



Effect of potassium and mulching ameliorants on soil enzyme activities in peanut (*Arachis hypogaea*) crop facing salinity stress

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Soil salinity has been a major threat to soil health and crop production. It often retards the soil microbial activities and hence restricts the soil nutrient availability and efficiency mediated by enzymes. Researchers around the world have been focused on this issue since salinity is not a local or regional problem, rather established as a global concern. However, some reclamation strategies are regularly suggested for minimizing the salinity stress. These measures sustain crop productivity under a particular saline soil by nullifying the excess salt ions.

Salt toxicity has many-fold negative effects on soil properties: high pH, high sodium adsorption ratio (SAR) and exchangeable sodium percentage (ESP), poor soil physical structure and low water permeability (Wong *et al.* 2008, Shukla *et al.* 2011, Singh *et al.* 2012). Moreover, salt deposition in surface soil also has unfavorable effects on soil biological parameters especially enzyme activities (Rietz and Haynes 2003, Karlen *et al.* 2008), limiting nutrient cycling and agriculture productivity as a whole (Rady 2011). Soil enzyme activities are closely linked with soil physical and chemical parameters, including soil properties, soil types and environmental conditions, and already established as important indicators of soil quality (Rietz and Haynes 2003, Yuan *et al.* 2007, Bhaduri *et al.* 2015). Therefore, any alterations happened in soil influencing soil enzyme activities is important to study (Dick and Burns 2011). Among all the enzymes, urease, alkaline phosphatase and catalase activities are more sensitive to environmental changes (Guangming *et al.* 2017).

Few studies have been undertaken in recent years suggesting that suitable management practices to curb down the salinity effect on soil microflora and their metabolic activities. Meena *et al.* (2016) concluded that integrated use of organic amendments and mineral fertilizer is beneficial for promoting MBC and enzyme activities in a mustard–pearl millet cropping system grown under saline soil, with a special mention of municipal solid waste compost. The combined effect of buried straw layer with plastic mulching has been an effective option for increasing the microbial biomass C and dissolved organic C in an arid saline soil (Huo *et al.* 2017). The role of potassium in peanut crop for minimizing the salinity effect was earlier evidenced by Chakraborty *et al.* (2016), whereas pronounced mulching effect has been observed in peanut (*Arachis hypogaea* L.) by Jain *et al.* (2017).

The majority of soil biological/biochemical parameters studied so far under imposition of salinity treatment or at the naturally saline soils primarily showed the restricted activity of enzyme concerned. But our study is one-step ahead where measures of salinity management, commonly practised for improving crop productivity, if at all impacts the soil health in terms of soil microbial activity. Hence the objectives of the study can be spelled out as: to assess the effect on four common soil enzymes at two levels of salinity, and the ameliorating effect of potassium and mulching towards soil enzymes.

A field experiment was conducted in summer, 2014 (February to May) at the research farm of ICAR-Directorate of Groundnut Research, Junagadh, India (21.52° N, 70.47° E, 100 m above MSL) having soil classified as Vertic Ustochrepts, medium black, clayey, shallow and slightly alkaline (pH 7.8–8.0) in nature. The experiment was laid out in a split-split plot design with 18 treatment combinations and three replications by using three levels of saline irrigation water (S₀: control, S₁: 2.0 and S₂: 4.0 dS m⁻¹) as main plot, two mulching treatments (M₀: without mulch, M₁: with mulch) as sub plot and three levels of potassium

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treatment (K_0 = no potassic fertilizer applied, K_1 = 30 kg K_2O ha^{-1} , K_2 = 60 kg K_2O ha^{-1}) as sub-sub plot. The size of each plot was 7 m $\times$ 6 m where peanut seeds (Variety TG 37A) were sown at a spacing of 30 cm $\times$ 10 cm. The crop was grown following standard agronomic management practices and recommended doses of N (25 kg ha^{-1}) and P (50 kg ha^{-1}) fertilizers applied to all plots at the time of sowing. During the whole cropping season the plants were irrigated with an average interval of 10 days and salinity treatment was started after 3 weeks of plants' emergence from seeds.

