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Effect of Magnetic Treatment on Enzyme Activation of Paddy (*Oryza sativa* L.)

Y.M. Yadav, S.G. Mahadik*, V.V. Dalvi, A.A. Deogirikar, M.M. Burondkar and P.B. Vanave

Dr. Balasaheb Sawant Konkan Krishi Vidyapeeth, Dapoli 415712, Dist. Ratnagiri, India

**Corresponding author*

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A B S T R A C T

A study was undertaken to investigate the effect of magnetic treatment on enzyme activation of paddy seed under laboratory and nursery conditions. A magnetic seed treater having aluminum container fitted with ten 250 gauss magnets on copper pipes in staggered fashion was used. The container could be rotated around the horizontal axis. The dry seeds were treated by putting the seeds in the container and rotated the container for different time periods (15 min without magnet (control) and 15, 30, 45, 60 and 90 min with magnet). In the germinating seeds, enzyme activities of α amylase, β amylase, catalase, and peroxidase were significantly higher in treated seeds as compared to control seeds. Results indicated that α and β amylase activity found higher in treatment T_1 (Rotation without magnet for 15 min). Catalase, and peroxidase activities found higher in T_5 (rotation with magnet 60 min), T_3 (rotation with magnet 30 min) and T_3 (rotation without magnet 30 min.) respectively.

Introduction

All organisms during their life are faced with two types of magnetic field (MF), one of them is natural magnetic field that is the result of earth magnetic field and the amount is between 0.03 and 0.07 mT, another is artificial magnetic field which is the result of application of electrical power at homes and industrial workshops. Magnetic field which is produced in a wide range has positive and negative effects on animal and plant life. Today an important question is if magnetic field has any distinctive effect on biological systems. There are reports about the short-term MF exposure. MF influences a variety of plant functions such as growth (Racuciu *et al.*, 2007), development (Yano *et al.*, 2004; Rakosy Tican *et al.*, 2005), protein biosynthesis and enzyme activity (Alikamanoglu *et al.*, 2011). But the interaction of such fields with the living cells is still unclear (Atak, 2007). MF causes an oxidative stress, that is, increases the activity, concentration, and lifetime of free radicals which are highly reactive byproducts of normal metabolism and immune defense (Scaiano, 1994). Accumulation of reactive oxygen species which are generated during stress can harm many cellular components such as lipids, proteins, carbohydrates and nucleic acids. Two wheat cultivars treated

with the magnetic field showed better germination results (Gholami and Sharafi, 2010). Seeds treated by magnetic stimulation seem to show higher enzyme activities which control the particular stages of seed germination (Vashisth and Nagarajan, 2010). It was also shown that the magnetic field improved cell division, prolongation and cell differentiation and influences most of the chemical factors involved in germination (Dao-lian *et al.,* 2009). Therefore, MF would change the antioxidant enzyme activity (Sahebjamei *et al.,* 2007; Alikamanoglu *et al.,* 2011). The purpose of this study was to investigate effect of MF on the enzyme activation of paddy during the various stages of growth.

Materials and Methods

The experiment was conducted at the Research Farm, Department of Botany, College of Agriculture, Dapoli, Dist. Ratnagiri, Maharashtra state during the *Kharif* 2014. The selection of site was considered on the basis of suitability of the land for cultivation of rice. The soil of experimental field was lateritic type slightly acidic. The seed of rice variety Ratnagiri-24 was used for this study as it is popular and fine commercial variety. The magnetic seed treater having a aluminum container of 5l volume capacity with 10 magnets of 250 gauss fitted in it on the copper pipes in the crisscross manner. The container is mounted on a horizontal shaft of a frame such that it could be rotated by handle around the horizontal axis. The magnetic treatment was given to the seeds by putting it in the container and rotating it at about 60 revolutions per minute for different durations like 15, 30, 45, 60 min. The experimental treatment details are as below.

