Seasonality and moisture regime control soil respiration, enzyme activities, and soil microbial biomass carbon in a semi-arid forest of Delhi, India

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Abstract
Soil respiration, soil enzymes, and microbial biomass are important in carbon cycling in the terrestrial ecosystem which is generally limited by environmental factors and soil carbon availability. Hence, we tried to assess the factors affecting the functional aspects of these processes in a semi-arid climate. We monitored soil respiration (surface) using a portable infrared gas analyzer (Q-Box SR1LP Soil Respiration Package, Qubit Systems, Canada) equipped with a soil respiration chamber (Model: G 180). Soil respiration was measured at midday during each season throughout the study period. Soil enzymatic activities and microbial biomass carbon (MBC) were analyzed following the standard protocol for a year during peak time in four seasons at 0–10 cm and 10–20 cm depth. Soil respiration shows significant variation with highest in monsoon (3.31 μmol CO₂ m⁻² s⁻¹) and lowest in winter (0.57 μmol CO₂ m⁻² s⁻¹). Similarly, β-glucosidase, dehydrogenase, and phenol oxidase activity ranged from 11.15 to 212.59 μg PNP g⁻¹ DW h⁻¹, 0.11 to 16.47 μg TFP g⁻¹ DW h⁻¹, and 4102.95 to 10187.55 μmol ABTS⁺ g⁻¹ DW min⁻¹, respectively. MBC ranged from 17.08 to 484.5 μgCg⁻¹. Besides, soil respiration, soil enzymes (except β-glucosidase), and MBC were significantly correlated with soil moisture. Seasonality, optimum moisture and temperature played a significant role in determining variations in soil microbiological processes (except β-glucosidase activity); the carbon cycling in the study area is assisted by enzyme activity; dehydrogenase and phenol oxidase played a significant role in soil respiration; hence, this landscape is sensitive to environmental changes.

Keywords: Soil respiration, Enzyme activities, Microbial biomass carbon, Seasons, Semi-arid forest

Background
Soil respiration accounts for the largest potential source of atmospheric carbon; hence, even small changes in soil respiration can increase or decrease the atmospheric carbon dioxide level (Schimel 1995). Soil respiration is carbon dioxide emission from soil surface which controls the primary carbon cycle in ecosystems (Jin et al. 2007). Soil respiration consists of two components, namely, autotrophic and heterotrophic respiration. Autotrophic respiration is carbon dioxide released from plant roots whereas heterotrophic respiration is carbon dioxide released from microbial activity associated with soil organic matter decomposition and constitute 54% of total respiration in the forest (Hanson et al. 2000; Ryan and Law 2005; Wei et al. 2015). Photosynthesis and heterotrophic respiration are the key processes that regulate terrestrial carbon balance (Xu et al. 2018). Soil heterotrophic respiration and decomposition are mainly associated with the microbial activity (Hanson et al. 2000). Soil enzymes catalyze many important biological processes involved in enhancing the rate of soil metabolism and promote the circulation of nutrient elements (Li et al. 2018).
The soil has a major role in the fertility and stability of forest ecosystems (Smith et al. 1992). Micro-organisms present in soil are responsible for decomposition and conversion of organic matter for vegetation development and plant growth (Aguilera et al. 1999). Soil microbial biomass can act as a source or sink of available nutrients (Singh et al. 1989) and changes in microbial biomass also affect soil organic matter turnover (Yang et al. 2010). The main component of soil microbial biomass is microbial biomass carbon (MBC), it is responsible for controlling the carbon and nutrient flows in ecological systems (Ross et al. 1995; Shao et al. 2015). Living microbial biomass carbon and dead microbial biomass both contribute to microbial biomass carbon pool (Xu et al. 2018). Necromass of dead micro-organisms represents a huge amount of carbon in soil and can act as a readily available source of carbon for living micro-organisms (Xu et al. 2018).

It is well known that the accumulation and decomposition of soil organic carbon (SOC) has a direct effect on carbon storage in the terrestrial ecosystem and global carbon balance (Liu et al. 2016). Hence, soil microbial properties such as soil respiration, enzyme activity, and microbial biomass are considered important in predicting SOC dynamics in many recent studies (Lawrence et al. 2009; Davidson et al. 2012; Wieder et al. 2013; Wei et al. 2015; Memoli et al. 2017; Panico et al. 2020). Various studies have shown seasonal variation in soil respiration (Borken et al. 2003; Huxman et al. 2004; Xu et al. 2004; Jin et al. 2007; Placella et al. 2012; Salazar et al. 2018; Meena et al. 2020), soil enzyme activity (Boerner et al. 2005; Bastida et al. 2008; Hedo et al. 2015), and soil microbial biomass carbon (Singh et al. 1989; Maithani et al. 1996; Bohlen et al. 2001; Ruan et al. 2004; Feng et al. 2009) in the various forest ecosystem. