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Effects of climate change on plant associated microbial communities and enzyme activities

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Environmental alterations leads to fluctuations in the soil microbial population and soil enzyme activities, as different weather parameters affect microbial biota and their activities in rhizospheric soil of crop. An agroclimatic study was carried out to study the effect of environmental alterations on soil microbial population and enzyme activities in rhizospheric soil of rice and wheat crop. Rice and wheat crop were grown under field conditions and under temperature gradient tunnel which was maintained at 4-5°C higher temperature than open field temperature. Soil samples were taken from rhizospheric soil of rice and wheat. Microbial population was enumerated by serial dilution spread plating technique. Statistically significant higher microbial population of total bacteria, nitrogen fixers and P-solubilizers was found in rhizospheric soil samples taken from temperature gradient tunnel as compared to soil samples taken from rice and wheat grown under field conditions. Fungal population was found to be statistically, significantly higher in soil samples taken from field conditions in case of rice but, higher in soil samples taken from temperature gradient tunnel in case of wheat crop. Activities of alkaline phosphatase and dehydrogenase were assessed by using para nitro phenl phosphate (PNPP) and tri phenyl tetrazolium chloride (TTC) as substrates, respectively. Enzymes activities were found to be significantly higher in rhizospheric soil samples taken from temperature gradient tunnel. Microbial population and enzyme activities were found over a broad range of temperature but, maximum microbial and enzyme activity was found only at and near optimum conditions.

Key words: Environmental alterations, microbial activities, rice, temperature gradient tunnel, wheat.

INTRODUCTION

From ice ages to long warming periods, Earth's climate has changed drastically, during the planet's history. Climate change is one of the major challenges in 21st century faced by agriculture. In recent years, natural and anthropogenic factors have dramatically altered the composition of the atmosphere, which ultimately may modify long-term climatic trends (Baei and Risbey, 2009). Much of the ecosystem climate change research conducted to date has focused on macroscale responses to climatic change such as changes in plant growth, plant community composition, and coarse scale soil processes, many of which may also indirectly interact with effect of

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microbial processes. So, general concern about climate change has led to growing interest in the responses of soil microbial communities and their activities to altered environmental conditions (Castro et al., 2010).

Soil microbial communities are an integral component of many ecosystem processes and are responsible for the cycling of carbon and nutrients in the ecosystem (Jackson et al., 2007). Activities of these microorganisms are regulated by various biotic and abiotic factors such as quantity and quality of litter inputs, temperature, moisture, etc. (Bardgett et al., 2008). Soil microbial populations are immersed in a framework of interactions which affect plant fitness and soil quality. The great array of root-microbe interactions results in the development of a dynamic environment known as the rhizosphere where microbial communities interact. The growth and colonization of bacteria on plant root can be influenced by several soil chemical, physical and biological factors (Harries, 1998). Gaining a detailed understanding of these microbial communities regarding their relationship to environmental factors and ecosystem function, and developing methods to accurately assess them has often proven difficult (Barns et al., 1999). The principal contributor to these difficulties is the opaque nature of the soil environment, which makes direct observation of soil communities difficult. Another reason is the high diversity of these communities (Fierer et al., 2007). Amongst the various environmental factors associated with climatic changes, temperature along with moisture content is the most important environmental factor influen-cing the various processes in soil including microbial activity and microbial growth in soil. Soil temperature greatly affects the microbial population. All microbes have a set of optimal environ-mental conditions under which their growth rate is maximal, the so-called growth optima (Pettersson, 2004). Increasing temperature can increase microbial activity, processing, and turnover, causing the microbial community to shift in favor of representatives adapted to higher temperatures and faster growth rates (Castro et al., 2010).