Treated seeds were immediately sown in field on raise bed for growing seedlings. The seedlings of 21 days were transplanted in the field in three replications. The seven treatments mentioned above were transplanted at spacing of 20 × 15 cm. The plot size for each treatment was 3.0 × 3.0 m. All the recommended cultivation practices including nutrient management was practiced during the growth of crop.

The experiment was conducted in randomized block design with three replication. The observation were recorded at 30, 50, 70 DAS and at harvest. The following methods were followed for the estimation of α –β amylase, catalase, nitrate reductase and peroxidase activity.

Alpha and beta amylase activity

The activity of alpha amylase was measured in reaction mixture containing starch solution, calcium acetate buffer, (pH 6.0) and enzyme extract similar for beta except buffer which is sodium acetate (0.1M.pH 3.6).

The activity of enzyme calculated from amount of starch hydrolysed read at 610 nm (Louis and Gifford, 1962).

Catalase activity

The activity of catalase was measured in a reaction mixture consisting of a tris-Glycine buffer (50 mM, pH 7.5), H_2O_2 (10 mM) and enzyme extract. The decomposition of H_2O_2 was followed by the decline in absorbance at 240 nm by a spectrophotometer (Pereira *et al.,* 2002).

Peroxidase activity

The peroxidase activity was measured in a reaction mixture containing acetate buffer (0.2mM, pH4.8), hydrogen peroxide (0.1mM), benzidine (0.04M) and enzyme extract. Enzyme activity was measured by a spectrophotometer at 530 nm (Koroi, 1989).

Results and Discussion

α amylase activity

In the present study, significant differences were observed in all treatments of the magnetic field with respect to the α amylase activity at all stages of crop i.e. at 30 DAS (126.67 to 169.67 µg/min), 50 DAS (128.33 to 193 µg/min), 70 DAS (115 to 187 µg/min) and at harvest (61.67 to 97.00 µg/min). In all treatments the treatment T_2 (169.67 µg/min), T_1 (193.33 µg/min) and T_1 (187.00 µg/min) showed the maximum α amylase activity at 30, 50 and 70 DAS respectively. Among all the treatments, the highest α amylase activity was observed in treatment T_2 (97 µg/min) which was at par with T_4 (96.33 µg/min) and T_5 (95.67 µg/min) and lowest α amylase activity was found in treatment T_0 (61.67 µg/min) at harvest stage. In general α amylase activities increased upto 50 DAS and then decreased

Considering the overall mean value of α amylase activity at different stages of growth, treatment T_1 (160.17 µg/min) showed maximum α amylase activity. The treatment T_0 showed minimum α amylase activity (107.92 µg/min) at different stages of growth (Table 1 and Fig. 1). The similar results were obtained from studying on *Satureia hortensis* by Pourakbar and Hatami *(2012*).

β amylase activity

There was a considerable variability amongst the treatments for β amylase activity at various stages of growth. It is evident from the data that β amylase activity decreased with the advancing age of the crop. The highest β amylase activity i.e. 163, 184.67 and 185 µg/min was found in treatment T_1 at 30, 50 and 70 DAS respectively. At 30 DAS, the treatment T_1 (163 µg/min) was at par with T_4 (161.67 µg/min), T_5 and T_6 (160.67 µg/min). The lowest β amylase activity was found in treatment T_0 at all stages of growth. At harvest, treatment T_2 (97.67 µg/min) recorded highest β amylase activity which was at par with treatment T_4 and T_1 (96 µg/min). The treatment T_1 (62.33 µg/min) showed maximum mean β amylase activity. There was no much difference in mean β amylase activity in treatment T_2, T_3, T_4 and T_5. The lowest mean β amylase activity recorded in treatment T_0 (110.25 µg/min) followed by treatment T_6 (122.0 9µg/min) (Table 1 and Fig. 2).

Catalase activity

Data indicated that there were significant differences amongst the treatment for catalase activity at various growth stages. The mean values were recorded 2.45, 2.28, 1.44 and 2.18 µmol/min at 30, 50,70 DAS and at harvest which showed the decreasing trend up to 70 DAS while it was increased at harvest stage of growth. The highest overall mean catalase activity was observed in treatment T_6 (2.47µmol/min) followed by treatment T_1 (2.42 µmol/min) whereas least mean catalase activity was observed in treatment T_0 (Control) (1.31 µmol/min).

At 30 DAS, the maximum catalase activity was observed by treatment T_2 (3.11µmol/min) which was at par with treatment T_3 (3.02 µmol/min). The lowest catalase activity was noted in treatment T_0 (1.38µmol/min).

The highest catalase activity was found in treatment T_2 (2.53 µmol/min) at 50 DAS which was superior over all treatments except treatment T_4 (2.47µmol/min). The lowest catalase activity was found in treatments T_6, T_5 and T_0 (2.10µmol/min).

At 70 DAS, significantly highest catalase activity was noticed in treatment T_5 (1.77µmol/min) followed by T_1 (1.57 µmol/min), T_3 and T_4 (1.53µmol/min).

Fig.1 Effect of magnetic seed treatment on α amylase activity

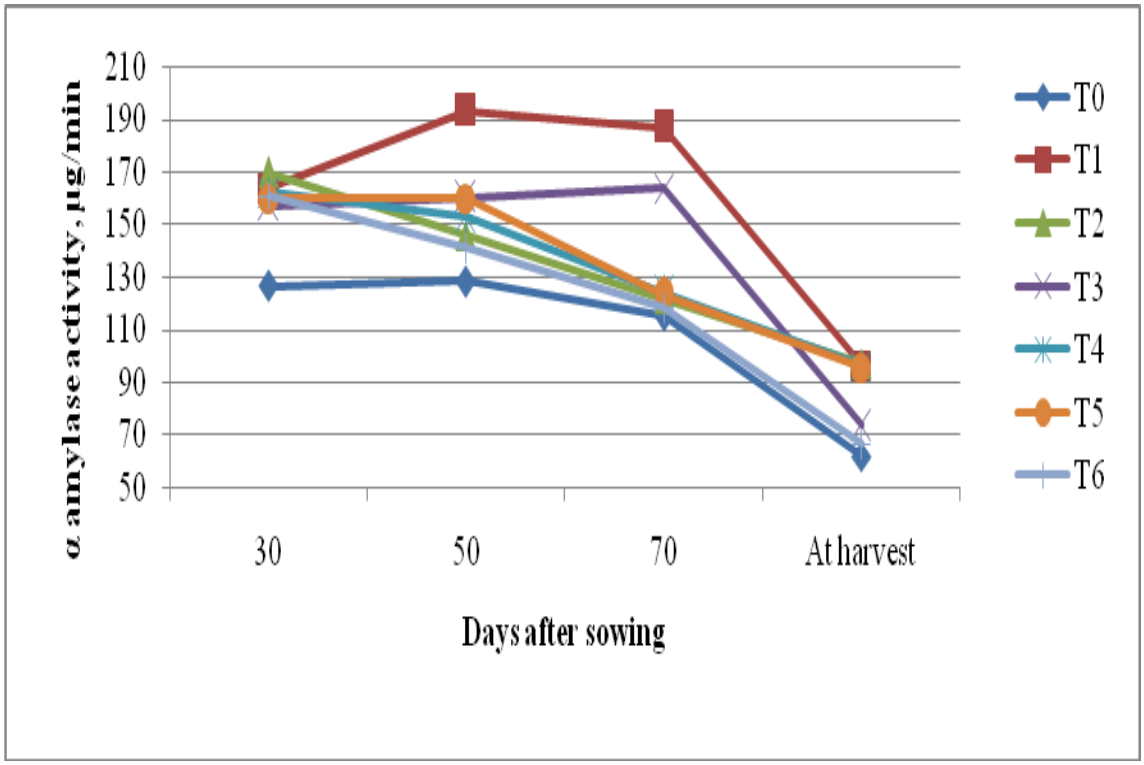

Fig.2 Effect of magnetic seed treatment on β amylase activity

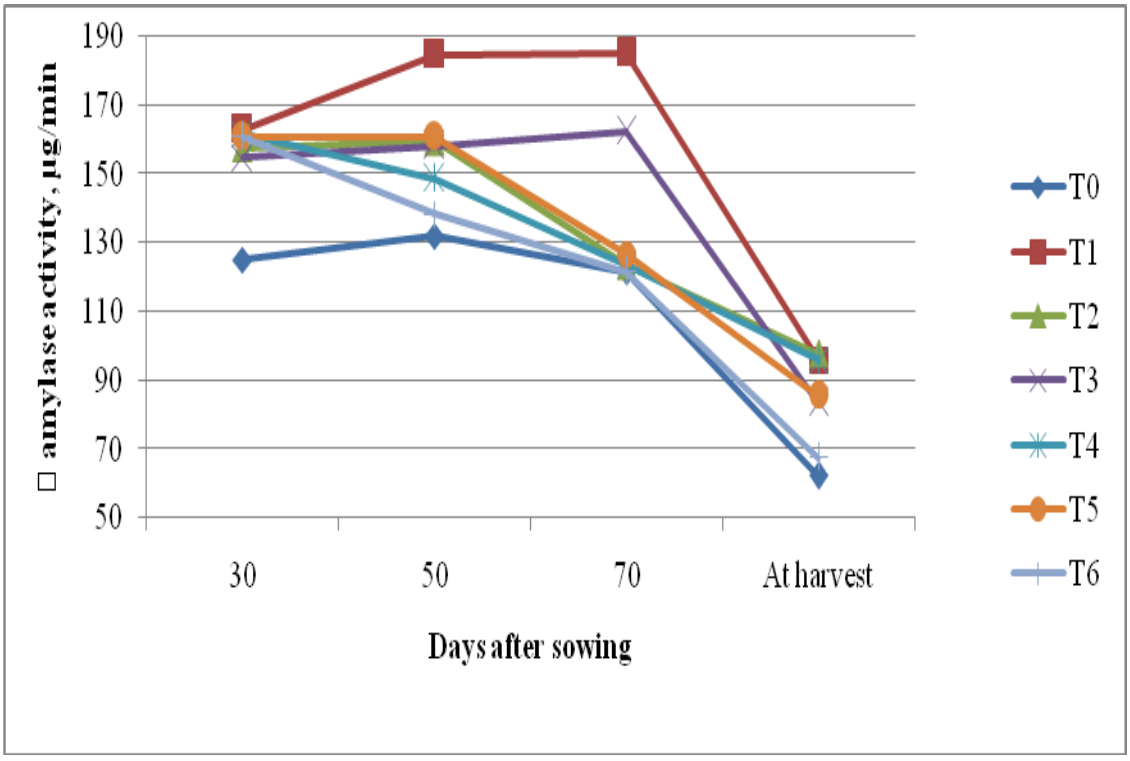

