 7.6% as compared to control. In stressed condition, treated plants showed improvement in RUE and WUE to the extent of 31 to 35% over the corresponding untreated control plants.

# DISCUSSIONS

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### 5.1. Standardization of magnetic field strength and duration for maximum enhancement in germination

Pre-sowing exposure of seeds of chickpea var. Pusa 256 to different magnetic fields showed that there was overall stimulating effect of magnetic field with respect to all germination characters (Fig.4.1a-f). Such enhanced performance of seeds in their germination characteristics have been reported in other crops (Pittman, 1965; Gubbels, 1982; Kato, 1988; Phirke, 1990; Anna, 2002; Fischer, 2004; Florez, 2007; Vashisth and Nagarajan, 2008a). In kabuli type chickpea, Vashisth and Nagarajan (2008b) reported enhanced performance in terms of laboratory germination characters and improved root and shoot growth of one month old plants in the field. They reported highest response when the seeds were exposed to a magnetic field of 100 mT for one hour. But the mechanism for such an increase has not been completely understood. In wheat, Pittman and Ormrod (1970) reported that the seedlings grown from magnetically treated seed absorbed more moisture, respired more slowly, released less heat energy and grew faster than the untreated controls. In soybean, Kavi (1977) observed that the seeds exposed to magnetic field had increased capacity to absorb moisture. The increased physiological activity due to greater absorption of moisture by treated seeds may be responsible for the increase in seedling length, seedling dry weight and vigour indices in our study. But some fields were more effective than others and there was no linear increase with increase in field strength. In the same way, the response to exposure time also varied and no direct relation between improvement in seedling parameters and time of exposure was observed. However, Florez *et al.* (2007) reported that the accumulation of dry weight of 10 day old seedling from seeds exposed to magnetic field increased logarithmically with duration of magnetic field induction. In their study the duration of magnetic field induction was increased from 1 minute to 24 h and also continuous exposure whereas we have gone until 3h of exposure only. Moreover, the seeds in their study were imbibed in water before the exposure to magnetic field whereas dry seeds were exposed to magnetic field in our study. Fischer *et al.* (2004) reported that sunflower

seedlings exposed to magnetic field showed small but significant increases in total fresh weight, shoot fresh weight and root fresh weight, whereas dry weight and germination rates remained unaffected. Experimentally treated wheat exhibited marginally (but significantly) higher root fresh and dry weights, total fresh weights and higher germination rates. Kiranmai (1994) found that 200 mT magnetic field produce positive mutations in sunflower.

The interaction of magnetic field and exposure time indicated that certain combinations of magnetic field and duration were highly effective in enhancing most of the germination characters compared to other combinations (Fig.4.1a-f). This observation suggests that there may be a resonance like phenomena which increases the internal energy of the seed and that occurs when there is appropriate combination of magnetic field and exposure time. In ragi (*Eleusine coracana* Gaertn) seeds, exposure to magnetic field changed its internal potential energy which could be used to get higher yields by suitably selecting the magnetic field and exposure time (Kavi, 1983). In case of desi type of chickpea, our results showed that an exposure of 100 mT for one hour gave maximum enhancement in seedling characters which matched with that reported for kabuli type chickpea earlier (Vashisth and Nagarajan, 2008b).

## **5.2. Seed water absorption, seed water activity and seed water diffusivity**

The seed water uptake pattern of magnetically treated and untreated chickpea seeds (Pusa 1053 and Pusa 256) were compared at different soil water potentials. At all soil water potentials, the seeds exhibited a general trend of higher rate of absorption initially which decreased gradually as the seed water content increased. Similar results were reported by Chatterjee *et al.* (1981) in seeds of gram, cotton, soybean and cowpea, by Hadas (1976) in leguminous seeds and by Chatterjee (2004) in wheat seeds. Hadas (1970) has also observed that germination will occur at a soil moisture content depending on the seed capabilities, seed moisture potential and conductivity. This indicated that the rate of water uptake by seeds depended mainly on the internal water potential of the seed. In the dry state, initially, the seed water potential is very low as compared to the potential of the surrounding medium and therefore, there is higher rate of water absorption. As the water enters the seed, the seed water potential increases and the difference between seed water and soil water potentials decreases and so the rate of absorption also reduces.

The temporal changes in seed water absorption showed an initial rapid increase which was followed by a steady increase until radical protrusion took place. In both varieties, seed water absorption was greater for magnetically treated seeds compared to unexposed control. A similar increase in absorption was reported in drought tolerant wheat genotype as compared to susceptible genotype (Chatterjee, 2004). It appears that exposure of seeds to magnetic field has improved water diffusivity through seed coat membrane as it has happened in drought tolerant cultivar. With decrease in soil water potential, time taken for radical protrusion increased and the seed water content at germination decreased. But, the radical protrusion was earlier in treated seeds of both varieties at all potentials and at slightly higher seed water content. Among the two varieties, Pusa 256 germinated earlier with lower seed water content. Water activity of seed water which describes the active part of seed water that is available for metabolic activities was in general higher for magnetically treated seeds. Water activity at germination was greater for Pusa 256 for both treated and untreated seeds compared to Pusa 1053 albeit lower seed water content. Hence, due to magnetic treatment, not only seed water absorption, but also the corresponding seed water activity increased which resulted in earlier germination of the treated seeds.

Seed water diffusivity increased with time in all treatments and soil water potential. But the absolute values of diffusivity decreased with decrease in soil water potential. Similar results were reported by Chatterjee *et al.* (1981) in two gram genotypes. Only in Pusa 1053 at -0.2 MPa potential, it decreased after 20 h duration. Different explanations have been put forward for the increase in diffusivity with time and with seed water content by many workers (Phillips, 1968; Hadas, 1970; Shakewich and William, 1971). The increase in diffusivity may be due to an increasing percentage of the seed surface becoming covered with a film of water as time increases. Secondly, it may be due to an increase in contact area as the water uptake proceeds. Also, when the seed water content increases, there is change in internal moisture status of the seeds resulting in a change in its internal water potential and conductivity to water. Since the external water potential is never zero, the seed has to overcome that potential for water to enter the seeds. Entry of water increases the seeds internal water potential and increases its conductivity to water. Hence, there is increase in diffusivity with time. Magnetically treated seeds showed higher

diffusivity at all potentials in both varieties. In a similar experiment with wheat seeds, drought tolerant cultivar showed higher diffusivity at different soil water potentials compared to susceptible cultivar (Chatterjee, 2004).

### **5.3. Root growth dynamics and root characteristics**

Periodic measurements of root parameters like total root length, root volume and root surface area under 20% and 12% soil moisture levels showed significant increase in plants raised from magnetically exposed seeds compared to unexposed control in both varieties only from 30 days after sowing. The relative increase due to magnetic treatment was greater for Pusa 1053 than Pusa 256. This is because in Pusa 256, even in untreated plants, the root characters are higher than those of Pusa 1053. Similar trend was observed for other seedling characters like longest root length, root and shoot dry weights. Pusa 256 being a desi type is hardier than the kabuli variety, Pusa 1053 and has inherently better root system. Therefore, the improvement due to magnetic field is not very high as in Pusa 1053. In the same chickpea variety Pusa 1053, Vasisth and Nagarajan (2008) have reported doubling of total root length, root surface area and root volume in one month old plants grown from seeds treated with 100 mT magnetic field for 1h. They also reported significant increase in longest root length, root and shoot dry weights of the plants. Rajendra *et al.* (2005) have observed a significant increase in mitotic index as well as  $^3\text{H}$ -thymidine incorporation into DNA in seeds of *Vicia faba* exposed to 100  $\mu\text{T}$  power frequency magnetic field. A similar mechanism may be operating in chickpea also wherein increased cell number of magnetically treated plants during initial sampling period might have led to greater expansion of these cells in the subsequent samplings. This would have resulted in higher growth rates of root and shoot parameters in plants from magnetically treated seeds. The significant increase in root and shoot weights and the greatly improved root characteristics in the plants from magnetically treated seeds has practical importance in chickpea which is a rainfed crop and generally grows under receding stored soil moisture. When large number of Ethiopian land races were evaluated for drought tolerance, it was found that the tolerant genotypes produced more root weight, root volume and rooting depth compared to susceptible check (Anbessa and Bejiga, 2004). Serraj *et al.* (2004) also suggested that deep and prolific root systems are associated with enhance tolerance to drought in chickpea.

#### 5.4. Photosynthesis, water relations and growth parameters

In the study conducted in pots maintained under two different soil water potentials showed significant improvement for various physiological traits in plants from magnetically treated seeds measured at flowering and podding stages of the crop. Photosynthesis reduced with stress as well as with maturity of the crop. This reduction from flowering to podding coincided with reduction in leaf water potential. The rate of net photosynthesis in chickpea leaves in rainfed condition decreased to values below  $5 \mu\text{mol m}^{-2}\text{s}^{-1}$  at the onset of seed filling compared to values above  $20 \mu\text{mol m}^{-2}\text{s}^{-1}$  in an adjacent irrigated crop (Turner *et al.*, 2001). They also reported that it reached a peak prior to podding and decreased rapidly during pod filling. The decrease in photosynthesis occurred as the leaf water potential decreased. Plants from magnetically treated seeds of both varieties in general maintained relatively higher rate of photosynthesis which may be attributed to better leaf water status as described by higher leaf water potential. In chickpea, it is observed that osmotic adjustment take place when the stress increased progressively (Lecoeur *et al.*, 1992). In our study, we find that irrespective of magnetic treatment, osmotic adjustment take place as the osmotic potential decreased in step with decrease in leaf water potential. This enabled the leaves to maintain positive turgour albeit at different levels. Plants from magnetically treated seeds, under both soil moisture conditions, maintained higher leaf water status in terms of leaf water and turgour potentials. Leaf chlorophyll content did not vary much due to soil moisture stress or due to advancement in growth stage. But other growth parameters like leaf area, specific leaf weight, shoot and root dry weights and total root length increased from flowering stage to podding stage and all of them reduced drastically due to water stress. Magnetic treatment was able to ameliorate the effect of stress to some extent which may be attributed to maintenance of better leaf water status by osmotic adjustment and greater root length and root surface area than the control. Increased uptake of  $\text{Ca}^{2+}$  ions in rice seedlings grown from seeds exposed to pulsed magnetic field was found responsible for better leaf growth, meristematic tissues in stems and roots (Saktheeswari and Subrahmaniyam, 1989). Chickpea is a cool-season legume, adopted to grow well on stored moisture in the post-rainy season with little or no irrigation (Turner, 2003). But, in our experiment in pot culture, in  $-0.2 \text{ MPa}$  soil moisture potential, both varieties did not produce any

nodules. The plants produced nodules in -0.1 MPa potential which increased with growth stage and with magnetic treatment.

### 5.5. Harvest maturity, RUE and WUE

The results obtained at harvest maturity showed drastic reduction in biomass, pod number and grain weight/pot of both varieties under stress conditions in magnetically treated and control plants. This may be attributed to a large number of physiological and physical factors. Heat or drought stresses if severe during reproductive development, particularly after the commencement of pod set, can cause significant pod abortion (Leport *et al.*, 1999) and decrease seed filling (Leport *et al.*, 2006). Reduced rate of photosynthesis, decreased leaf water potential and lesser development of root characters must have led to poor development of leaf area, specific leaf weight and yield components like pod number and seed weight. Among the two varieties, desi variety, Pusa 256 produced greater biomass and grain weight than the kabuli variety, Pusa 1053. Davies *et al.*, (2000) have shown that in desi chickpea, C and N assimilated prior to podding can supplement the supply of current assimilates to the filling of seeds in both well watered and water stressed plants. Wang *et al.*, (2006) reported 26% greater seed yield in desi chickpea than in kabuli type cultivar under same conditions. Plants from magnetically treated seeds gave marginal advantage over plants from untreated plants of both varieties. In a study on the relationship of different physiological traits, Singh *et al.*, (2004) reported a positive correlation of yield with biomass and chlorophyll content in chickpea. Higher biomass/pot and greater chlorophyll content in leaves of plants from magnetically treated seeds at flowering and podding stages might have resulted in relatively better grain weight at harvest.

Accurate measurement of Radiation use efficiency (RUE) in potted plants using point quantum sensor is difficult as the canopy is not evenly covered. However, our measurements give a rough idea about the effect of soil moisture stress and magnetic field on the values of RUE. It was computed as the slope of the linear regression of cumulative dry matter versus cumulative intercepted photosynthetically active radiation (PAR) until flowering, podding and maturity. This kind of linear relationship between crop growth and cumulative intercepted radiation has been



reported in many crops (Monteith, 1994). Variability in RUE can be understood in terms of physical and biological process, which determine the state of plant canopy (Rouphael and Colla, 2005). Weather parameters like solar radiation, air temperature and vapour pressure deficit influence the physical and physiological process of the crops (Vijaya Kumar *et al.*, 1996) which intern affect the RUE. This may be the reason for the poor RUE of plants grown under -0.2 MPa potential as compared to plants grown under -0.1 MPa potential due to low vapour pressure deficit. Climatic and plant growth conditions affect the plant water use and therefore, Water use efficiency (WUE). It has been shown in greenhouse rose crop that WUE is inversely proportional to the vapour pressure deficit and temperature (Duchein *et al.*, 1995). This explains the low values obtained for WUE in stressed plants of our experiment. In both varieties, except for Pusa 1053 at -0.1 MPa potential, magnetic field exposure has improved RUE and WUE over the unexposed controls. This may be attributed to better shoot and root development of the plants raised from treated seeds.

## Chapter - 6

### SUMMARY & CONCLUSION

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Experiments were conducted to study the effect of pre-sowing seed exposure to selected magnetic field on two chickpea varieties with a view to understand their mechanism of action in terms of seed water absorption, root dynamics, growth and yield. A desi variety, Pusa 256 and a kabuli variety, Pusa 1053 were selected for this study. Initial experiments were conducted to find out the optimum dose of magnetic field and duration of exposure for maximum enhancement of seedling characters in Pusa 256. An exposure of 100 mT for 1h was found optimum which was the same for the kabuli variety Pusa 1053 which has been already reported. Therefore, the seeds of both varieties exposed to 100 mT static magnetic field for 1h were used as the experimental material and the following experiments were conducted:

1. Temporal changes in seed water absorption, seed water activity and seed water diffusivity when placed in soils maintained at different soil water potentials.
2. Dynamics of root characteristics, root and shoot growth in sand culture maintained under two moisture levels.
3. A pot culture experiment in the green house where soil moisture potentials were maintained at -0.1 MPa and -0.2 MPa to study physiological and growth parameters at different phenological stages and relate them to yield and energy and water use efficiencies.

Major findings of the study are given below:

1. Temporal changes in seed water absorption exhibited similar pattern of initial rapid increase followed by steady increase until radical protrusion in all seed lots. But, magnetically exposed seeds of both varieties had higher rate of absorption which may be attributed to magnetically induced greater seed membrane permeability to water.
2. Seed water activity, which is the functional part of cellular water that participates in metabolic activities increased in general with seed water

absorption and was greater for magnetically exposed seeds. This indicated that in treated seeds, not only seed water content increased, but also its corresponding water activity that resulted in early germination.

3. Seed water diffusivity increased in general with time and the values were greater at high soil water potential compared to low soil water potential. In magnetically treated seeds, seed water diffusivity values were greater than untreated controls and hence better water absorption in these seeds.
4. Dynamics of root characters like, total root length, root surface area and root volume studied under two soil water regimes (12 & 20%) in sand culture showed consistent increases in all parameters in treated plants over unexposed control. The relative increase over untreated control was more for Pusa 1053 than Pusa 256. The desi variety, Pusa 256 inherently had better root system than kabuli variety Pusa 1053 and therefore, effect of magnetic field was marginal.
5. Periodic measurements in root and shoot weight and the length of the longest root in the same experiment also showed similar trend.
6. Study conducted in pots maintained under -0.1 MPa and -0.2 MPa soil water potentials exhibited significant improvement for various physiological traits (leaf photosynthesis, stomatal conductance, transpiration, leaf water potential, osmotic potential, turgour potential) in plants raised from magnetically treated seeds. All the above said parameters reduced significantly from flowering to podding stage and with soil moisture stress.
7. Growth parameters such as leaf area, specific leaf weight, root and shoot weight and root parameters like total root length, root surface area and root volume increased from flowering to podding and decreased with soil moisture stress. However, in plants from treated seeds, adverse effect of stress was ameliorated as they maintained relatively higher photosynthesis and leaf water status through osmotic adjustment and greater root length and root surface area.
8. At harvest maturity, irrespective of magnetic treatment, drastic reduction in grain weight of stressed plants was observed due to severe reduction in

pod number per plant. However, in well watered plants, plants from magnetically treated seeds produced more biomass, pod number and grain weight than untreated controls. Pusa 256 produced more biomass and grain weight than Pusa 1053.