Measurement of the soil microbial population and size of soil microbial biomass does not indicate microbial activity. Microbial activities include activities of general soil enzymes (Amador et al., 1997). Soil microorganisms are surrounded by organic matter rich in carbon and other nutrients that are required for growth and cell maintenance. But, microbes cannot directly transport these macromolecules into the cytoplasm. Rather, they rely on the activity of array of enzymes to utilize various nutrients. The rate at which soil organic matter (SOM) is decomposed is strongly affected by temperature and moisture, and thus should be sensitive to climate change. Activities of the soil enzymes are critical to the soil functioning and for maintenance of the vast biodiversity of organisms present in the soil. Soil enzymes drive soil organic matter decomposition, nutrient transformations and therefore, considered as indicator of soil health and quality. Temperature and moisture can affect both the overall rate of enzyme production and microbial activity as well as the relative rate of production of different enzymes due to effects on enzyme efficiency, substrate availability and microbial efficiency. Thus, changes in the soil microclimate, whether they occur within hours, weeks, seasonally, or over decades in response to climate change, will affect enzyme pool sizes (Steinweg et al., 2013). The link between soil microbes and their function can be made by studying the activity of enzymes involved in the C, N and P cycling (Caldwell, 2005). Soil enzymes are suitable measure to assess the effect of changing climate because they are very sensitive and respond quickly to environmental alterations. Moreover, soil enzymes are strongly associated with microorganisms. Soil enzyme activities are believed to indicate the extent of specific soil processes and in some cases they act as indicator of soil fertility. Among all enzymes in the soil environment, dehydrogenases are one of the most important, and are used as an indicator of overall soil microbial activity, because they occur intracellularly in all living microbial cells (Yuan and Yue, 2012). Alkaline phosphatase enzyme is also considered as representative of soil microbial activity. The activities of various enzymes that degrade the principle component of detrital organic matter have been extensively studied from many prospective (Kaur, 2013).

Rice and wheat are the major cereal crops cultivated in India and their growth depends upon climate, seed type and soil conditions. These both crops are primarily grown in Punjab due to rice-wheat cropping system. Although these both crops have been extensively studied but, the fundamental role of changing climatic conditions in regulating enzyme activities and microbial population of these crops under field conditions has been examined in relatively few studies. Keeping all these points in view, this research was carried out to study the effect of altered environmental conditions on microbial population and enzyme activities in rhizospheric soil of wheat and rice grown under field conditions and under temperature gradient tunnel.

MATERIALS AND METHODS

Soil sample collection

Soil samples were collected from rhizospheric soil of rice and wheat crops grown under field conditions and under Temperature Gradient Tunnel (TGT) located at the research farm of PAU, Ludhiana. The dimensions of the TGT were 30 m length × 5 m width and the meteorological parameters at the various depths within the TGT and outside were monitored at hourly interval by automated weather station manufactured by Delta -T devices, UK. Temperature and moisture within the TGT were maintained using fans, exhaust fans and coolers. TGT does not have fixed temperature. But, it always has 3-5°C higher temperature than the open field because it entraps heat radiations thus leading to higher temperature in it. TGT works on the phenomenon of greenhouse effect.

Soil temperature and moisture data recorded at different time intervals in wheat crop is represented in Table 1. Accurate recording of these parameters in rice crop was difficult due to continuous flooding in rice crop. Soil samples were collected at different time

Table 1. Weather conditions at different time intervals within TGT and open field in wheat.

Time Interval (DAS)		Wheat	
		Soil temperature (°C)	Moisture content (%)
0	F	17.5	21.6
	TGT	20.0	25.2
60	F	13.2	15.5
	TGT	17.2	16.4
120	F	23.2	15.8
	TGT	28.5	17.9
Harvesting	F	30.8	06.5
	TGT	34.7	08.7

intervals, 0, 60, 120 days after sowing (DAS) and at harvest from rhizosphere of rice crop grown during Kharif 2012 (10 June, 2012 - mid October, 2012) to wheat crop grown during rabi 2012-13 (November, 2012 - April, 2013). The crops were raised within the TGT and in open by following the crop management practices recommended in the Package of Practices of PAU, Ludhiana. Soil samples were collected by composite sampling method. Several soil samples were randomly taken from different areas of the same field. These soil samples were mixed together to get a representative sample (Walworth, 2006). A total of 16 soil samples, 4 each from rice grown under field conditions, rice grown under TGT, wheat grown under field conditions and wheat grown under TGT were collected.