Fig.3 Effect of magnetic seed treatment on catalase activity

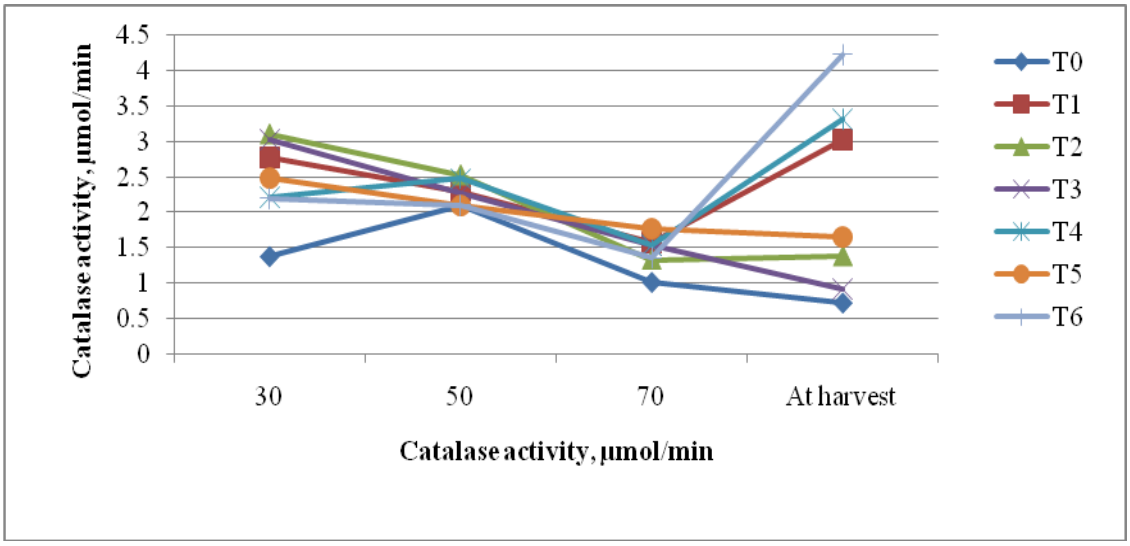

Fig.4 Effect of magnetic seed treatment on peroxidase activity

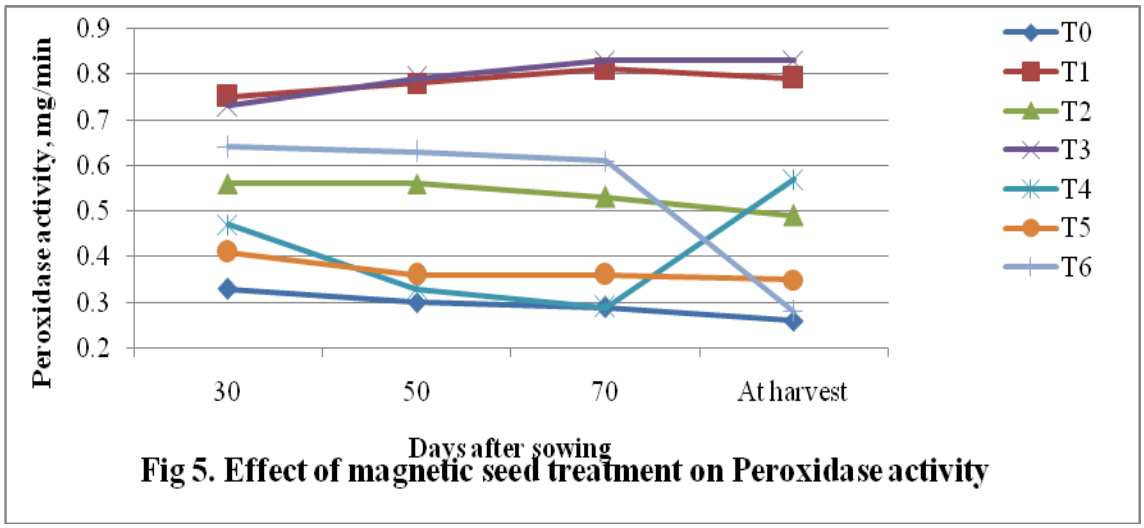

Fig 5. Effect of magnetic seed treatment on Peroxidase activity

Experimental treatment details of magnetic seed treatments

Treatment details	Symbol
Control	T_0
Rotation without magnet for 15 minutes	T_1
Rotation with magnet for 15 minutes	T_2
Rotation with magnet for 30 minutes	T_3
Rotation with magnet for 45 minutes	T_4
Rotation with magnet for 60 minutes	T_5
Rotation with magnet for 90 minutes	T_6

Table.1 Effect of magnetic seed treatment on α amylase activity and β amylase activity