9. In both varieties, Radiation Use Efficiency (RUE) and Water Use Efficiency (WUE) decreased sharply in severe soil moisture stress (-0.2 MPa) conditions. Except for Pusa 1053 at -0.1 MPa potential, magnetic field exposure has improved RUE and WUE over the unexposed controls. This may be due to better shoot and root development of the plants raised from treated seeds.

Therefore, it may be concluded that exposure of dry seeds to static magnetic field of 100 mT for 1h improved seed water absorption characteristics in desi and kabuli varieties of chickpea. These resulted in early germination and early vigour of seedlings in terms of root and shoot weight and root characteristics. Improved root system coupled with superior leaf water status led to increased photosynthesis in adult plants and produced greater biomass and grain weight. Also, when these plants were subjected to severe water stress (-0.2 MPa), the adverse effect was ameliorated partially.

**Future study:**

1. The studies can be extended to other soil types with different compaction levels.
2. Magnetic treatment of seeds can be compared with other conventional seed priming treatments to evaluate its superiority.
3. Large scale field trials may be conducted to understand the beneficial effect of magnetic treatment under farmers' conditions.
4. The mechanism of magnetic treatment in enhancing root development should be studied at hormonal and molecular level which can be exploited in molecular plant breeding.

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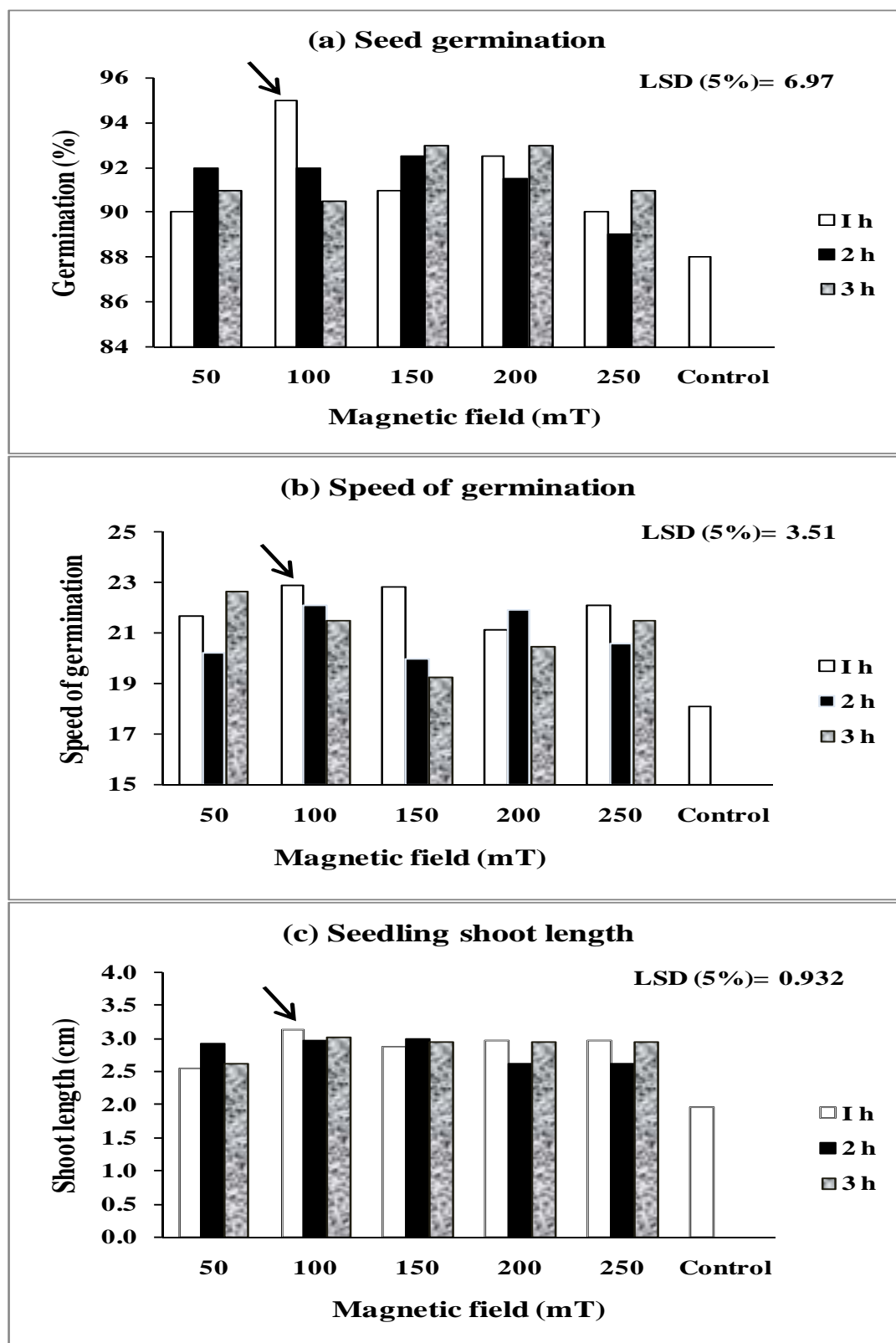


Fig.4.1: Effect of different magnetic field and duration of exposure on (a) Germination% (b) Speed of germination and (c) Shoot length of 8 days old seedling of chickpea var. Pusa 256

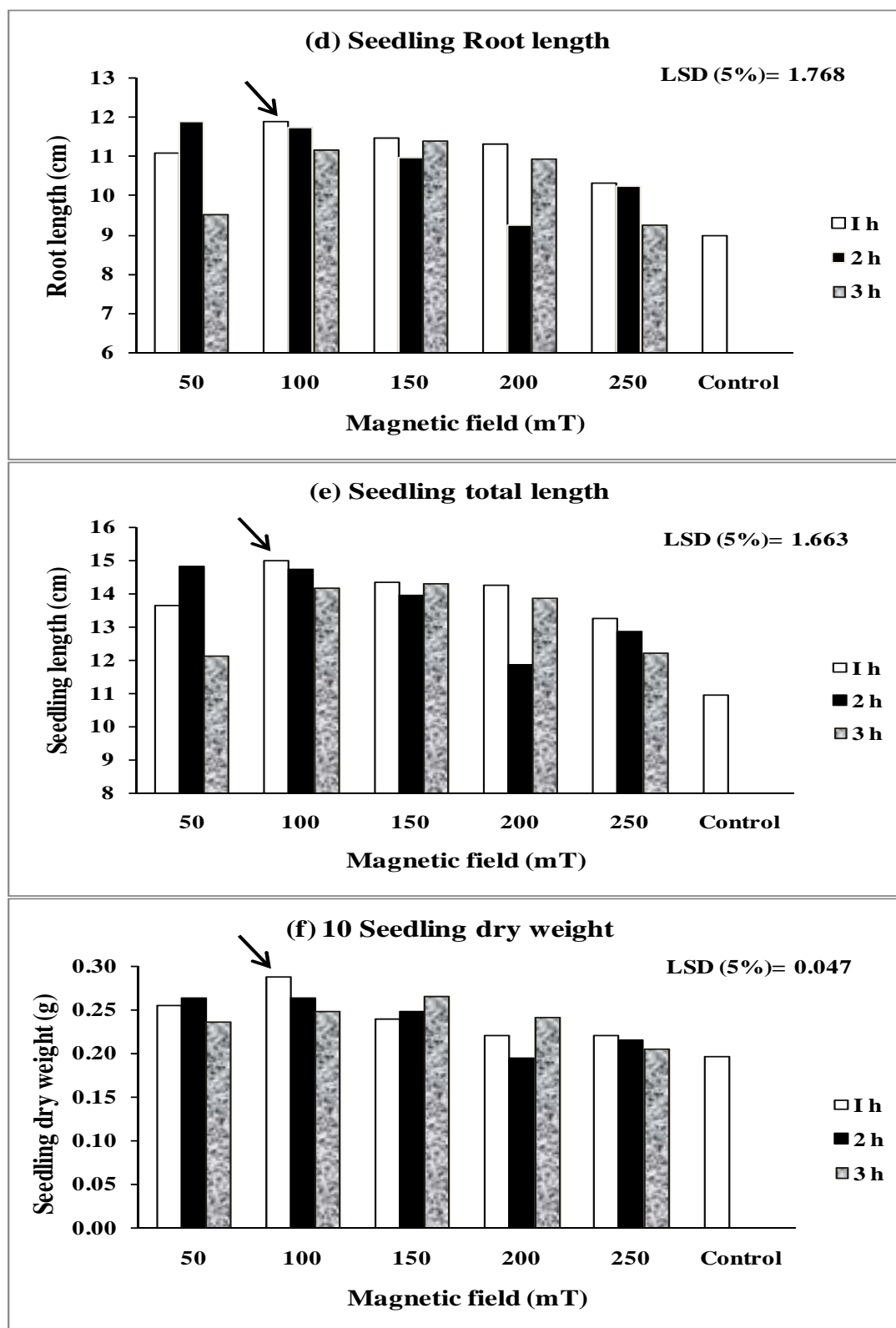
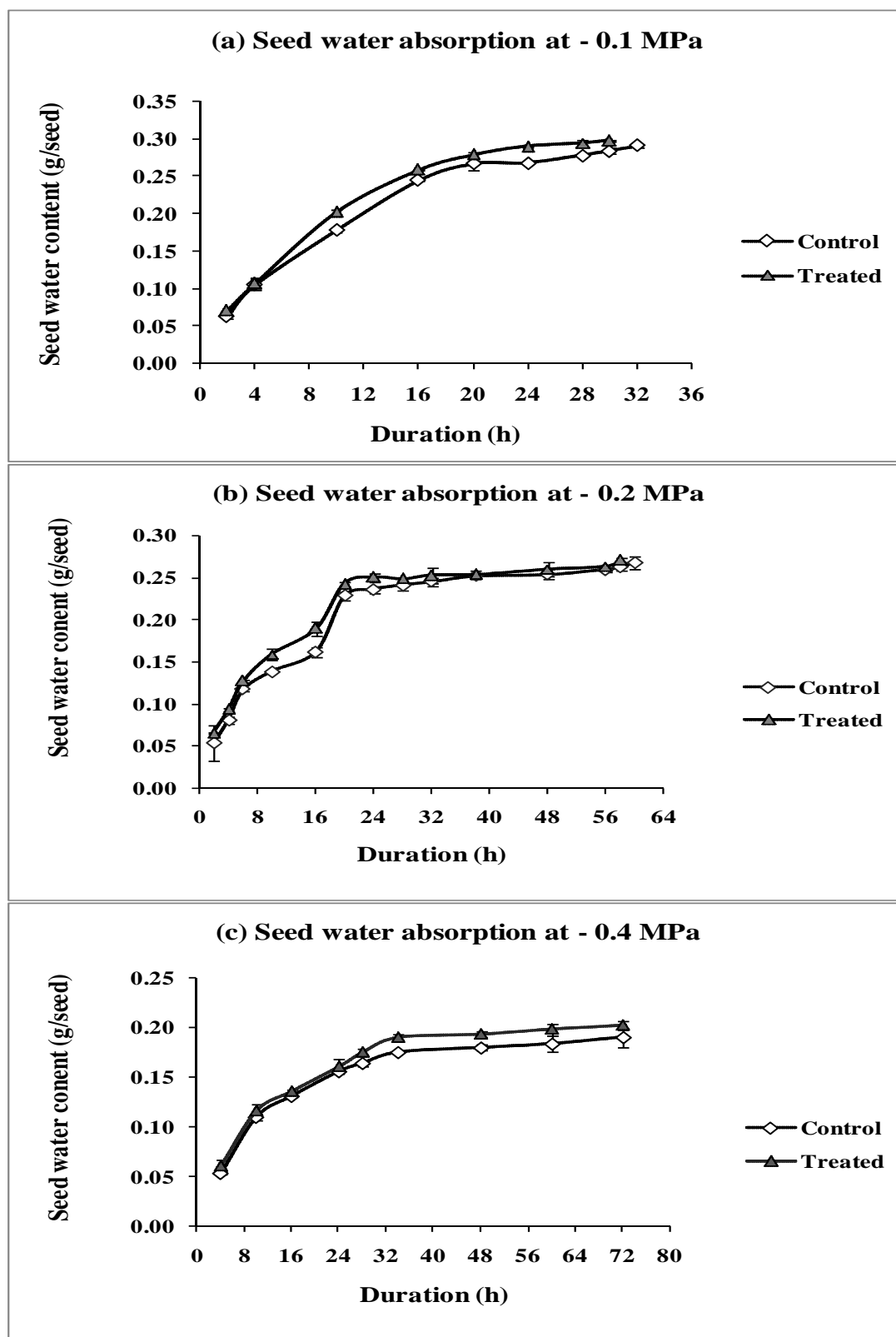


Fig.4.1: Effect of different magnetic field and duration of exposure on (d) root length (e) seedling total length and (f) 10 seedling dry weight of 8 days old seedling of chickpea var. Pusa 256





**Fig.4.2.1: Seed water absorption of magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 at (a) -0.1 MPa, (b) -0.2 MPa and (c) -0.4 MPa soil moisture potentials**