The S_1 (2.0 dS m^{-1}) and S_2 (4.0 dS m^{-1}) salinity levels in the irrigation water was created by dissolving required amount of commercially available sodium chloride salt (2.6 and 5.2 kg in 2000 l irrigation water to get 22 and 44 M NaCl solution, respectively which ideally represented the desired salinity levels, 2.0 and 4.0 dS m^{-1}). The irrigation water used for the present study belongs to the classes from C_1S_1 to C_2S_1 (USDA classification, Richards 1954) having no sodium hazard ($SAR < 10$) and residual sodium carbonate ($RSC < 1.00$ me/l) with moderate soluble sodium percentage.

Soil sampling (at 0-15 cm depth) was done at two stages of crop growth (50 DAS and 100 DAS) from crop rhizosphere region and moist, undisturbed soil samples was kept in refrigerator (4°C) after proper labeling for further enzymatic assays. Dehydrogenase activity (DHA) was estimated by monitoring the rate of production of triphenyl formazan (TPF) from substrate 2,3,4-tri-phenyl tetrazolium chloride (TTC) (Klein *et al.* 1971). Both acid and alkaline phosphatase activities in soil were assayed from the amount of *p*-nitrophenol released (Tabatabai and Bremner 1969). Urease activity in soil sample was estimated according to 'determination of urea remaining' methodology (Douglas and Bremner 1971). Fluorescein diacetate hydrolytic activity (FDHA) was estimated by measuring the released fluorescein from fluorescein diacetate by the action of hydrolytic enzymes in soil as per the method outlined by Green *et al.* (2006). The electrical conductivity of soil samples was measured using portable Hanna-make pH-EC meter in saturation extract with distilled water at a ratio of 1:2.5. The measured EC was converted to ECe by multiplying with suitable factor.

The analysis of variance (ANOVA) was carried out using Microsoft-Excel based software where least significant difference was computed to test the significant differences between treatment means at 5% probability ($P = 0.05$), to determine the main and interaction effects of saline irrigation, mulch and potassium treatments as applicable to split-split plot design.

Measuring electrical conductivity (in present study, it is ECe) is an integral part of any experiment where salinity is a component. In both the observation intervals (50 and 100 DAS) the treatment with mulching (M1) could significantly reduce the soil ECe at all three salinity levels (S_0 , S_1 and S_2) with respect to control, M_0 (Fig 1a, c). With positive effect of mulching, level of reduction of ECe was higher for S_2 (15%) at 50 DAS while it was maximum for S_4 (49%)

at 100 DAS. As previously reported, mulching has some beneficial impact in reducing salinity developed in soil. Both straw and plastic mulches helped to curb down the effect of salinity during sunflower growing period by reducing the salt content in the topsoil as well as altering salt distribution in the soil profile (Zhao *et al.* 2016). Similarly, Tan *et al.* (2017) also observed the reduced risk of soil salinization under film-mulched drip irrigation condition.

However, for potassium treatments the changing behavior of ECe was not as uniform as mulching treatment. At 50 DAS, K_{30} and K_{60} showed the significant drop in ECe (5.53 and 6.38 dS m^{-1} , respectively) over the control (9.93 dS m^{-1}). This is mainly because the excess of K^+ ions could replace the ample Na^+ ions at clay exchange sites and may help to reduce ECe by leaching excess Nations (Dhillon and Dhillon 1990). At later crop growth stage (100 DAS), the situation reflected in a different way since the highest ECe of respective treatment were observed at S_2 and S_4 , due to the more number of saline irrigations imposed and abundance of K^+ ions added up the measurable salinity in the form ECe. Similar effect on soil ECe was also observed by Chakraborty *et al.* (2016).