Enumeration of microbial flora

Microbial populations such as total bacteria, nitrogen fixers, P-solubilizers and fungi were enumerated on Soil Extract Agar medium (Subba Rao, 1977), Jensen's medium (Jensen, 1942), Pikovaskaya's Agar medium (Pikovskaya, 1948) and Oxytetracycline Yeast Extract Agar medium (Mossel, 1970), respectively, using serial dilution spread plate technique (Kaur, 2013). All the media were prepared and sterilized in an autoclave at 15 psi pressure and 121°C temperature for 20 min.

Soil enzyme activities assay

Soil samples were analyzed for alkaline phosphatase activity and dehydrogenase activity. Alkaline phosphatase activity of soil samples was measured using para-nitrophenyl phosphate (pNPP) as substrate (Tabatabai and Bremner, 1969). It is based on colorimetric estimation of the p-nitrophenol released by phosphatase activity when soil is incubated with buffered (pH 11) sodium p-nitrophenyl phosphate solution and toluene. The colorimetric procedure used for the estimation of p-nitrophenol is based on the fact that alkaline solution of this phenol has a yellow color whereas alkaline solution of p-nitrophenyl phosphate is colorless. The intensity of yellow color is measured at 420 nm wavelength. Dehydrogenase activity of soil samples was measured using triphenyl tetrazolium (TTC) chloride as substrate (Klein et al., 1971). Measurement of dehydrogenase activity involves determi-nation of triphenyl formazan (TPF) produced by the reduction of Triphenyl tetrazolium chloride (TTC). TTC is a yellow colored water soluble salt and possesses the property of being easily transformed into intensely colored, water insoluble, methanol soluble formazan by reduction. TPF formed is extracted with methanol. The intensity of pinkish color is measured at 480 nm wavelength.

RESULTS AND DISCUSSION

Microbial population

Soil temperature greatly affects the microbial population within the soil profile. Both rice and wheat belong to C3 plant family and thus share similar metabolic characteristics. In rice crop, maximum bacterial population (310×10^6 cfu/g soil) was found at 60 DAS in soil sample taken from TGT. In case of N_2- fixing bacteria, maximum population (243×10^4 cfu/g soil) was found in soil sample taken from TGT at 120 DAS. Maximum fungal population (40×10^2 cfu/g soil) was found at 60 DAS in soil sample taken from rice grown under field conditions. Whereas, maximum P-solubilizers population of 29×10^3 cfu/g soil was found at 60 DAS in soil sample taken from tunnel (Table 2). Microbial population such as total bacteria, nitrogen fixers and P-solubilizers was found to be less in soil samples taken from rice grown under field conditions as compared to the microbial population in soil samples taken from TGT, at different time intervals. But, fungal population was found to be significantly higher in soil samples taken from rice grown under field conditions than in rice gown under TGT as indicated by the *p*-value (4.7×10^{-5}). This might be due to high temperature in tunnel which decreases fungal population as fungi is more inhibited by higher temperature than bacteria. Similar results have been reported by Pietikainen et al. (2005). They also found that fungal populations are more negatively affected by higher temperature than bacteria. This drastic decrease in fungal population at higher temperature results in an increase in the ratio of bacterial to fungal growth rate at higher temperatures. Another reason for low fungal population in the soil samples taken from TGT is high relative humidity in rice crop. Jensen et al. (2003) also stated that fungi as a group are more adapted to low soil moisture conditions than bacteria.

In the case of wheat crop, microbial population (total bacteria, nitrogen fixers, P-solubilizers and fungi) was found to be less in soil samples taken from wheat grown under field conditions as compared to the microbial population in soil samples taken from wheat grown under

Table 2. Microbial population in rhizospheric soil of rice crop grown under field conditions and under TGT at different time intervals during Kharif 2012.

Time interval (DAS)	Soil temperature (°C)	Bacterial population (cfu g^{-1}soil×10^6)	Diazotrophic population (cfu g^{-1} soil×10^4)	Fungal population (cfu g^{-1}soil × 10^2)	P-solubilizers population (cfu g^{-1}soil × 10^3)
0	33.5 (F)	59	41	19	11
	37.2 (TGT)	97	72	07	15
p-value (< 0.05)		1.86×10^{-8}	2.22×10^{-7}	4.7×10^{-5}	1.62×10^{-2}
60	27.1 (F)	286	208	40	20
	30.9 (TGT)	310	229	36	29
p-value (< 0.05)		1.65×10^{-6}	1.41×10^{-6}	4.36×10^{-5}	4.36×10^{-5}
120	28.4 (F)	219	209	29	17
	32.3 (TGT)	243	243	17	19
p-value (< 0.05)		1.69×10^{-7}	2.03×10^{-8}	1.2×10^{-5}	3.2×10^{-3}
Harvesting	25.5 (F)	112	91	31	14
	30.4 (TGT)	129	117	13	17
p-value (< 0.05)		4.96×10^{-6}	7.51×10^{-6}	5.48×10^{-6}	1.67×10^{-2}

F refers to field; TGT refers to temperature gradient tunnel.

Table 3. Microbial population in rhizospheric soil of wheat crop grown under field conditions and under TGT at different time intervals.

Time interval (DAS)	Soil temperature (°C)	Bacterial population (cfu g^{-1}soil×10^6)	Diazotrophic population (cfu g^{-1} soil×10^4)	Fungal population (cfu g^{-1}soil×10^2)	P-solubilizers population (cfu g^{-1}soil × 10^3)
0	17.5 (F)	14	13	06	03
	20.0 (TGT)	39	30	11	09
p-value (< 0.05)	(< 0.05)	1.05×10^{-6}	5.16×10^{-6}	2.7×10^{-3}	8.0×10^{-4}
60	13.2 (F)	57	136	29	24
	17.2 (TGT)	79	167	31	27
p-value (< 0.05)	(< 0.05)	7.53×10^{-5}	1.96×10^{-6}	1.3×10^{-3}	1.3×10^{-3}
120	23.2 (F)	219	111	33	07
	28.5 (TGT)	279	143	40	12
p-value (< 0.05)		2.48×10^{-9}	3.08×10^{-8}	1.0×10^{-4}	2.0×10^{-4}
Harvesting	30.8 (F)	207	97	17	11
	34.7 (TGT)	215	113	07	08
p-value (< 0.05)		4.38×10^{-9}	8.77×10^{-6}	5.0×10^{-5}	1.4×10^{-3}

F refers to field; TGT refers to temperature gradient tunnel.

TGT, at all different time intervals. At 60 DAS, the microbial count of all the microbial populations was observed to be higher than the microbial count taken at zero day of the experiment in the case of both field and tunnel soil samples. In case of total bacteria, maximum population (279 × 10^6 cfu/g soil) was found at 120 DAS. Maximum nitrogen fixers population of 167 × 10^4 cfu/g soil was found in soil samples taken at 60 DAS which may be due to increase in release of root exudates released by the crop. Maximum fungal population (40 × 10^2 cfu/g soil) was found at 120 DAS and P-solubilizers population (27 × 10^3 cfu/g soil) was found to be maximum at 60 DAS (Table 3). Maximum population of bacteria was observed whereas, P-solubilizers were very less in number. Along with the time interval, microbial populations were also found to be significantly correlated with the soil temperature at that time interval.

Changes in soil temperature affect microbial population and their activity. Data analysis revealed that maximum microbial count was found in the rhizospheric soil samples

taken from TGT than in the soil samples taken from wheat grown under field conditions (Table 3). It might be due to the fact that the soil samples were taken from wheat in winter season. Wheat crop was cultivated from November to April in Punjab state. During this period, the temperature decreases from month of November to January and then starts increasing. The minimum temperature was observed in the month of January (13.2°C) which was not optimum for the growth of micro-organisms. So, the outside temperature was very low and unfavorable for the growth of micro-organisms. The sun's energy passed through the tunnel and heated the air and soil beneath the crop. Therefore, temperature gradient tunnel resulted in increase of soil temperature from 3 to 4°C and created a favorable microenvironment for the growth of soil micro-organisms, which helped to improve and maintain the biological and physico-chemical qualities of the soils, thereby improving the growth of microbial population.