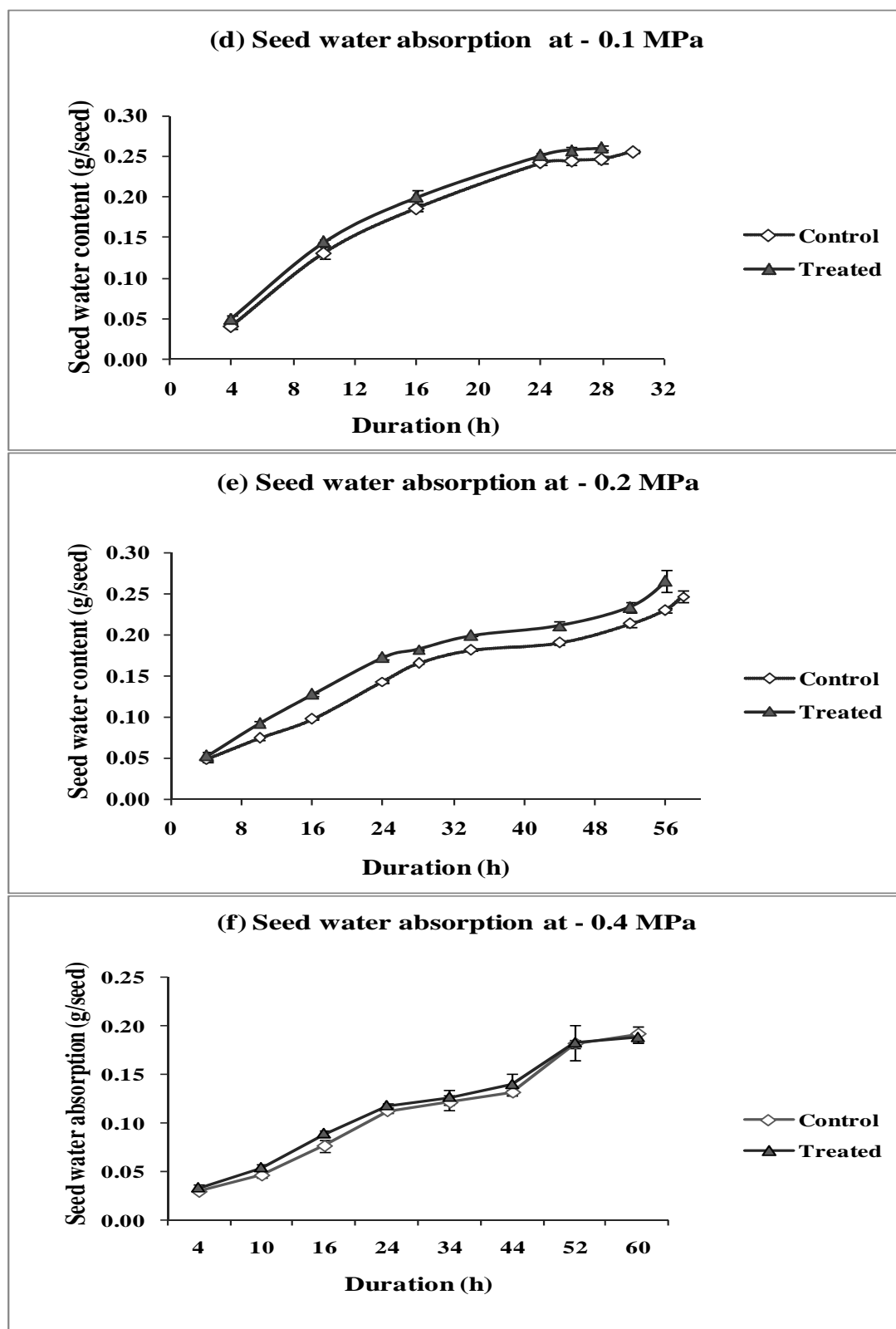
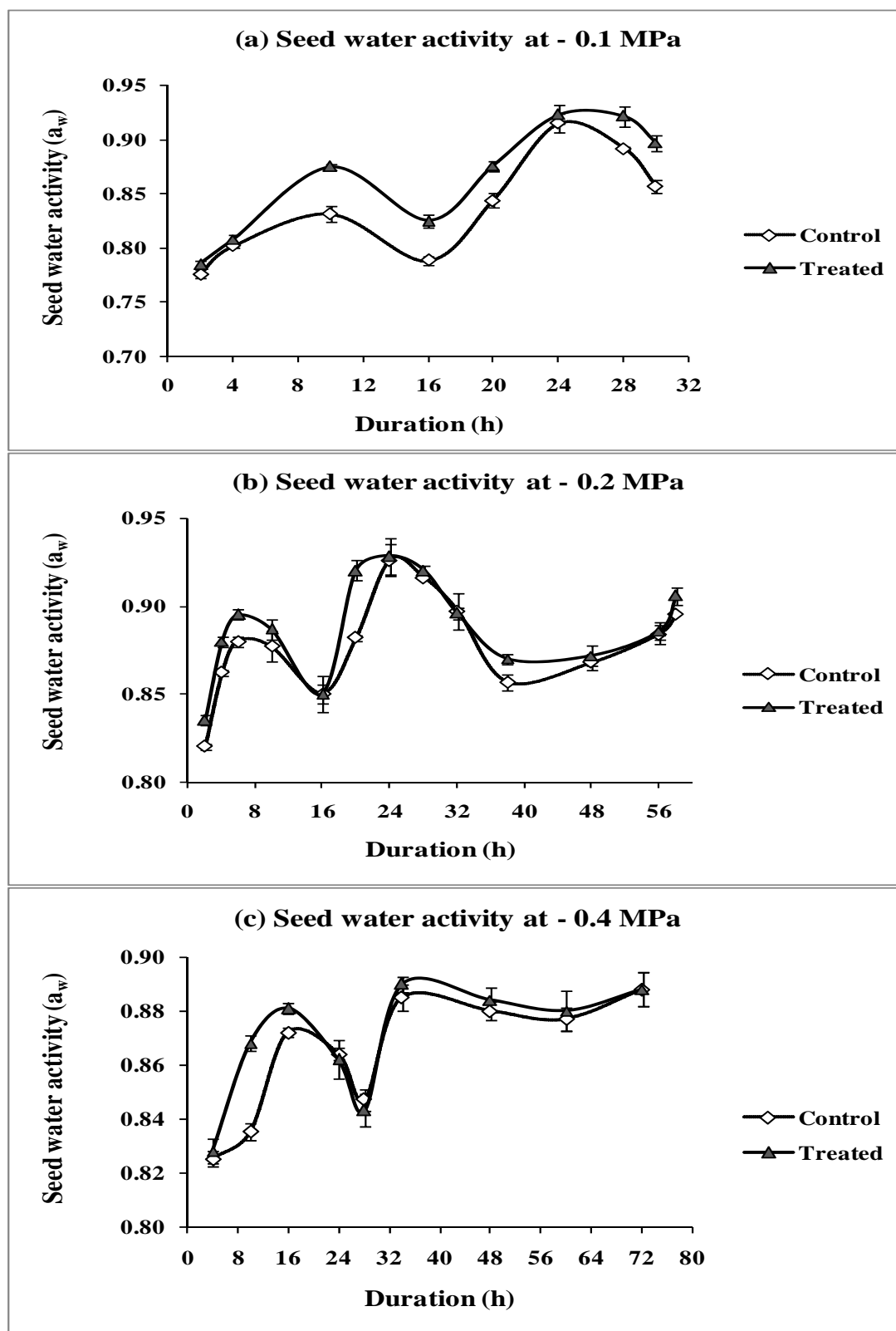
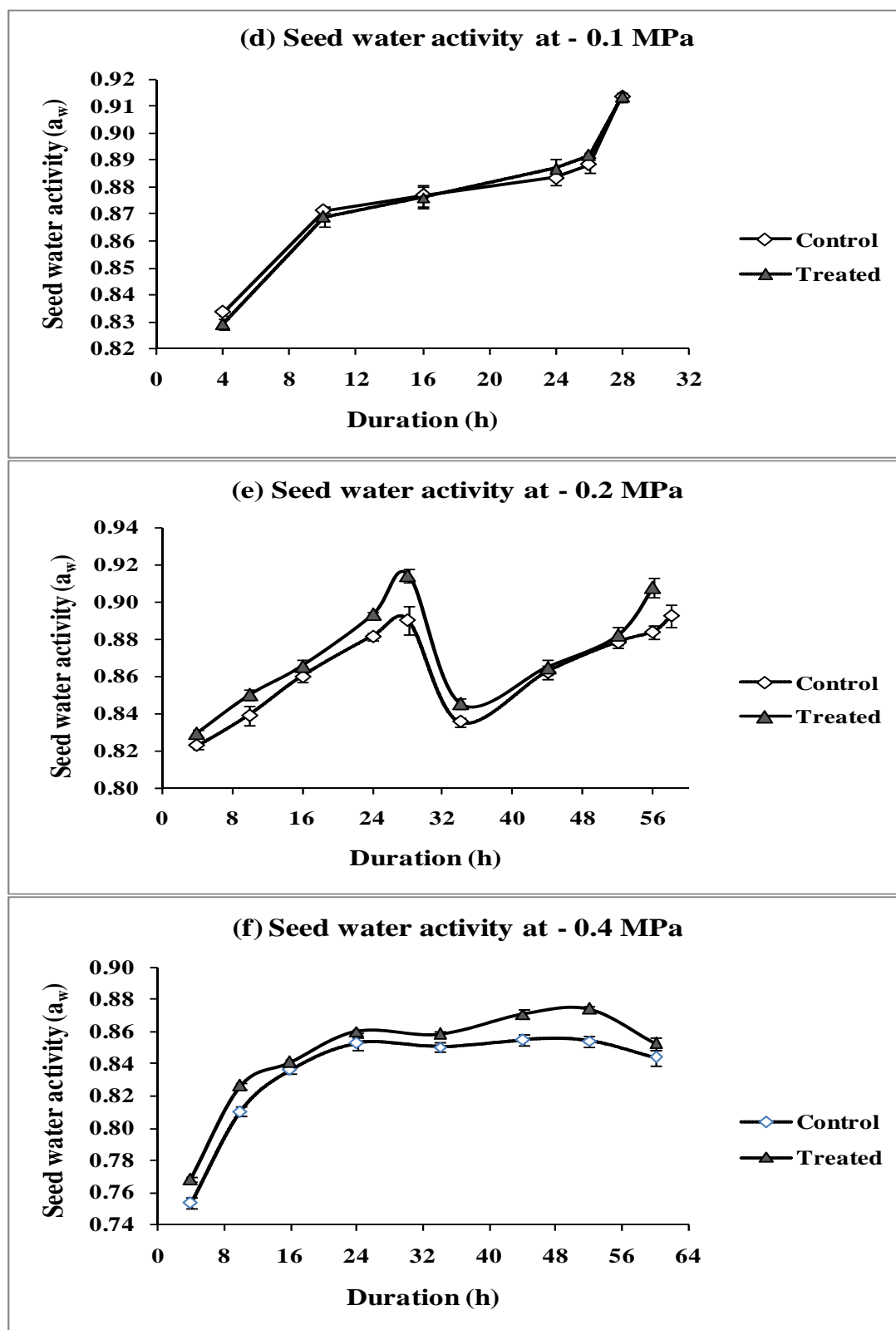


Fig.4.2.1: Seed water absorption of magnetically exposed and unexposed seeds of chickpea var. Pusa 256 at (d) -0.1 MPa, (e) -0.2 MPa and (f) -0.4 MPa soil moisture potentials



**Fig.4.2.2:** Changes in seed water activity with time in magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 at (a) -0.1 MPa, (b) -0.2 MPa and (c) -0.4 MPa soil moisture potentials



**Fig.4.2.2:** Changes in seed water activity with time in magnetically exposed and unexposed seeds of chickpea var. Pusa 256 at (d) -0.1 MPa, (e) -0.2 MPa and (f) -0.4 MPa soil moisture potentials

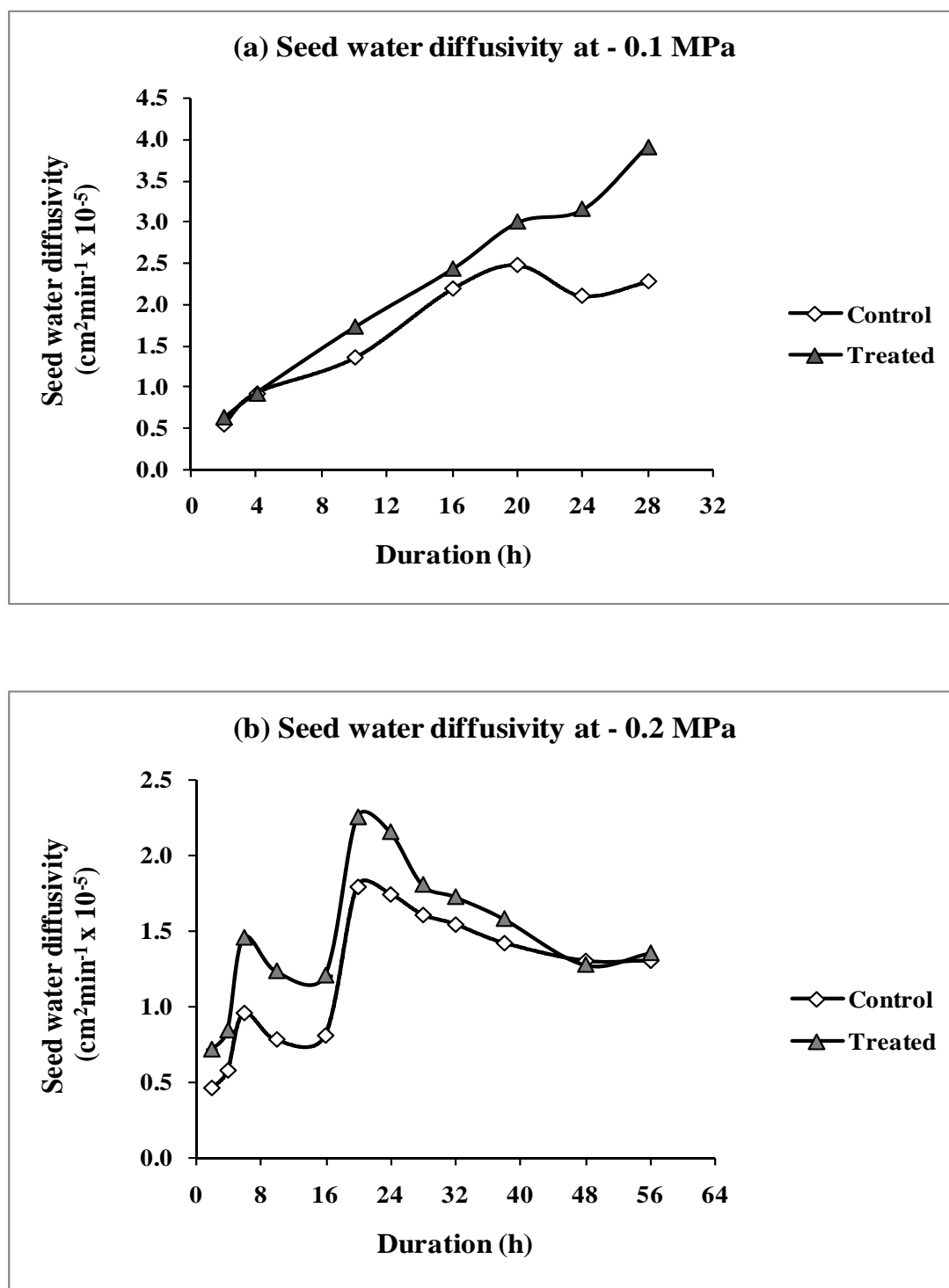
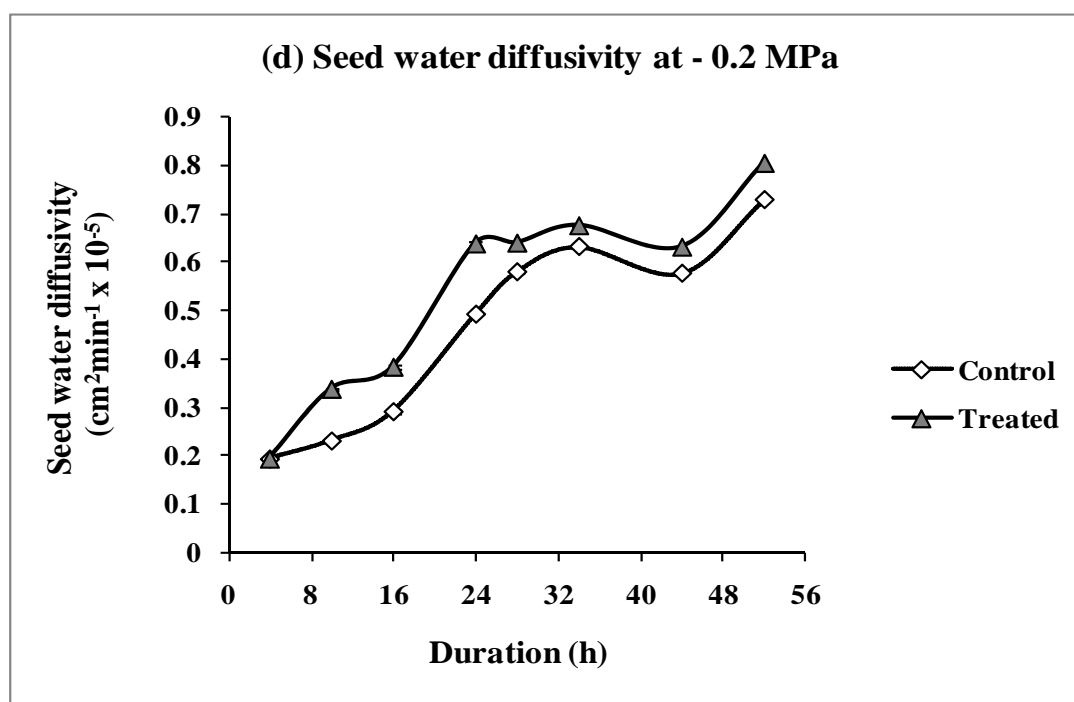
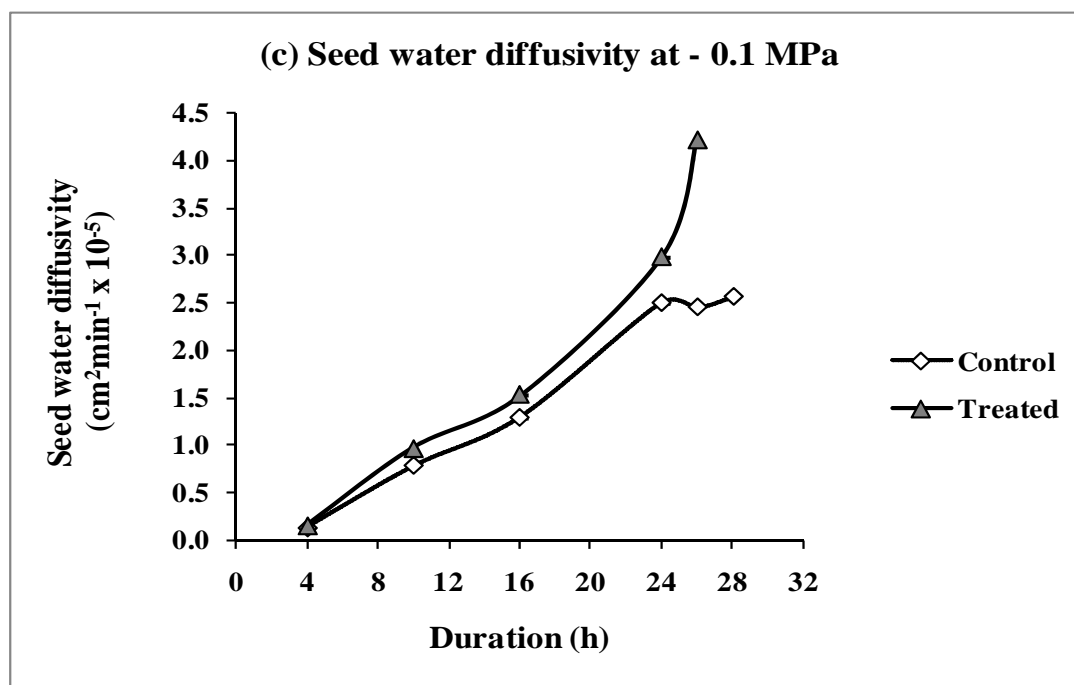
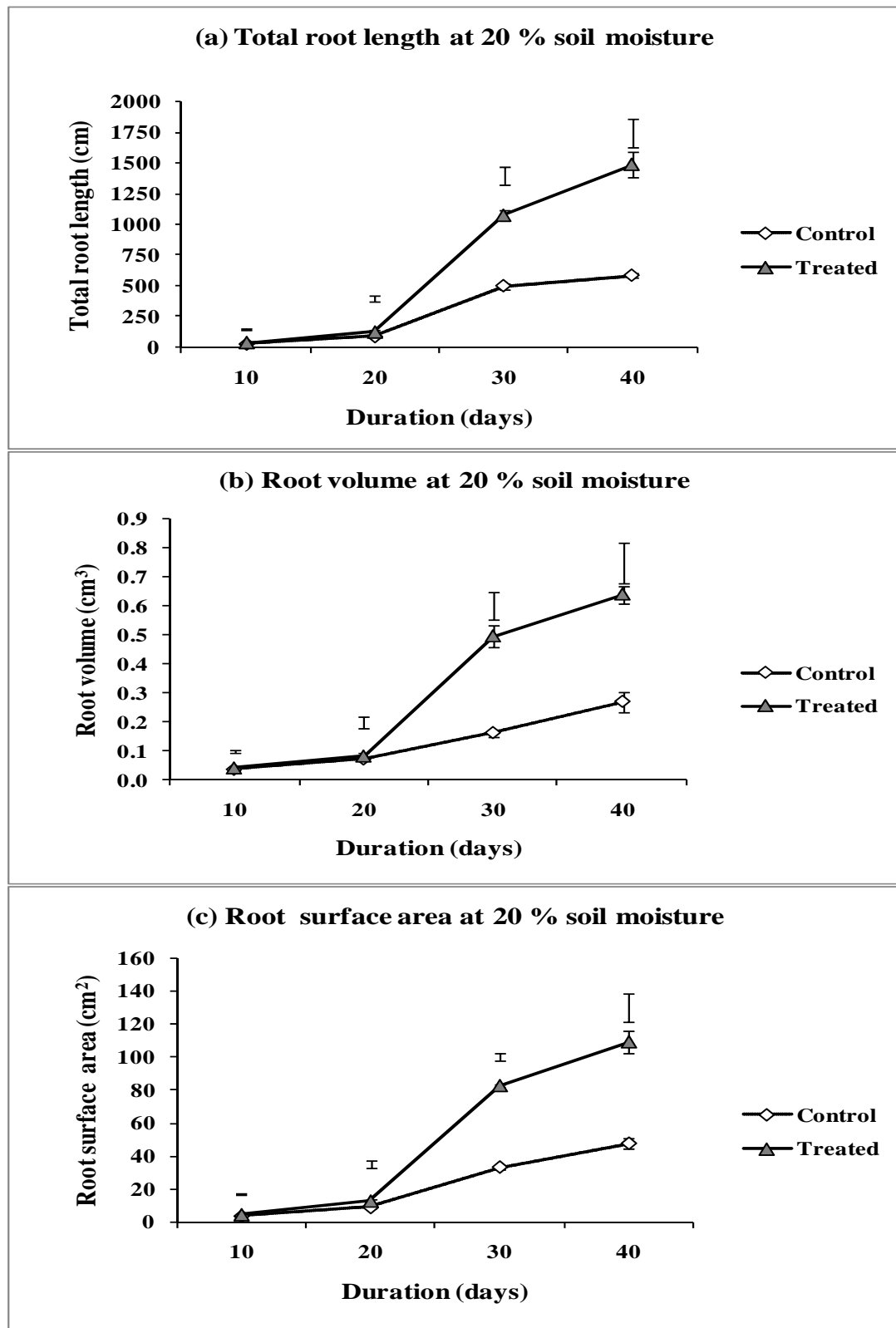


Fig.4.2.3: Change in seed water diffusivities of magnetically exposed and unexposed chickpea var. Pusa 1053 at (a) -0.1 MPa and (b) -0.2 MPa soil water potentials



**Fig.4.2.3: Change in seed water diffusivities of magnetically exposed and unexposed chickpea var. Pusa 256 at (c) -0.1 MPa and (d) -0.2 MPa soil water potentials**



**Fig.4.3.1: Changes in (a) total root length, (b) root volume and (c) root surface area of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 grown in sand culture with 20% moisture**

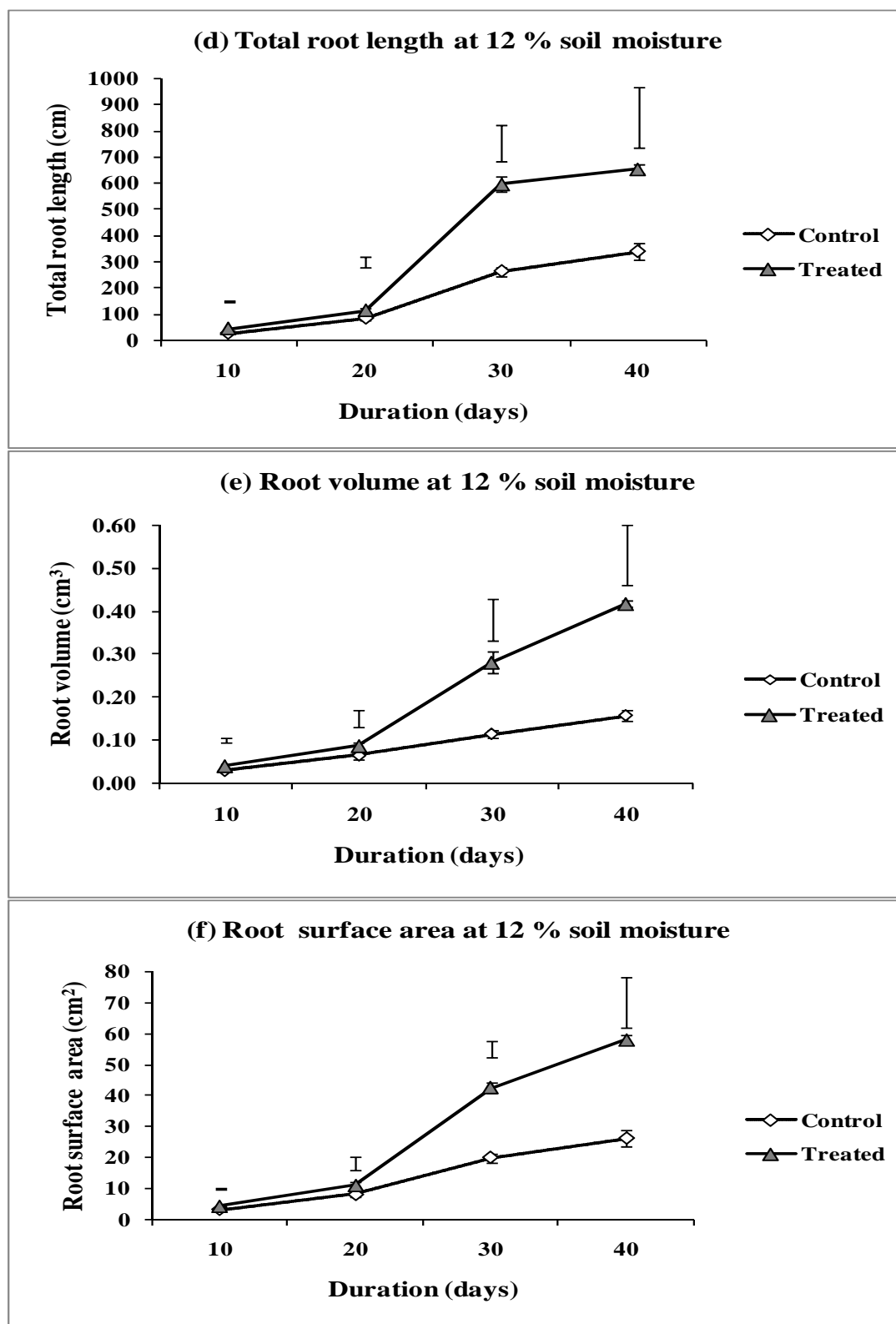
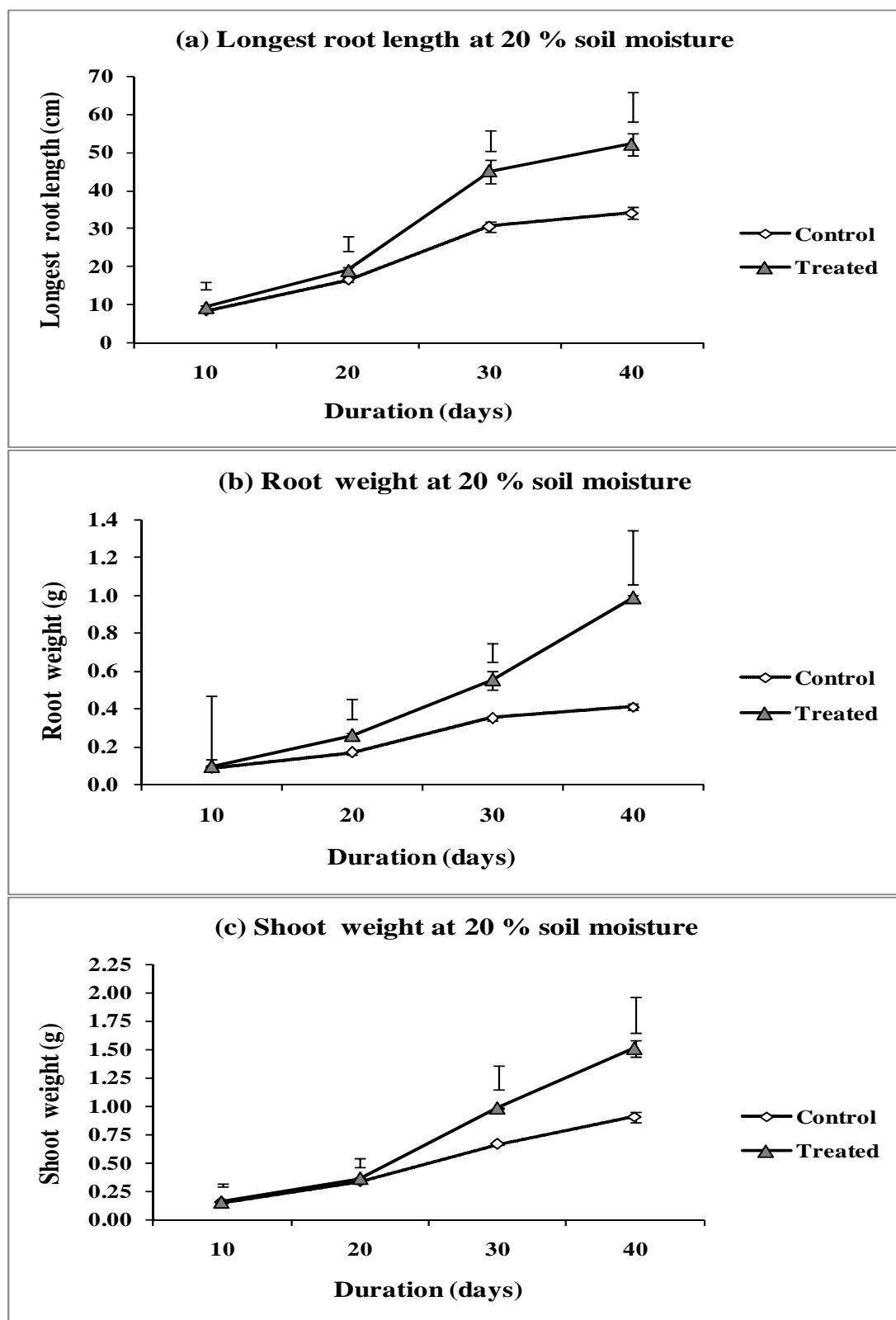
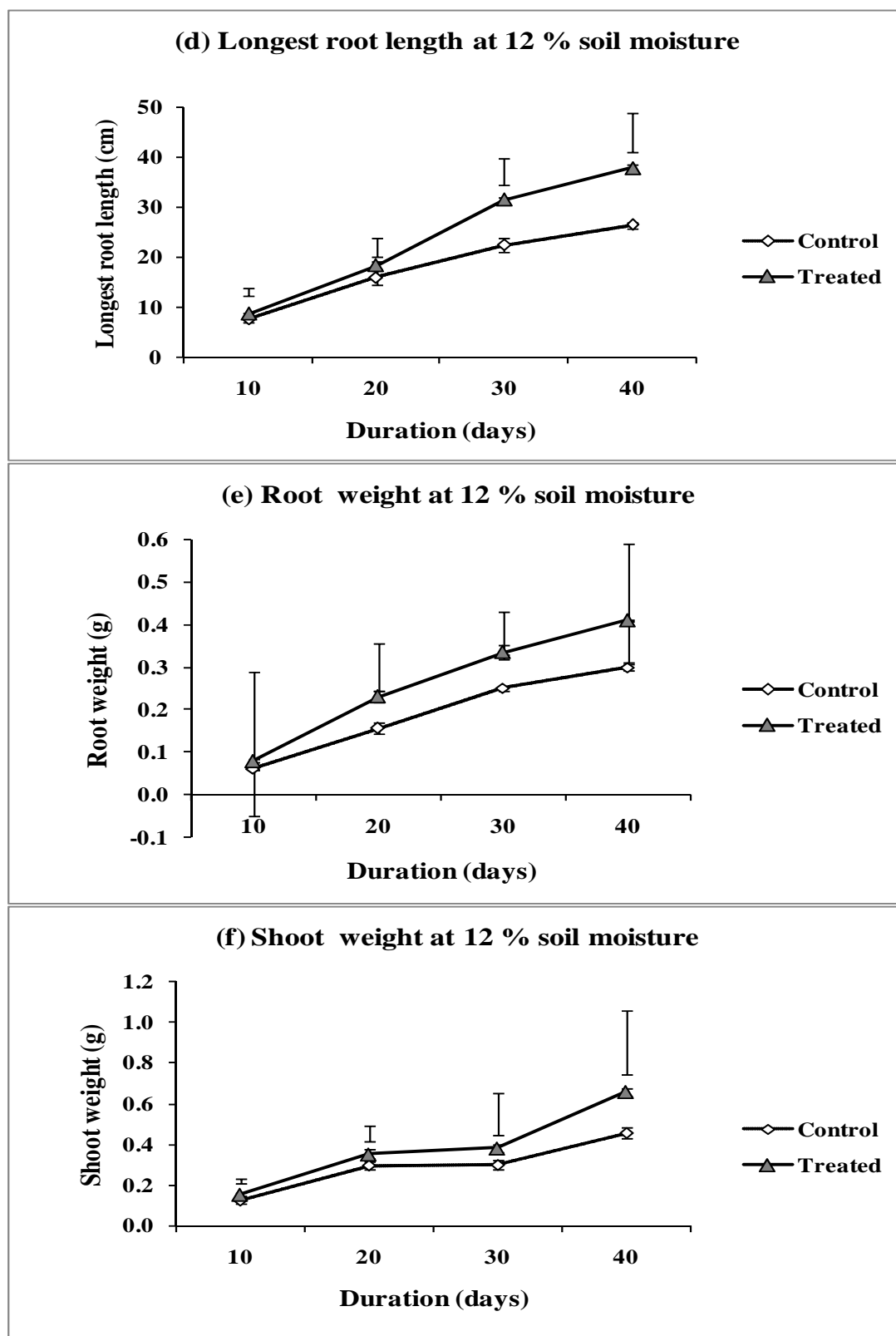