Dehydrogenase activity (DHA) showed a good response with crop growth stages, mulching and potassium doses under saline and non-saline treatments imposed under the experimental peanut crop (Fig 2). The value obtained for the enzyme was invariably higher at 50 DAS than that obtained at 100 DAS. At higher salinity level (both S_2 and S_4), mulching effect for DHA was pronounced, thus varied from 10-29% in S_2 and 8-66% in S_4 at 100 DAS soil samples, and the difference between potassium doses seemed to be nullified. Whereas at non-mulching condition potassium played an important role to reduce the salt stress created at rhizosphere, and inclined DHA value at K_{60} proved the hypothesis. However, at S_0 situation, the DHA values followed a dissimilar trend.

Mulching and potassium both, individually can be a well-opted strategy for reducing the ill-effect of salinity; however, it cannot be predicted because their combined effect can go well or not for soil enzymatic study. While reporting the present work, we even could not find a single evidence of their combined effect in any crop. Although there are few studies which revealed the combined use of straw and plastic film mulching (Li *et al.* 2016) and ridge-furrow planting method with plastic mulching (Qin *et al.* 2016) showed healthy impact on soil microbial community and crop production by alleviating salt stress. Increased DHA content in soil might be highlighted as another reflection of the sustained microbial activity under mulching at saline soil environment. In addition, potassium took care the soil biological activity (in terms of DHA) where mulching was seen absent. So it can be inferred that role of potassium and mulching could be complementary in this case.

Activities of acid and alkaline phosphatases (AcPA and AlkPA) were also studied under the present experimental setup at two different crop growth intervals (Table 1). Overall the inclined trend for AcPA was observed at 50 DAS over

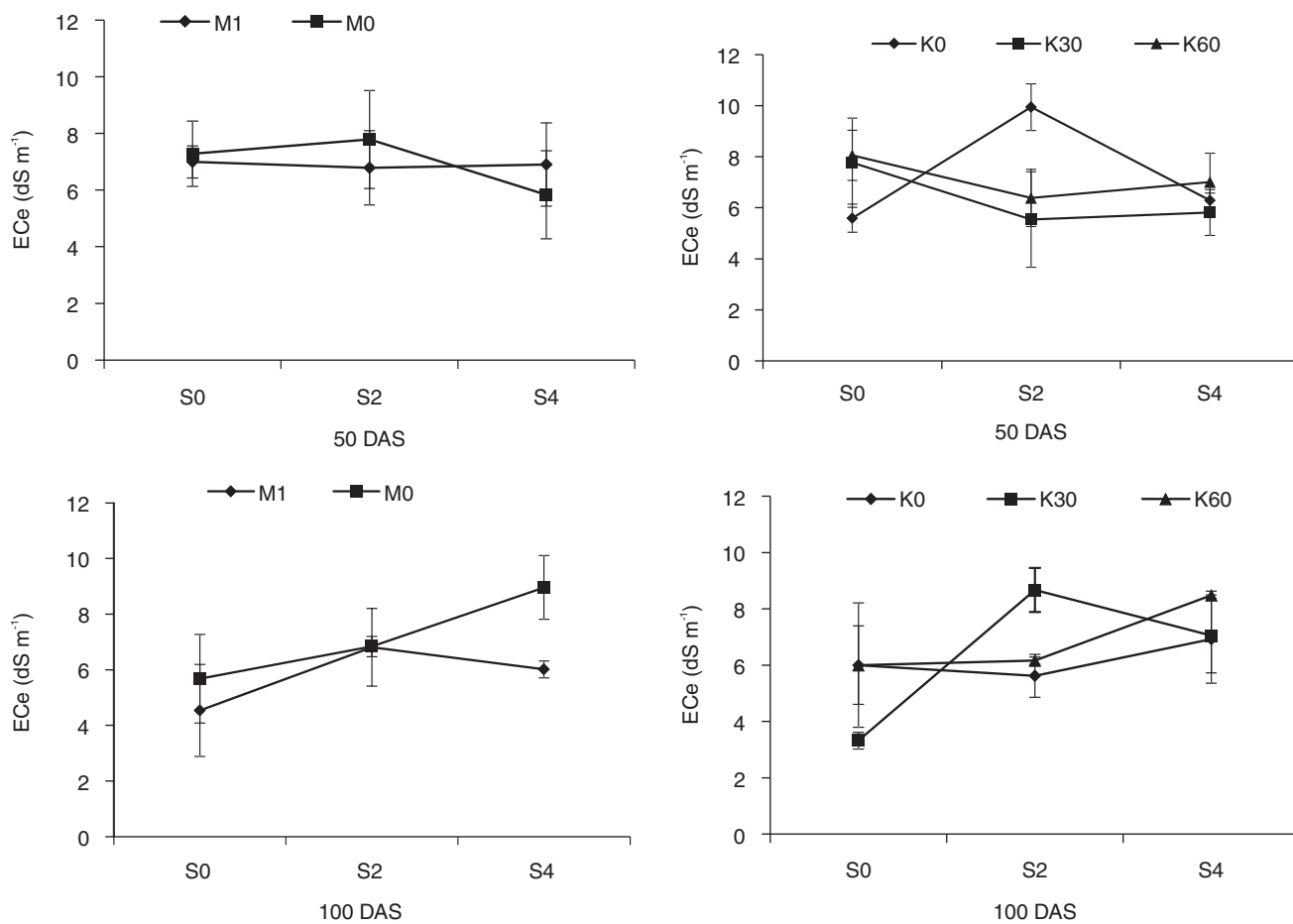


Fig 1 Measured electrical conductivity (ECe) varied with applied mulching (M) and potassium doses (K) at two crop growth stages (D) i.e. 50 DAS (a, b) and 100 DAS (c, d) under salinity treatment (S). LSD ($p < 0.05$) values obtained for main and interaction effects were 0.07 (S), 0.028 (M), 0.065 (K), 0.23 (D), 0.049 ($S \times M$), 0.092 ($M \times K$), 0.113 ($S \times K$), 0.16 ($S \times M \times K$) and 0.978 ($S \times M \times K \times D$).