In winter, when temperature is low, the number and activity of microorganisms falls down, and as the soils warms up in summer, they increases in number as well as activity (http://www.agriinfo.in). Therefore, more microbial population was observed in rice crop as compared to wheat crop. High microbial population (total bacteria, nitrogen fixers and P-solubilizers) in rice crop (Table 2) can be attributed to presence of high moisture content in rice crop. Melentev et al. (2000) also reported similar results. They found that the moisture content and temperature of the soil considerably affect the microbial cenosis of the soil and the rhizosphere, since these factors can essentially change the amount and the pattern of nutrient secreted by plant roots. In both crops, maximum population of bacteria was observed whereas, P-solubilizers were very less in number. In the present study, significant improvement in soil microbial counts (except fungal population in rice crop) was observed in soil samples taken from rice and wheat grown under temperature gradient tunnel.

Alkaline phosphatase activity

Enzyme activities of the soil are result of enzyme producing activity of soil microbes present in it. In rice crop, at zero day, alkaline phosphatase activity in rhizospheric soil samples taken from field conditions was observed to be 32.8 µg/g soil/hour at 33.5°C soil temperature. Whereas, higher alkaline phosphatase activity of 39.0 µg/g soil/hour was found in rhizospheric soil samples taken from TGT at soil temperature of 37.2°C (p-value- 2.05×10^{-10}). At 60 and 120 DAS, similar trend of significantly higher phosphatase activity in soil samples taken from TGT was observed. At the time of harvesting, alkaline phosphatase activity in rhizospheric soil samples taken from field conditions was observed to be 25.0 µg/g soil/hour at soil temperature of 25.5°C. Whereas, alkaline phosphatase activity in rhizospheric soil samples taken from TGT was found to be 27.9 µg/g soil/hour at 30.4°C soil temperature (p- value- 4.17×10^{-9}). Higher alkaline phosphates activity was found in rhizospheric soil samples taken from TGT at all the different time intervals (Figure 1a).

Similarly in wheat crop, at zero day, alkaline phosphatase activity of rhizospheric soil samples taken from field conditions was observed to be 17.0 µg/g soil/hour at 17.5°C soil temperature. Whereas, higher alkaline phosphatase activity of 22.1 µg/g soil/hour was found in rhizospheric soil samples taken from TGT at 20.0°C soil temperature (p-value- 3.49×10^{-7}). Similar trend of increased alkaline phosphatase activity in soil samples taken from TGT was observed at 60 and 120 DAS. At harvest, alkaline phosphatase activity in rhizospheric soil samples taken from field conditions was observed to be 28.3 µg/g soil/hour at 30.8°C soil temperature. Whereas, alkaline phosphatase activity in rhizospheric soil samples taken from TGT was observed to be 33.0 µg/g soil/hour at 34.7°C soil temperature (Figure 1b). Data analysis at different time intervals clearly indicated that significantly higher alkaline phosphatase was found in the rhizospheric soil of TGT than in the soil samples taken from field conditions.

An increasing trend in the alkaline phosphatase activity with increase in soil temperature was observed in both field and tunnel samples in the case of both crops because soil temperature was increasing towards the optimum temperature. The optimum temperature for soil enzyme alkaline phosphatase is 37°C. Beyond this optimum temperature, activity start decreasing with increase in temperature and at very high temperature, enzyme gets denatured. Changes in soil temperature above or below optimum temperature of enzyme would result in decrease in soil phosphatase activity. Our results are also in agreement with the study conducted by Banerjee et al. (2012). They measured p-nitro phenyl phosphate degradation over a range of temperature from 17 to 67°C and observed that degradation of p-nitro phenyl phosphate demonstrated a broad range of activity. Maximum enzyme activity was attained at 37°C. Inhibitory effects on phosphatase activity were observed above or below this temperature.