Treatments	α amylase activity, µg/min					β amylase activity, µg/min				
	Days after sowing					Days after sowing				
	30	50	70	At harvest	Mean	30	50	70	At harvest	Mean
T0	126.67	128.33	115.00	61.67	107.92	125.00	132.00	121.67	62.33	110.25
T1	164.00	193.33	187.00	96.33	160.17	163.00	184.67	185.00	96.00	157.17
T2	169.67	145.67	121.67	97.00	133.50	157.33	159.33	123.33	97.67	134.42
T3	157.00	160.33	164.00	73.33	138.67	154.67	158.00	162.33	83.33	139.58
T4	162.33	152.67	124.33	96.33	133.92	161.67	148.67	123.33	96.00	132.42
T5	160.00	160.00	123.33	95.67	134.75	160.67	160.67	126.67	85.67	133.42
T6	160.67	141.00	118.33	66.33	121.58	160.67	138.33	121.68	67.67	122.09
Mean	157.19	154.47	136.23	83.80	132.92	154.71	154.52	131.71	84.10	131.26
S.Em±	2.45	2.37	2.21	1.10		1.95	2.54	2.18	1.07	
CD at 5%	7.35	7.12	6.64	3.31		5.85	7.63	6.55	3.21	

Table.2 Effect of magnetic seed treatment on catalase activity and peroxidase activity

Treatments	Catalase activity, µmol/min					Peroxidase activity, mg/min				
	Days after sowing					Days after sowing				
	30	50	70	At harvest	Mean	30	50	70	At harvest	Mean
T0	1.38	2.10	1.02	0.73	1.31	0.33	0.30	0.29	0.26	0.30
T1	2.77	2.30	1.57	3.03	2.42	0.75	0.78	0.81	0.79	0.78
T2	3.11	2.53	1.33	1.38	2.09	0.56	0.56	0.53	0.49	0.54
T3	3.02	2.27	1.53	0.92	1.94	0.73	0.79	0.83	0.83	0.80
T4	2.21	2.47	1.53	3.31	2.38	0.47	0.33	0.29	0.57	0.42
T5	2.48	2.10	1.77	1.65	2.00	0.41	0.36	0.36	0.35	0.37
T6	2.20	2.10	1.36	4.23	2.47	0.64	0.63	0.61	0.28	0.54
Mean	2.45	2.28	1.44	2.18	2.09	0.56	0.54	0.53	0.51	0.54
S.Em±	0.062	0.062	0.16	0.30		0.03	0.011	0.014	0.016	
CD at 5%	0.19	0.19	0.48	0.90		0.11	0.035	0.042	0.050	

Table.3 Effect of magnetic seed treatment on α amylase activity, β amylase activity and catalase activity

Treatments	α amylase activity, µg/min					β amylase activity, µg/min					Catalase activity, µmol/min				
	Days after sowing					Days after sowing					Days after sowing				
	30	50	70	At harvest	Mean	30	50	70	At harvest	Mean	30	50	70	At harvest	Mean
T0	126.67	128.33	115.00	61.67	107.92	125.00	132.00	121.67	62.33	110.25	1.38	2.10	1.02	0.73	1.31
T1	164.00	193.33	187.00	96.33	160.17	163.00	184.67	185.00	96.00	157.17	2.77	2.30	1.57	3.03	2.42
T2	169.67	145.67	121.67	97.00	133.50	157.33	159.33	123.33	97.67	134.42	3.11	2.53	1.33	1.38	2.09
T3	157.00	160.33	164.00	73.33	138.67	154.67	158.00	162.33	83.33	139.58	3.02	2.27	1.53	0.92	1.94
T4	162.33	152.67	124.33	96.33	133.92	161.67	148.67	123.33	96.00	132.42	2.21	2.47	1.53	3.31	2.38
T5	160.00	160.00	123.33	95.67	134.75	160.67	160.67	126.67	85.67	133.42	2.48	2.10	1.77	1.65	2.00
T6	160.67	141.00	118.33	66.33	121.58	160.67	138.33	121.68	67.67	122.09	2.20	2.10	1.36	4.23	2.47
Mean	157.19	154.47	136.23	83.80	132.92	154.71	154.52	131.71	84.10	131.26	2.45	2.28	1.44	2.18	2.09
S.Em±	2.45	2.37	2.21	1.10		1.95	2.54	2.18	1.07		0.062	0.062	0.16	0.30	
CD at 5%	7.35	7.12	6.64	3.31		5.85	7.63	6.55	3.21		0.19	0.19	0.48	0.90	