Fig.4.3.1: Changes in (d) total root length, (e) root volume and (f) root surface area of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 grown in sand culture with 12% moisture

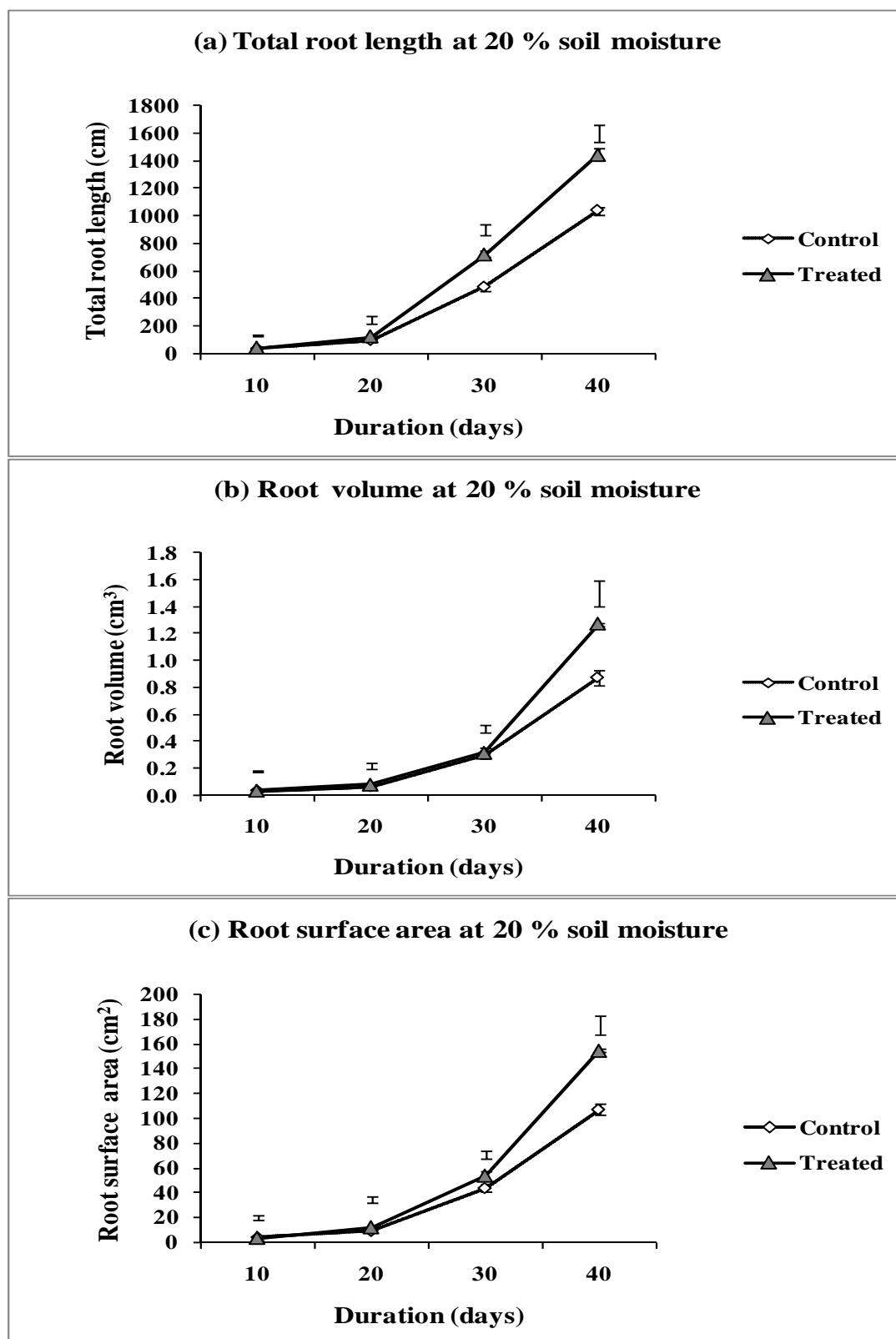




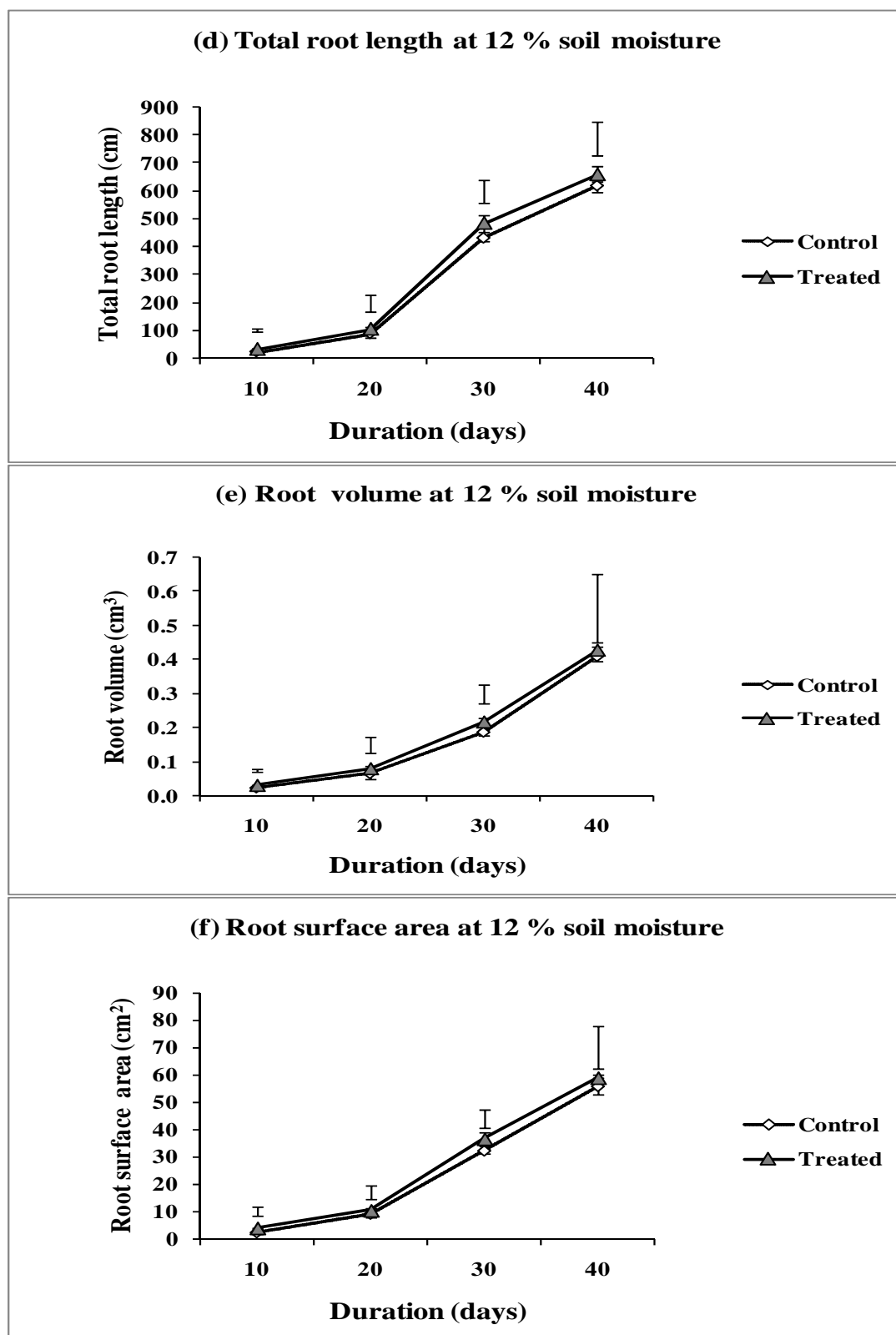
**Fig.4.3.2: Changes in (a) longest root length, (b) root weight and (c) shoot dry weight of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 grown in sand culture with 20% moisture**



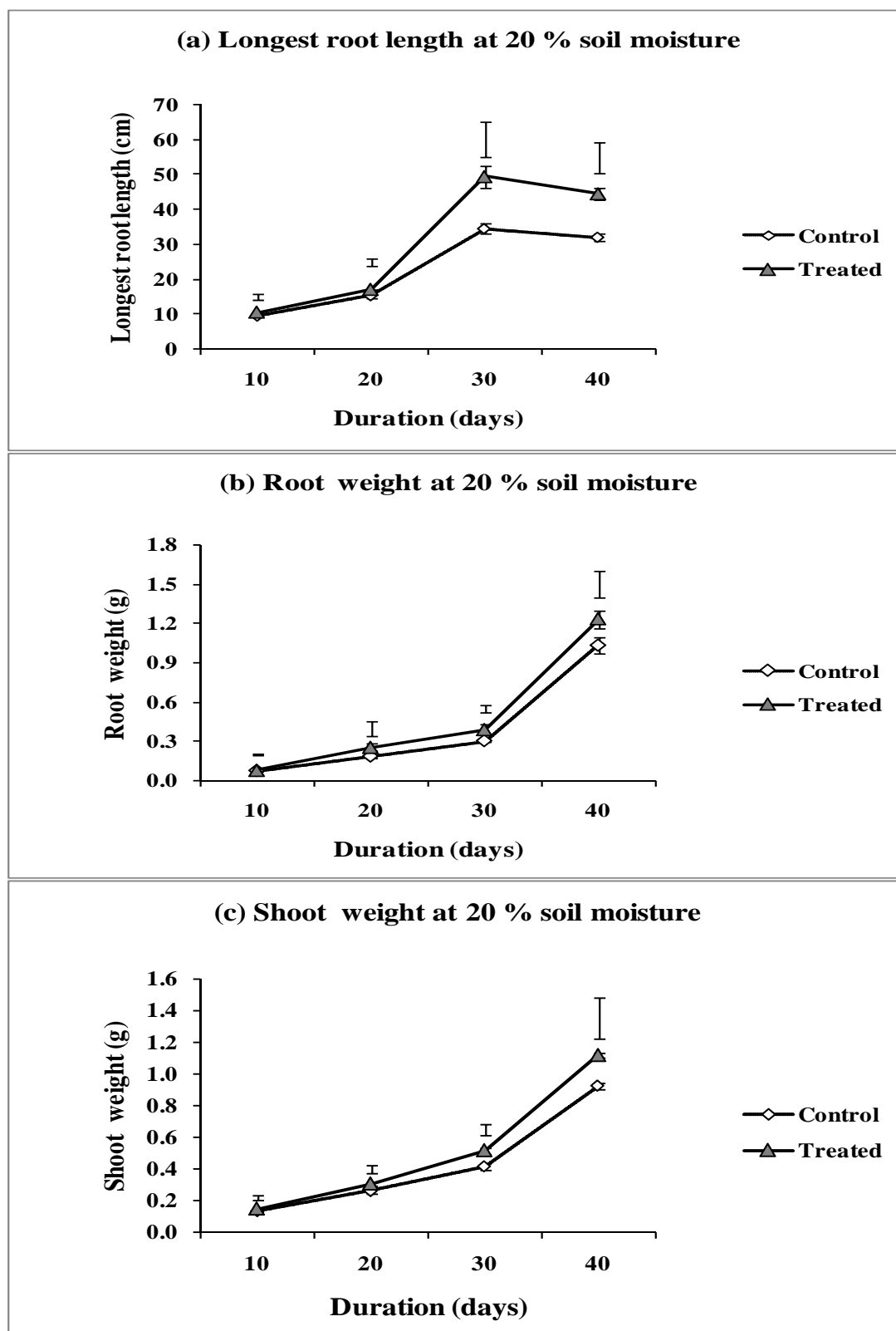
**Fig.4.3.2: Changes in (d) longest root length, (e) root weight and (f) shoot dry weight of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 grown in sand culture with 12% moisture**



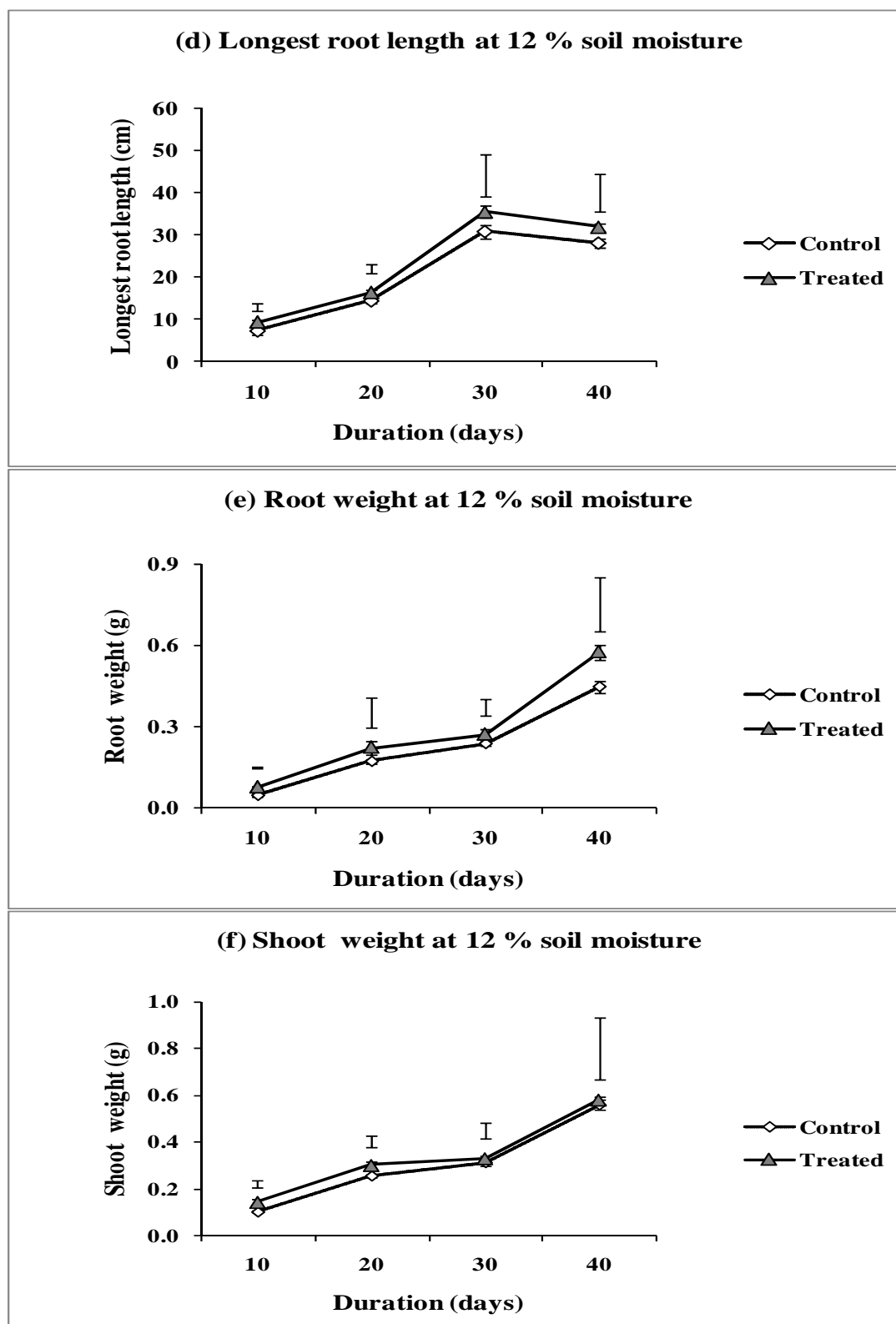
**Fig.4.3.3: Changes in (a) total root length, (b) root volume and (c) root surface area of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 256 grown in sand culture with 20% moisture**