Dehydrogenase activity

Dehydrogenase enzyme activity is considered as an indicator of the oxidative metabolism in soils and thus of the microbiological activity. In rice crop, at zero day, dehydrogenase activity of rhizospheric soil samples taken from field conditions was observed to be 8.02 µg TPF/g soil/hour at 33.5°C soil temperature. Whereas, higher dehydrogenase activity of 10.94 µg TPF/g soil/hour was found in rhizospheric soil samples taken from TGT at 37.2°C soil temperature (p-value- 6.17×10^{-14}). Similar trend of significantly higher dehydrogenase activity in soil samples taken from TGT was observed at 60 and 120 DAS. At harvest, dehydrogenase activity in rhizospheric

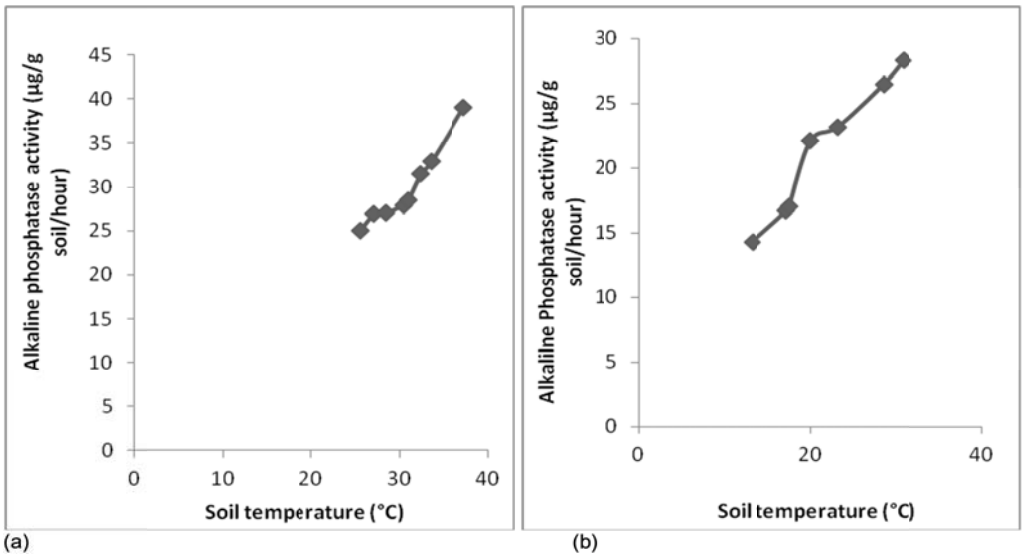

Figure 1. Variation of soil alkaline phosphatase activity with soil temperature (a) Rice crop and (b) Wheat crop.

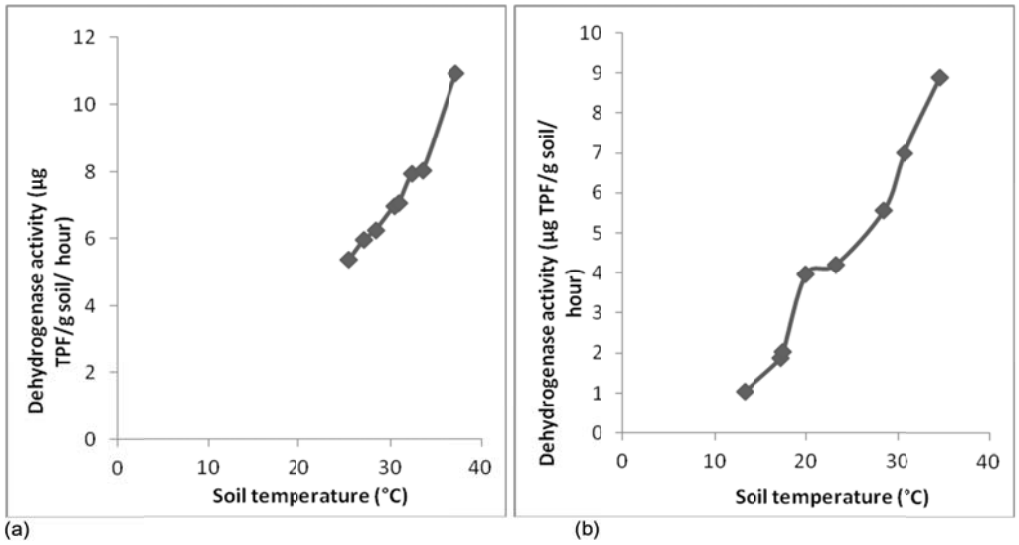

Figure 2. Variation of soil dehydrogenase activity with soil temperature (a) Rice crop and (b) Wheat crop.