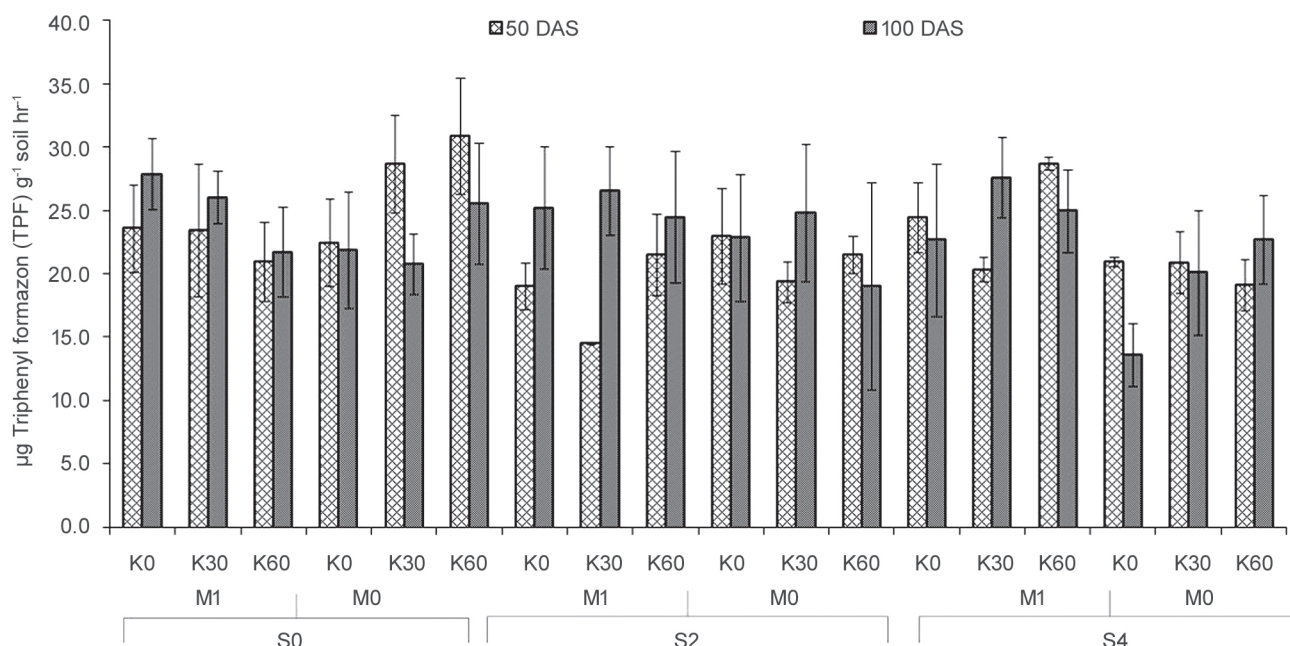


Fig 2 Effect of salinity (S), mulch (M) and potassium doses (K) on soil dehydrogenase activity at two crop growth stages (D). LSD ($p < 0.05$) values obtained for main and interaction effects were 4.98 (S), 2.12 (M), 1.46 (K), 2.95 (D), 3.66 ($S \times M$), 2.07 ($M \times K$), 2.53 ($S \times K$), 3.58 ($S \times M \times K$) and 12.53 ($S \times M \times K \times D$).

100 DAS. Both the mulching (M1) and potassium (K30 and K60) treatments had profound impact on AcPA at lower salinity level, S2 for both 50 and 100 DAS, however that seemed to disappear at higher salinity level, S4. A reverse trend was observed for AlkPA while a higher set of values was observed at later growth stage (100 DAS) than at 50 DAS. However, at S0 and S2 situations, no distinct effect of mulching and potassium was observed. At S4 in 100 DAS, the combined beneficial effect of mulching along with K30 and K60 were pointed (~31% and ~20% increases, respectively) which nullified the ill-effect of salinity on soil enzymes. It is interesting that the activities of both AcPA and AlkPA showed significant differences from one sampling time to another but did not show similar trends of minimum and maximum activity. It is therefore apparent that the activity of each enzyme is affected differently by soil and environmental factors. Temporal fluctuations

of soil enzymatic activities including phosphatase under polyethylene mulches has also been reported (Haynes 1987).

AcPA seemed to be more sensitive soil enzyme over AlkPA and that could be managed up to S2 level, while the latter was managed up to S4 treatment. Earlier study with living mulches (white clover, crown vetch and ryegrass) could improve the soil alkaline phosphatase activity levels (Qian *et al.* 2015). Whereas Singh *et al.* (2016) underlined the role of potassium in microbial and enzyme activities in saline soil. Both the management seemed to be effective in maintaining the P dynamics in saline soil by up-regulating the phosphatase activities.

Activity of urease enzyme (UA) mostly showed the trend of inclined range at 100 DAS over 50 DAS (except for few treatments) (Fig 3). Once comparing the values of UA among the treatments, at S2 condition, mulching and potassium management could not manage the adverse

Table 1 Effect of salinity, mulch and potassium doses on soil acid and alkaline phosphatase activities at two crop growth stages

Treatment			Acid phosphatase (µg PNP g ⁻¹ soil h ⁻¹)		Alkaline phosphatase (µg PNP g ⁻¹ soil h ⁻¹)	
			Days of observation (D)			
Salinity	Mulch (M)	Potassium dose (K)	50 DAS	100 DAS	50 DAS	100 DAS
S ₀	M ₁	K ₀	27.3	18.9	13.7	24.6
		K ₃₀	23.0	18.3	12.8	18.3
		K ₆₀	23.5	17.0	12.9	15.2
	M ₀	K ₀	27.3	18.2	17.0	25.1
		K ₃₀	22.6	20.1	14.4	23.8
		K ₆₀	23.5	20.5	15.6	16.7
S ₂	M ₁	K ₀	26.9	18.5	12.1	17.6
		K ₃₀	28.4	20.2	14.5	17.9
		K ₆₀	24.9	21.5	14.1	17.2
	M ₀	K ₀	23.9	20.0	12.2	23.6
		K ₃₀	24.5	15.7	12.2	14.6
		K ₆₀	20.8	18.9	17.2	19.0
S ₄	M ₁	K ₀	28.8	19.0	15.2	17.9
		K ₃₀	25.2	18.3	14.1	23.3
		K ₆₀	24.3	17.4	13.2	21.4
	M ₀	K ₀	29.2	9.5	12.9	16.6
		K ₃₀	26.2	15.9	8.6	16.6
		K ₆₀	26.0	16.1	17.5	19.1
CD (<i>P</i> <0.05)		AcPA		AlkPA		
S		2.22		1.77		
M		1.34		1.65		
K		0.93		1.48		
D		1.88		2.34		
S×M		2.32		2.85		
M×K		1.31		2.09		
S×K		1.61		2.56		
S×M×K		2.28		3.62		
S×M×K×D		7.97		9.93		

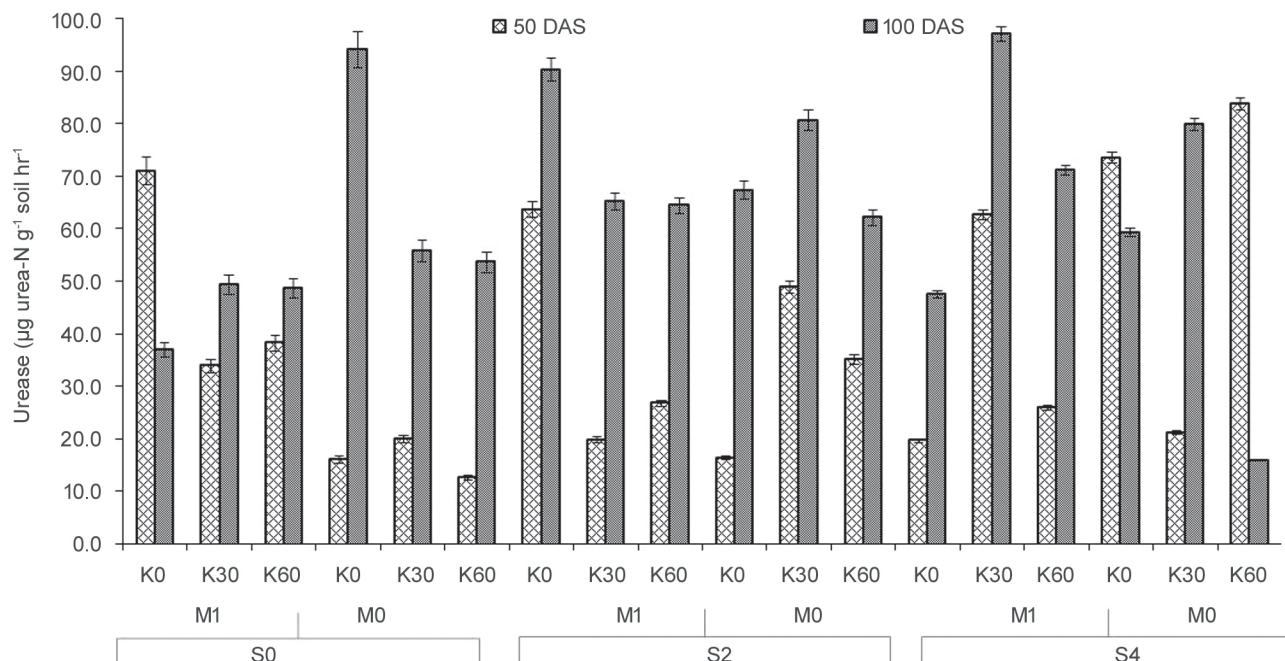


Fig 3 Effect of salinity (S), mulch (M) and potassium doses (K) on soil urease activity at two crop growth stages (D). LSD ($p < 0.05$) values obtained for main and interaction effects were 5.11 (S), 0.15 (M), 0.40 (K), 0.72 (D), 0.25 (S×M), 0.56 (M×K), 0.68 (S×K), 0.97 (S×M×K) and 3.05 (S×M×K×D).

effect of salinity at both crop growth stages; while at M0 effect of potassium (K30) was evidenced at both stages, so increased the UA from 16.6 to 49 units, and 67.5 to 81 units at 50 DAS and 100 DAS, respectively. At highest salinity level, S4, imposition of both mulching and potassium could improve the UA for both stages at K30 and K60 doses, however, M0 showed a discrete trend for UA.