**Fig.4.3.3: Changes in (d) total root length, (e) root volume and (f) root surface area of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 256 grown in sand culture with 12% moisture**



**Fig.4.3.4:** Changes in (a) longest root length, (b) root weight and (c) shoot dry weight of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 256 grown in sand culture with 20% moisture



**Fig.4.3.4: Changes in (d) longest root length, (e) root weight and (f) shoot dry weight of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 256 grown in sand culture with 12% moisture**

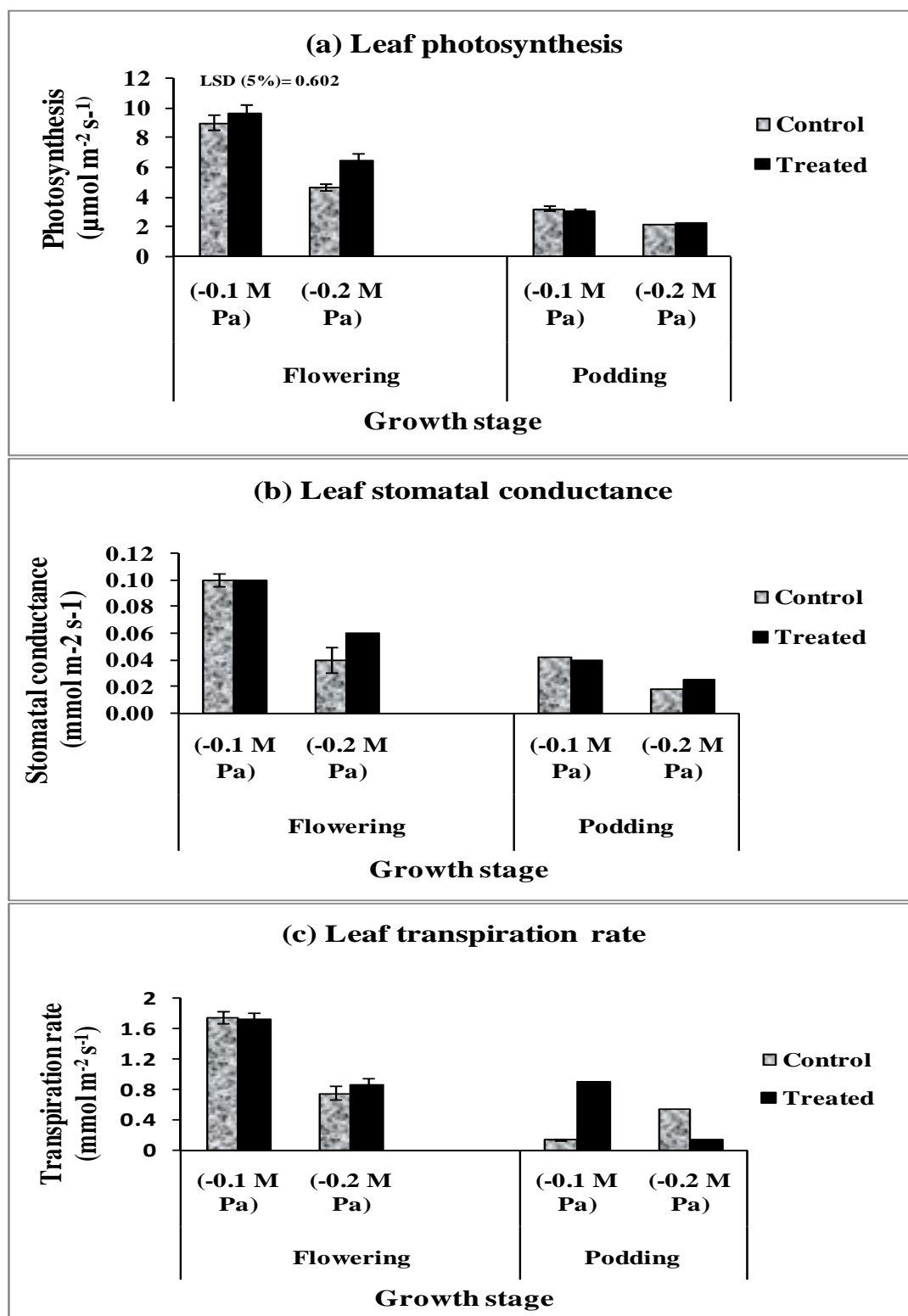
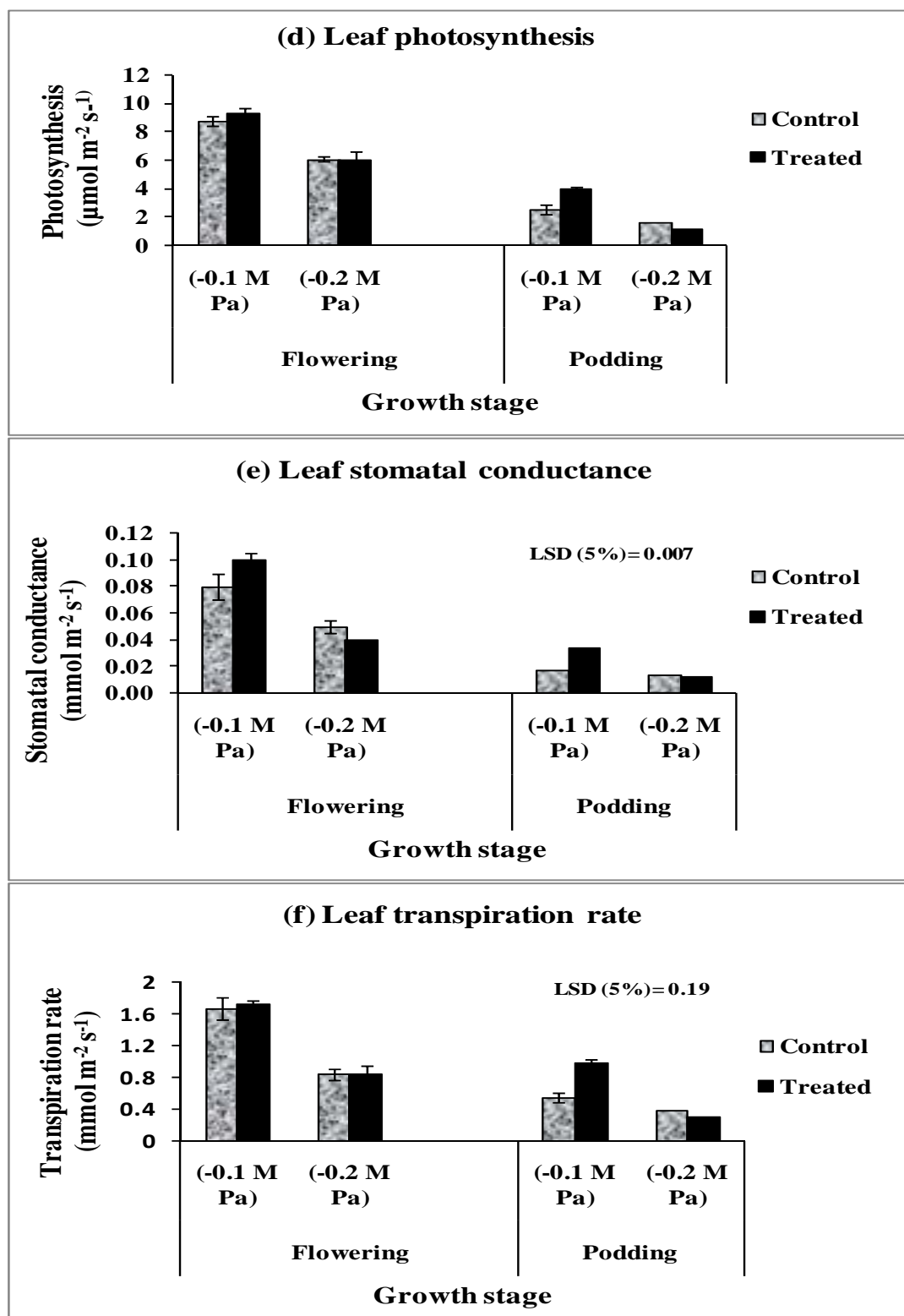
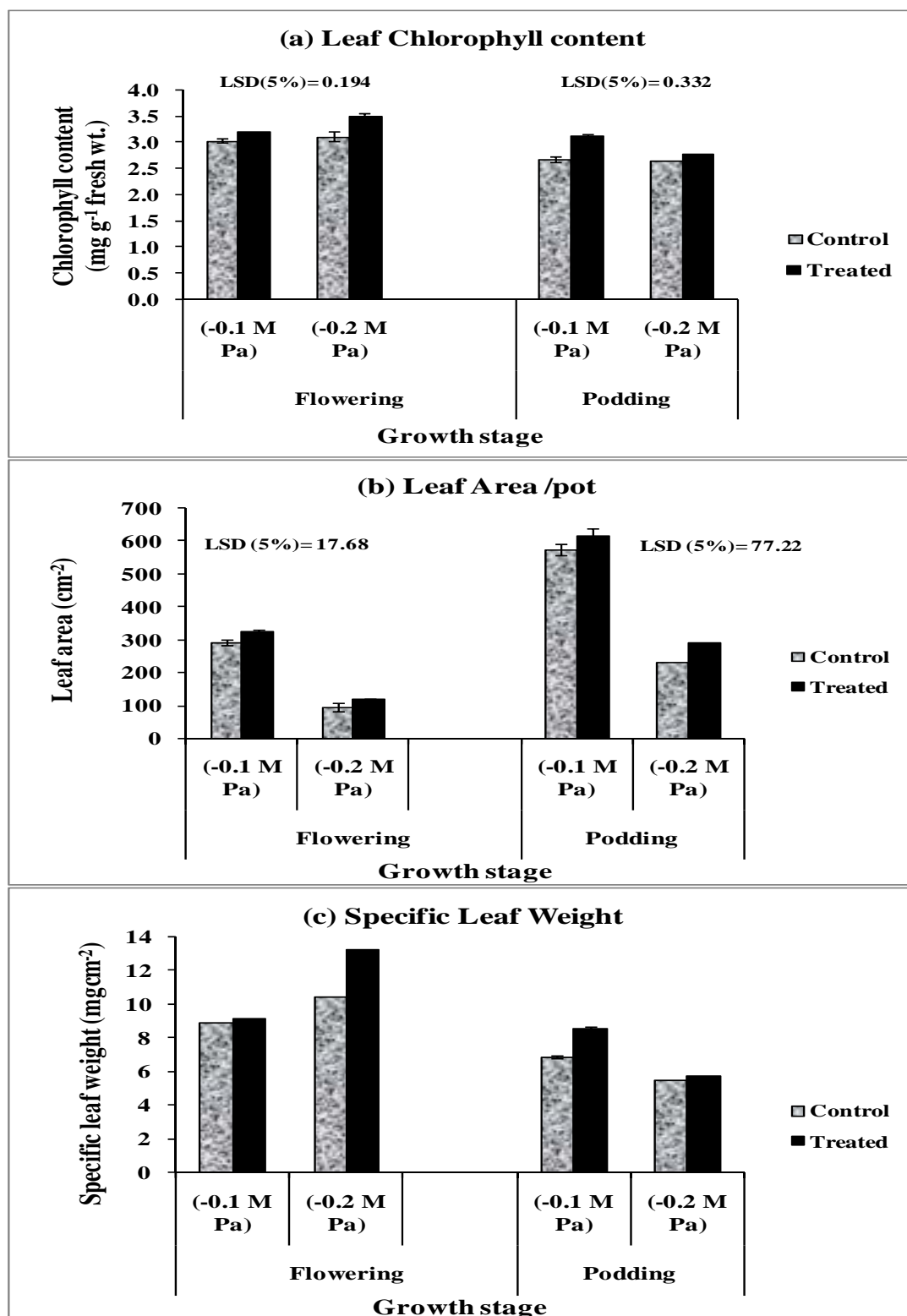


Fig.4.4.1: (a) Leaf photosynthesis, (b) Stomatal conductance and (c) Transpiration rate at two growth stages of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 grown in pots at two soil moisture potentials



**Fig.4.4.1: (d) Leaf photosynthesis, (e) Stomatal conductance and (f) Transpiration rate at two growth stages of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 256 grown in pots at two soil moisture potentials**





**Fig.4.4.2: (a) Leaf chlorophyll content, (b) Leaf area/pot and (c ) Specific leaf weight at two growth stages of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 grown in pots at two soil moisture potentials**