soil samples taken from field conditions was observed to be 5.35 µg TPF/g soil/hour at soil temperature of 25.5°C. Whereas, dehydrogenase activity in rhizospheric soil samples taken from TGT was 6.96 µg TPF/g soil/hour at 30.4°C soil temperature. Higher dehydrogenase activity was found in rhizospheric soil samples taken from TGT at different time intervals (Figure 2a). Similarly in wheat crop, at zero day, dehydrogenase activity in rhizospheric soil samples taken from field conditions was observed to be 2.02 µg TPF/g soil/hour at 17.5°C soil temperature. Whereas, higher dehydrogenase activity of 3.97 µg TPF/g soil/hour was found in rhizospheric soil samples taken from TGT at soil temperature of 20.0°C (p-value- 4.5×10^{-13}). Similar trend of significantly higher dehydrogenase activity in soil samples taken from TGT was observed at 60 and 120 DAS. At harvest, dehydrogenase activity in rhizospheric soil samples taken from field conditions was observed to be 7.01 µg TPF/g soil/hour at 30.8°C soil temperature. Whereas, dehydrogenase activity of 8.88 µg TPF/g soil/hour was found in rhizospheric soil samples taken from TGT at soil temperature of 34.7°C (p-value- 1.0×10^{-12}). Data clearly indicated that significantly higher dehydrogenase activity was found in the soil samples taken from rhizospheric soil of TGT than in the soil samples taken from field conditions (Figure 2b).

An increasing trend in the dehydrogenase activity was

observed with increase in soil temperature in the case of both crops, because, in the present study, soil temperature were increasing towards the optimum temperature (Figure 2). So, activity of soil enzyme dehydrogenase increases significantly upto the optimum temperature beyond which activity start decreasing with increase in temperature. At a very high temperature, enzyme gets denatured.

At different time intervals, maximum dehydrogenase activity was found in the soil samples taken from TGT as compared to soil samples taken from field conditions. Results are in accordance with the study carried out by Trevors (1984) who found positive correlation between soil temperature and soil dehydrogenase activity. On comparison of enzyme activities of rice and wheat crop, it was found that activities of both enzymes were more in rice crop as compared to wheat. More enzyme activity in rice crop is also due to high moisture content in rice crop (flooding conditions in rice crop). Results are in agreement with the study conducted by Banerjee et al. (2000) and Brzezinska et al. (1998). According to them, soil water content and soil temperature influence soil dehydrogenase activity by affecting the soil oxidation-reduction status. Ross and Roberts (1970) also reported that dehydrogenase activities vary with season and are dependent on soil temperature. Present results are also supported by the study conducted be Yuan and Yue (2012). They also found lowest value of enzyme activity in winter.

Conclusion

It is concluded that soil temperature had profound effect on microbial population and enzyme activities. Though microorganisms can tolerate extreme environmental conditions, but the optimum environmental conditions at which soil microorganisms can grow and function actively is rather narrow. Soil microbial population and enzyme activities are suitable measure to assess the relationship between environment alteration and the total microbial activity but, this relationship is not always obvious, especially in the case of complex systems like soils.

Conflict of Interests

The authors have not declared any conflict of interests.

REFERENCES

Amador JA, Gluksman AM, Lyons JB, Gorres JH (1997). Spatial distribution of soil phosphatase activity within riparian forest. Soil Sci. 162:808-825.

Baei P, Risbey JS (2009). Uncertainity and assessment of issues posed by urgent climate change. An editorial comment. Clim. Chang. 92:31-36.

Banerjee A, Sanyal S, Sen S (2012). Soil phosphatase activity of agricultural land: A possible index of soil fertility. J. Agric. Res. 2:412-419.

Banerjee MR, Burton DL, McCaughery WPP, Grant CA (2000). Influence of pasture management on soil biological quality. J Range Manage. 53:127-133.

Bardgett RD, Freeman C, Ostle NJ (2008). Microbial contributions to climate change through carbon cycle feedbacks. ISME J. 2: 805-814.