Similar to phosphatases, UA also showed responsiveness

towards mulching and potassium. But that effect worked well under higher salinity. Urease activity was found to be more tolerant to salinity (Das *et al.* 2017) hence the management also played a role at higher salinity.

Fluorescein diacetate hydrolyzing activity (FDHA) is another sensitive biological indicator of soil system under differential management practices. Interestingly, FDHA could maintain its level at S2 and S4 that is comparable

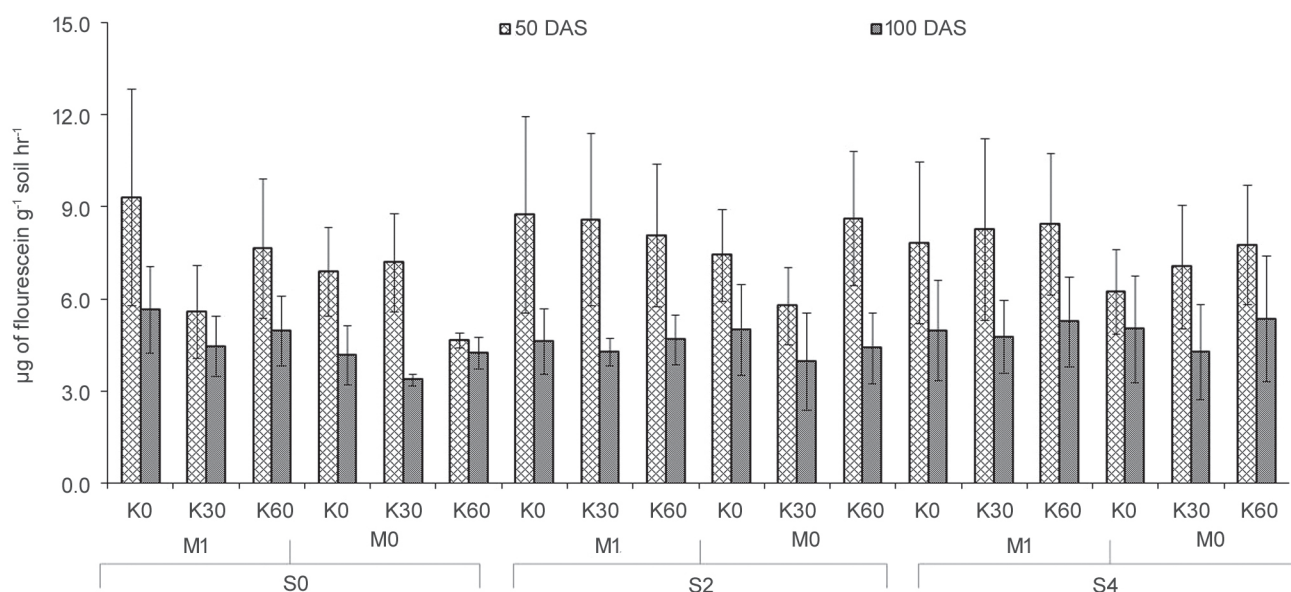


Fig 4 Effect of salinity (S), mulch (M) and potassium doses (K) on soil fluorescein diacetate hydrolyzing activity at two crop growth stages (D). LSD ($p < 0.05$) values obtained for main and interaction effects were 5.11 (S), 0.15 (M), 0.40 (K), 0.72 (D), 0.25 (S×M), 0.56 (M×K), 0.68 (S×K), 0.97 (S×M×K) and 3.05 (S×M×K×D).