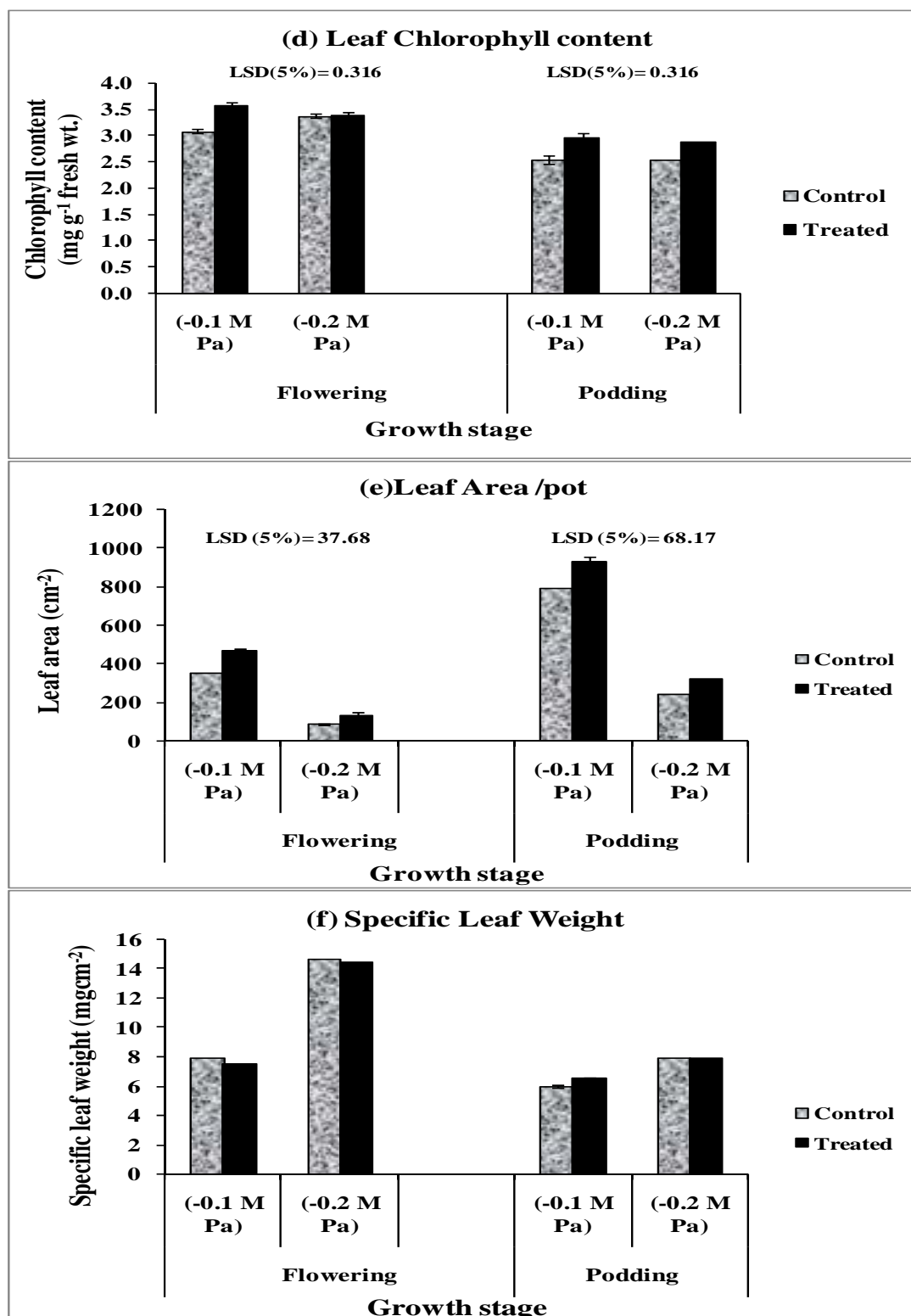
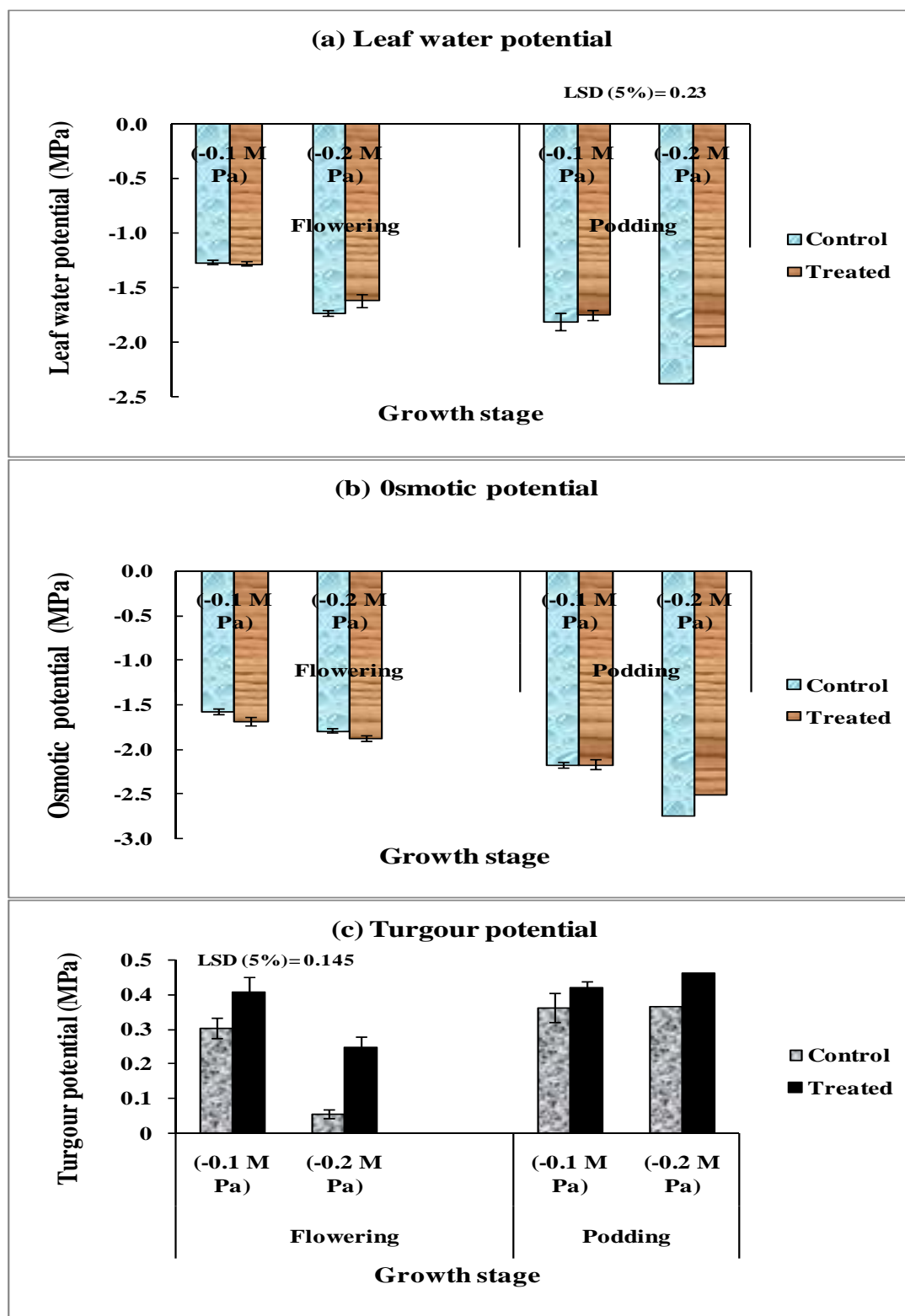
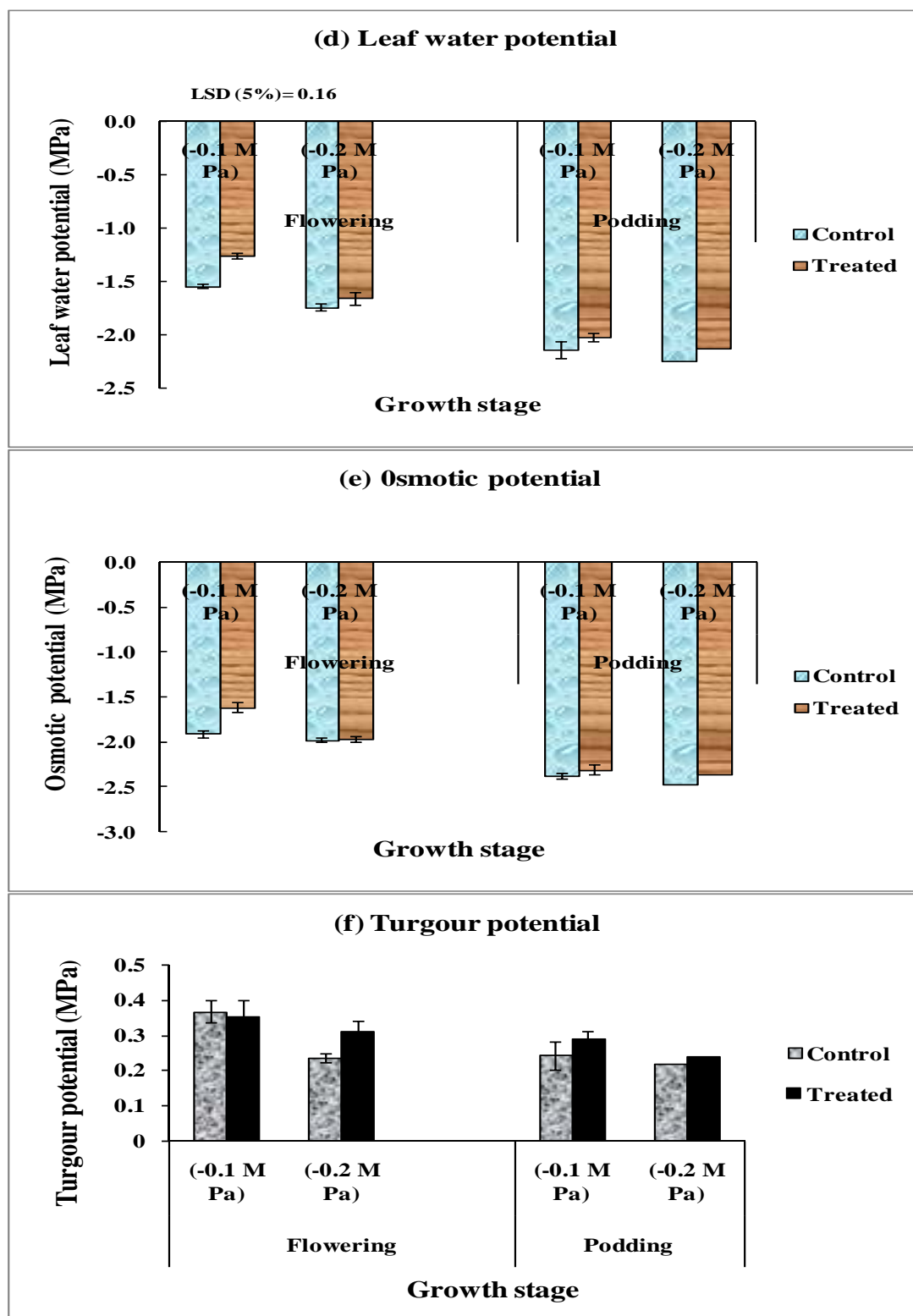


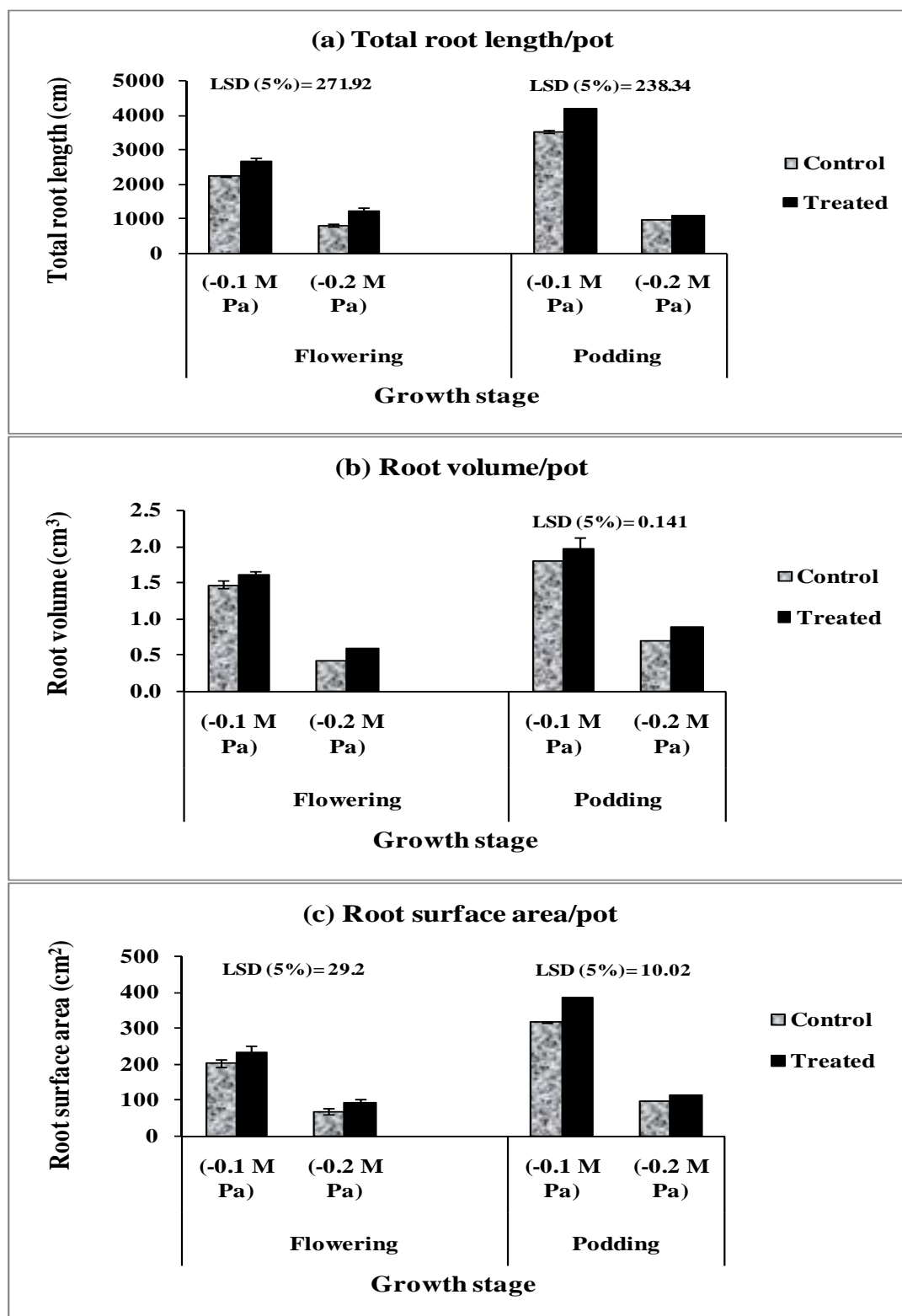
Fig.4.4.2: (d) Leaf chlorophyll content, (e) Leaf area/pot and (f) Specific leaf weight at two growth stages of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 256 grown in pots at two soil moisture potentials



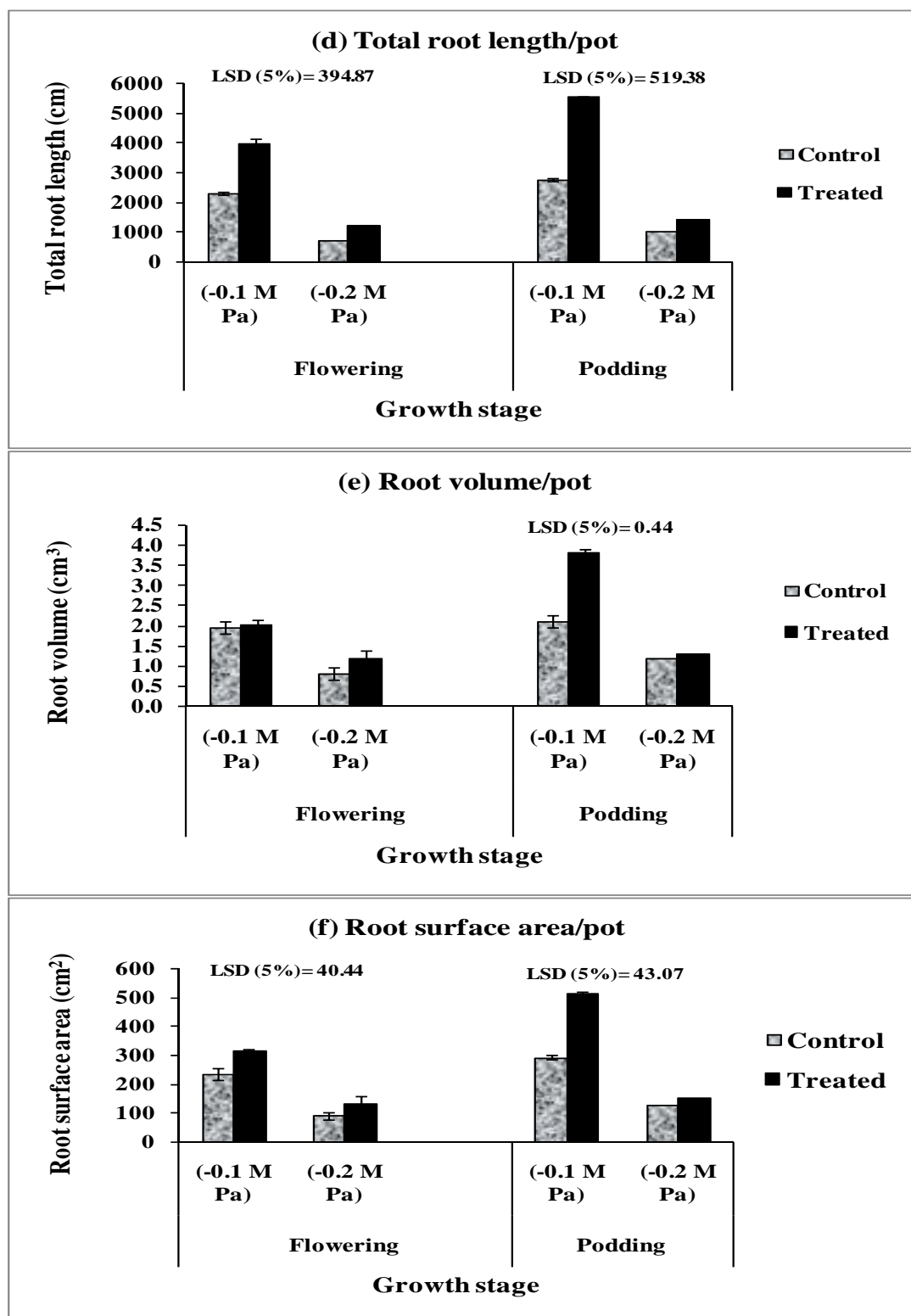
**Fig.4.4.3: (a) Leaf water potential, (b) Osmotic potential and (c) Turgour potential at two growth stages of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 grown in pots at two soil moisture potentials**