Table.4 Effect of magnetic seed treatment on nitrate reductase activity and peroxidase activity

Treatments	Nitrate reductase activity, mg/min					Peroxidase activity, mg/min				
	Days after sowing					Days after sowing				
	30	50	70	At harvest	Mean	30	50	70	At harvest	Mean
T0	0.07	0.08	0.11	0.05	0.08	0.33	0.30	0.29	0.26	0.30
T1	0.14	0.16	0.19	0.12	0.15	0.75	0.78	0.81	0.79	0.78
T2	0.17	0.18	0.23	0.12	0.18	0.56	0.56	0.53	0.49	0.54
T3	0.21	0.23	0.26	0.10	0.20	0.73	0.79	0.83	0.83	0.80
T4	0.12	0.13	0.22	0.09	0.14	0.47	0.33	0.29	0.57	0.42
T5	0.15	0.16	0.17	0.12	0.15	0.41	0.36	0.36	0.35	0.37
T6	0.15	0.16	0.17	0.12	0.15	0.64	0.63	0.61	0.28	0.54
Mean	0.13	0.14	0.14	0.09	0.13	0.56	0.54	0.53	0.51	0.54
S.Em±	0.14	0.15	0.19	0.10		0.03	0.011	0.014	0.016	
CD at 5%	0.010	0.006	0.009	0.009		0.11	0.035	0.042	0.050	

The lowest catalase activity was found in treatment T_0 (1.02µmol/min). At harvest, the highest catalase activity was observed in treatment T_6 (4.23 µmol/min) which was superior over all rest of the treatments. The lowest catalase activity was observed in treatment T_0 (0.73µmol/min) (Table 1 and Fig. 3). Similar results were reported in wheat by Alikamanoglue and Sen (2011).

Peroxidase activity

The data on mean peroxidase activity showed considerable variability amongst the treatments at different growth stages. The mean peroxidase activity was decreased with advanced stages of growth i.e. from 30 DAS to harvest stage. At 30 DAS, treatment T_1 showed maximum peroxidase activity (0.75 mg/min) which was at par with T_3 (0.73 mg/min) and T_6 (0.64 mg/min). The treatment T_3 showed maximum peroxidase activity (0.79, 0.83, 083 mg/min) which was at par with treatment T_1 (0.78, 0.81 and 0.79 mg/min) at 50, 70 DAS and at harvest respectively.

The mean highest peroxidase activity was noticed in treatment T_3 (0.80 mg/min) followed by treatment T_1 (0.78 mg/min). The treatment T_0 showed mimimum peroxidase activity 0.33, 0.30, 0.29 and 0.26 mg/min at 30, 50, 70 DAS and at harvest stage of growth. The peroxidase activity decreased with increase in the days after sowing till harvest stage. Javed *et al.,* (2013) also reported that the decline in peroxidase activity above 12 hrs was seen for 250mT magnetic field strength. The peroxide activity increased in all the treatment than control (T_0) (Table 2 and Fig. 4). Peroxidase activity increased in magnetic field was also reported by Pourakbar and Hatami (2012) in *Satureia hortensis*, Farzpournachaini *et al.,* (2013) in *Valeriana officiandis*, and Atak *at al.* (2007) in soybean (Table 3 and 4).

α amylase activity, β amylase activity, Catalase activity, and Peroxidase activity were highly influenced by the magnetic treatments. All these enzyme activities increased up to 50 to 70 DAS and then declined. There was no any co-linearity between time of treatment and expression of any enzyme activity.

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