or higher, as of S0 (Fig 4). Across the treatments FDHA showed inclined values at 50 DAS as compared to 100 DAS but the magnitude of difference obtained varied from treatment to treatment. The impact of mulching over potassium doses was more prominent for FDHA under saline treatments, S2 and S4, hence the values remained at par across K0, K30 and K60 within M1. However, at 50 DAS the effect of potassium was more evidenced where K60 had a clear-cut increase in FDHA values, ~8% under mulching and ~25% without mulching conditions at higher salinity level, S4. The magnitude of same increase at 100 DAS was comparatively much less.

FDHA is less studied under saline soil as compared to previously mentioned soil enzymes. As per the recent report, crop residue retention in maize-wheat system had beneficial effect on soil FDHA (Kumawat *et al.* 2017). Srivastava *et al.* (2013) also reported an almost 200% increase in FDHA activity during amelioration of wheat crop grown saline soil by using bio-augmented organic amendment in combination with half the recommended doses of chemical fertilizer. Contradictory to this, Wang *et al.* (2016) opined that plastic film mulching has reduced the soil FDHA activity irrespective of the color of mulch applied.

The present study can be summarized as: use of polythene mulch is beneficial to overcome the salinity stress imposed through varied levels of saline irrigation for regaining the normal soil microbial activity; which is further assured by the application of potassium. Again without polythene mulch, application of potassium alone showed a positive effect towards the soil enzymes under salinity vis-à-vis assuring the better nutrient availability in soil. In a nutshell, the study emphasized the role of mulching as well as potassium to reduce or nullify the effect of salinity, more specifically to soil enzymes. The mulching had its established role in regulating soil temperature and moisture regimes, and that may surely help in better hydro- and thermo- sustenance of soil microbes even under influence of salinity, which can be considered as natural soil contamination. While on the other hand, potassium fertilizer is often recommended as a useful amendment for reclaiming saline soil. In the present study, external potassium (sourced from muriate of potash) also might have created a favorable soil environment, both in terms of soil physical structure and fertility point of view. Hence both the factors played stimulants for soil enzyme activity, which has earlier been suffered by salt stress. Our findings highlighted that most enzymes showed positive impact under either mulching or potassium treatment, or both which further reiterated the role of soil enzymes in relation to microbial respiration and nutrient cycling (N, P). This management practice can be a viable option for retaining soil health for salt-affected soils belonging from arid and semi-arid zones.

SUMMARY

Salinity in agriculture creates a menace when exceeds the threshold limit, impacting heavily both soil and plants grown on it. K-fertilizer and mulching are often proposed

to control salinity. In this backdrop, our study was aimed to analyze the effect of four important soil enzymes by combined application of muriate of potash as potassium fertilizer (control, 30, 60 kg K ha⁻¹) and mulching (with and without polythene mulch) under saline irrigation (control, 2.0, 4.0 dS m⁻¹) during the active crop growth stages (50 and 100 DAS) in peanut (*Arachis hypogaea* L.) crop during summer, 2014. Results revealed that polythene mulching could effectively reduce the electrical conductivity of saturation extract of soil (ECe) across the salinity levels. The differentiating effects under mulching and potassium treatments were also observed for each of the enzymes studied. Dehydrogenase, acid phosphatase and FDH enzymes recorded more activity at 50 DAS than 100 DAS, while alkaline phosphatase and urease showed reverse trend. However, the benefits of using ameliorants like mulching and potassium fertilizer for soil biological activity under peanut crop were found to be salinity treatment specific, either at higher irrigation salinity (4.0 dS m⁻¹) or at non-saline or lower irrigation salinity (2.0 dS m⁻¹) plots. To reduce the impact of salinity both the ameliorants may or may not act jointly as observed in the present study. Overall, our study inevitably established the usefulness of mulching and potassium fertilizer for sustenance of soil biological health under salt-prone arid and semi-arid regions.

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