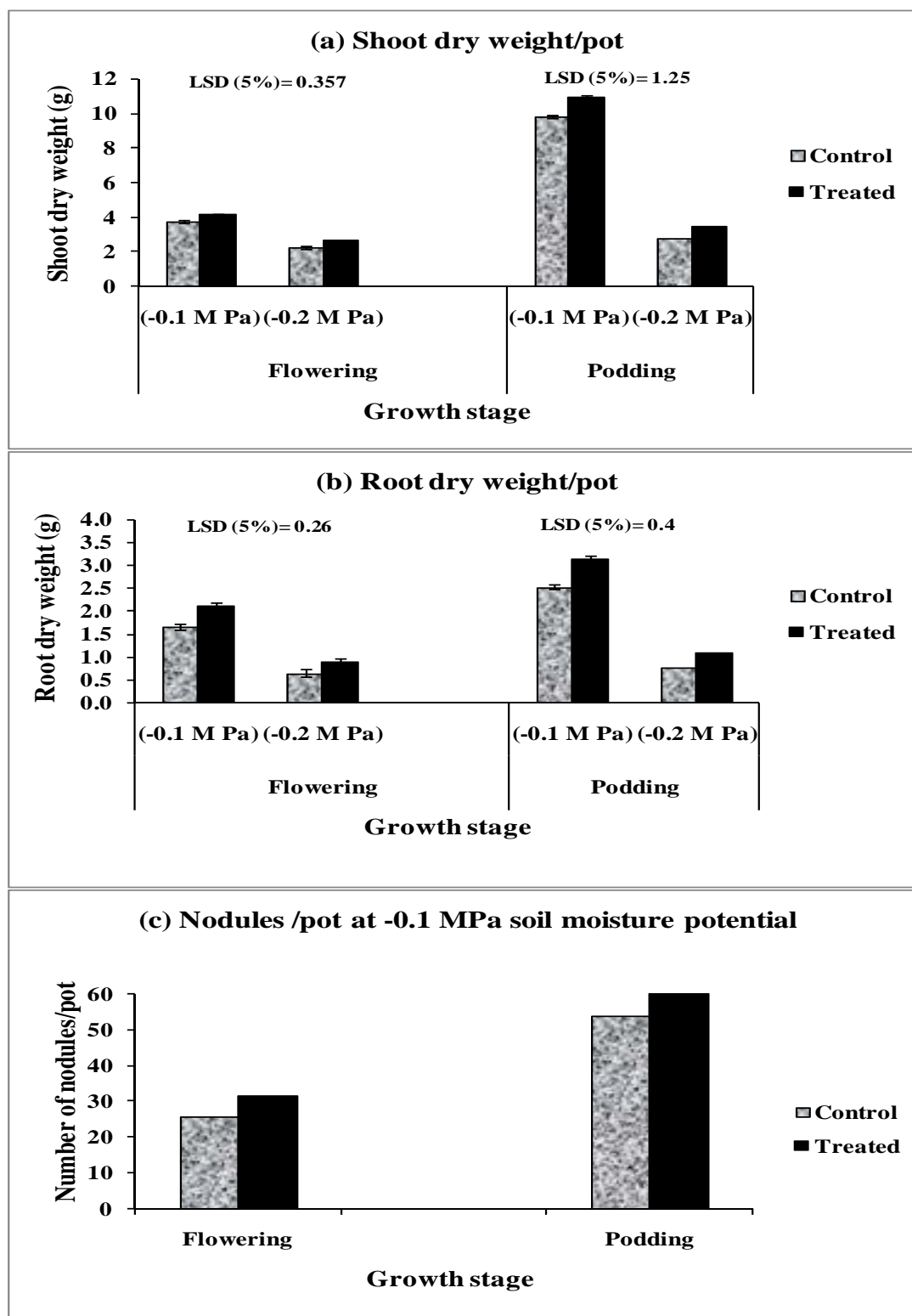
**Fig.4.4.3: (d) Leaf water potential, (e) Osmotic potential and (f) Turgour potential at two growth stages of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 256 grown in pots at two soil moisture potentials**



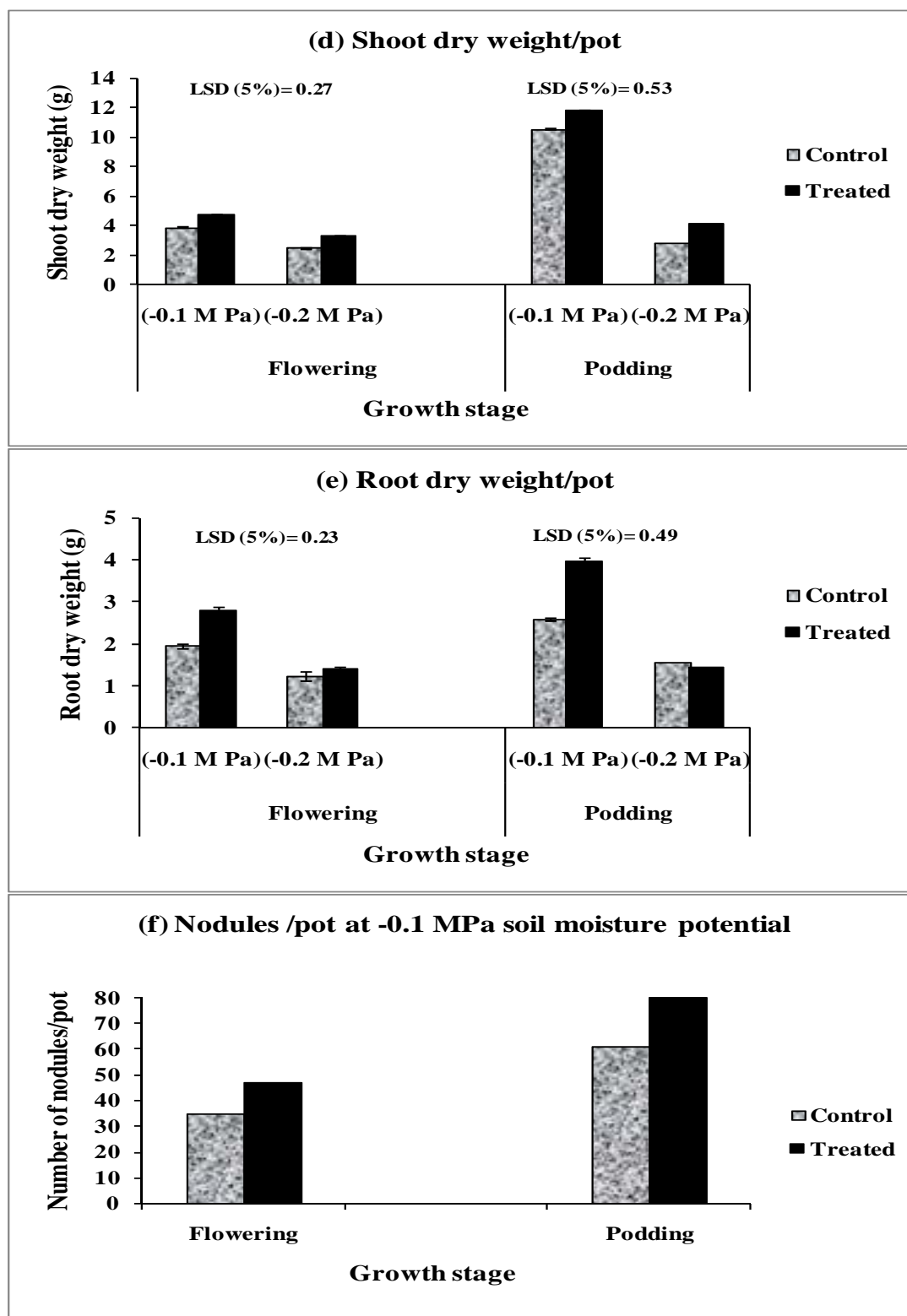
**Fig.4.4.4: (a) Total root length, (b) Root volume and (c) Root surface area/ pot at two growth stages of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 grown in pots at two soil moisture potentials**



**Fig.4.4.4: (d) Total root length, (e) Root volume and (f) Root surface area/ pot at two growth stages of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 256 grown in pots at two soil moisture potentials**



**Fig.4.4.5: (a) Shoot dry weight, (b) Root dry weight and (c) Nodule number/pot at two growth stages of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 1053 grown in pots at two soil moisture potentials**



**Fig.4.4.5: (d) Shoot dry weight, (e) Root dry weight and (f) Nodule number/pot at two growth stages of plants from magnetically exposed and unexposed seeds of chickpea var. Pusa 256 grown in pots at two soil moisture potentials**





**“Biophysical characterization of magnetic field induced enhancement in chickpea (*Cicer arietinum* L.) seeds in relation to soil moisture stress”**

**ABSTRACT**

Experiments were conducted to study the effect of pre-sowing seed exposure to static magnetic field on two chickpea varieties with a view to understand their mechanism of action in terms of seed water absorption, root dynamics and growth and yield. A desi variety, Pusa 256 and on a kabuli variety, Pusa 1053 were selected for this study. Seeds of both varieties exposed to 100 mT static magnetic field for 1h, standardized by preliminary experiments were used as the starting material for all experiments.

Temporal changes in seed water absorption in magnetically exposed seeds of both varieties had higher rate of absorption which may be attributed to magnetically induced greater seed membrane permeability to water. Seed water activity was greater for magnetically exposed seeds. This indicated that in treated seeds, not only seed water content increased, but also its corresponding water activity that resulted in early germination. Seed water diffusivity increased in general with time and in magnetically treated seeds, seed water diffusivity values were greater than untreated controls and hence better water absorption in these seeds.

Dynamics of root characters under two soil water regimes (12 & 20%) clearly demonstrated consistent increases in all parameters in treated plants over unexposed control. The relative increase over untreated control was more for Pusa 1053 than Pusa 256. Periodic measurements in root and shoot weight and the length of the longest root in the same experiment also showed similar trend.

Study conducted in pots maintained under -0.1 MPa and -0.2 MPa soil water potentials exhibited significant improvement for various physiological traits in plants grown from magnetically treated seeds and reduced significantly from flowering to podding stage and with soil moisture stress. Similar trend was observed for growth and root parameters. However, in plants from treated seeds, adverse effect of stress was ameliorated as they maintained relatively higher photosynthesis and leaf water status through osmotic adjustment and greater root length and root surface area. In both varieties, RUE and WUE decreased sharply under stress conditions. Magnetic field exposure has improved RUE and WUE over unexposed controls except for Pusa 1053 at -0.1 MPa potential. At harvest irrespective of magnetic treatment, drastic reduction in grain weight of stressed plants was observed but in well watered condition, plants from magnetically treated seeds produced more biomass, pod number and grain weight than untreated controls.

It may be concluded that exposure to static magnetic field of 100 mT for 1h improved seed water absorption characteristics in desi and kabuli varieties of chickpea that resulted in early germination and early vigour of seedlings. Improved root system coupled with superior leaf water status led to increased photosynthesis in mature plants and produced greater biomass and grain weight. Also, when these plants were subjected to severe water stress (-0.2 MPa), the adverse effect was ameliorated partially by magnetic field treatment.

## मृदा-नमी प्रतिबल के संदर्भ में चने ( *साइसर एरीटिनम* एल. ) के बीजों में चुम्बकीय क्षेत्र द्वारा प्रेरित वृद्धि का जैव-भौतिकीय अभिलक्षणन

सार

बीज द्वारा जल अवशोषण, जड़ गतिकी तथा वृद्धि एवं उपज के संदर्भ में चने की दो किस्मों के साथ बुआई-पूर्व बीज पर स्थिर चुम्बकीय क्षेत्र के प्रभाव की क्रियाविधि को समझने के उद्देश्य से प्रयोग किए गए। एक देशी किस्म पूसा 256 तथा एक काबुली किस्म पूसा 1053 का अध्ययन हेतु चयन किया गया। सभी प्रयोगों में दोनों किस्मों के बीजों को प्राथमिक प्रयोगों द्वारा मानकीकृत, आरम्भिक पदार्थ के रूप में 100 एम टी स्थिर चुम्बकीय क्षेत्र में 1 घंटे के लिए रखा गया।

दोनों किस्मों के चुम्बकीय क्षेत्र में रखे बीजों में जल-अवशोषण की दर अधिक थी, बीज द्वारा जल-अवशोषण में ये कालिक परिवर्तन चुम्बकीय क्षेत्र द्वारा प्रेरित, जल के लिए बीज-झिल्ली की पारगम्यता बढ़ जाने के कारण हो सकते हैं। चुम्बकीय क्षेत्र में रखे बीजों में बीज-जल सक्रियता अधिक थी। यह दर्शाता है कि उपचारित बीजों में न केवल बीज-जल अंश बढ़ा बल्कि अधिक जल सक्रियता के फलस्वरूप अंकुरण भी अगेता हुआ। सामान्य रूप से, समय के साथ बीज जल विसरणशीलता बढ़ गई और चुम्बकीय रूप से उपचारित बीजों में अनुपचारित बीजों की तुलना में बीज जल विसरणशीलता मान अधिक थे और इसलिए इन बीजों में जल अवशोषण बेहतर हुआ।

दो मृदा जल प्रक्षेत्रों (12 एवं 20%) में जड़ गुणों की गतिकी ने स्पष्ट रूप से दर्शाया कि अनुपचारित कंट्रोल की तुलना में उपचारित पौधों के सभी प्राचलों में सतत रूप से वृद्धि हुई अनुपचारित कंट्रोल की तुलना में पूसा 256 की अपेक्षा, पूसा 1053 में आपेक्षिक वृद्धि अधिक पाई गई। उसी प्रयोग में जड़ एवं प्ररोह के भार एवं जड़ की सर्वाधिक लम्बाई के समय-समय पर आमापन भी यही ट्रेंड दर्शाते हैं।

-0.1 एम पी ए एवं -0.2 एम पी ए मृदा-जल विभवों पर अनुरक्षित गमलों में चुम्बकीय रूप से उपचारित बीजों पर किए गए अध्ययन दर्शाते हैं कि कई पादपकार्यिकीय गुणों में मत्वपूर्ण रूप से सुधार हुआ तथा मृदा नमी प्रतिबल के साथ पुष्पन अवस्था से लेकर फलियाँ बनने की अवधि में महत्वपूर्ण रूप से कमी आई। वृद्धि एवं जड़ प्राचलों में भी इसी प्रकार का ट्रेंड देखा गया। वैसे उपचारित बीजों से उगाए गए पौधों में प्रतिबल के प्रतिकुल प्रभाव में सुधार हुआ क्योंकि ऐसे पौधों में आपेक्षित रूप से प्रकाशसंश्लेषण अधिक हुआ और परासरण समायोजन के माध्यम से पत्तियों में जल-स्तर अधिक बना रहा तथा साथ ही जड़ की लम्बाई एवं जड़-सतह क्षेत्रफल भी अधिक रहा। दोनों ही किस्मों में प्रतिबल अवस्थाओं में विकिरण उपयोग क्षमता (वि. उ. क्षमता) एवं जल उपयोग क्षमता (ज. उ. क्षमता) तेजी से कम हुए। -0.1 एम पी ए विभव र पूसा 1053 को छोड़कर अनुपचारित कंट्रोल की तुलना में चुम्बकीय क्षेत्र में रखने से विकिरण उपयोग क्षमता (वि. उ. क्षमता)

एवं जल उपयोग क्षमता (ज. उ. क्षमता) में सुधार हुआ। कटाई के समय चुम्बकीय उपचार को ध्यान में न रखते हुए प्रतिबल के साथ उगने वाले सभी पौधों के बीज-भार में अत्यंत कमी देखी गई किन्तु भली-भाँति सिंचित अवस्था में उगे पौधों में चुम्बकीय उपचार किए गए बीजों से उगे पौधों से अनुपचारित कंट्रोल की तुलना में अधिक जैवमात्रा अधिक संख्या में फलियाँ और अधिक बीज भार प्राप्त हुए।

इन प्रयोगों से यह निष्कर्ष निकलता है कि 100 एम टी पर 1 घंटे, स्थिर चुम्बकीय क्षेत्र में उपचारित करने से चने की देशी एवं काबुली किस्मों के बीज-जल अवशोषण गुणों में सुधार होता है जिसके परिणामस्वरूप अंकुरण जल्दी होता है एवं नवोद्भिद अधिक ओजस्वी होते हैं। बेहतर जड़-तंत्र और साथ ही बेहतर पर्ण-जल स्तर होने से वयस्क पौधों में प्रकाश संश्लेषण अधिक होता है तथा परिणामस्वरूप अधिक जैवमात्रा एवं बीज-भार उत्पन्न होते हैं। इसके साथ ही, जब इन पौधों को गंभीर जल प्रतिबल ( $-0.2$  एम पी ए) का सामना करना पड़ता है तो चुम्बकीय क्षेत्र उपचार से आंशिक रूप से दुष्प्रभाव में सुधार हुआ।