Barns SM, Takala SL, Kuske CR (1999). Wide distribution and diversity of members of the bacterial kingdom Acidobacterium in the environment. Appl. Environ. Microbiol. 65:1731-1737.

Brzezinska M, Stepniewska Z, Stepniewski W (1998). Soil oxygen status and dehydrogenase activity. Soil Biol. Biochem. 30: 1783-90.

Caldwell BA (2005). Enzyme activities as a component of soil biodiversity: a review. Pedobiologia 49: 637-644.

Castro HF, Classen AT, Austin EE, Norby RJ, Schadt CW (2010). Soil microbial community responses to multiple experimental climate change drivers. Appl. Environ. Microbiol. 76: 999-1007.

Fierer N, Breitbart M, Nulton J, Salamon P, Lozupone C, Jones R, Robeson M, Edwards RA, Felts B, Rayhawk S, Knight R, Rohwer F, Jackson RB (2007). Metagenomic and small-subunit rRNA analyses reveal the genetic diversity of bacteria, archaea, fungi, and viruses in soil. Appl. Environ. Microbiol. 73:7059-7066.

Harries PJ (1998). Ecology of the soil population. In wild. A (ed.), Russels Soil conditions and plant growth. pp. 472-500. http://www.agriinfo.in/

Jackson RB, Fierer N, Schimel JP (2007). New directions in microbial ecology. Ecology 88: 1343-1344.

Jensen HL (1942). Pro Line Soc. N.S.W. 57:205-212.

Jensen KD, Beier C, Michelsen A, Emmett BA (2003). Effects of experimental drought on microbial processes in two temperate heathlands at contrasting water conditions. Appl. Soil Ecol. 24: 165-176.

Kaur J (2013). Screening of microbial communities for high temperature and CO_2 tolerant diazotrophs from wheat rhizosphere. M.Sc thesis, Punjab Agricultural University, Ludhiana, India.

Klein DA, Loh TC, Goulding RL (1971). A rapid procedure to evaluate dehydrogenase activity of soils low in organic matter. Soil Biol. Biochem. 3:385-387.

Melentev AL, Kuzmlna LY, Gallmzyanova NF (2000). Effect of Temperature and Soil Moisture Content on the colonization of the Wheat Rhizosphere by Antiphytopathogenic Bacilli. Microbiology 69(3):351-356.

Mossel DAA (1970). O.G.Y.E. for Selective Enumeration of Moulds and Yeast in Foods and Clinical Material. J. Appl. Bact. 35: 454-457.

Pettersson M (2004). Factor affecting rates of change in soil bacterial communities. Ph.D. thesis, Lund University, Sweden.

Pietikainen J, Pettersson M, Baath E (2005). Comparison of temperature effects on soil respiration and bacterial and fungal growth rates. FEMS Microbiol. Ecol. 52:49-58.

Pikovskaya R I (1948). Mobilization of phosphate dissolving bacteria in rhizosphere of some cultivated legumes. Plant Soil 35: 127-32.

Ross DJ, Roberts HS (1970). Enzyme activities and oxygen uptakes of soils under hard pasture in temperature and rainfall sequences. J. Soil. Sci. 21:368-381.

Steinweg JM, Dukes JS, Paul EA, Wallenstein MD (2013). Microbial responses to multifactor climate change: effects on soil enzymes. Front. Microbiol. 4:146

Subba Rao NS (1977). Soil Microorganisms and Plant Growth, Oxford and IBH Publishing Co., New Delhi. p. 251.

Tabatabai MA, Bremner JM (1969). Use of p-nitrophenyl phosphate for assay of soil phosphatase activity. Soil Biol. Biochem. 1: 301-307.

Trevors J (1984). Effect of Substrate Concentration, Inorganic Nitrogen, O_2 Concentration, Temperature and pH On Dehydrogenase Activity In Soil. Plant Soil 77:285-293.

Walworth JL (2006). Soil sampling and analysis. The university of Arizona publication, “Laboratories Conducting Soil, Plant, Feed or Water Testing” (AZ1111).

Yuan B, Yue D (2012). Soil Microbial and Enzymatic Activities Across a Chronosequence of Chinese Pine Plantation Development On The Loess Plateau of China. Pedosphere 22:1-12.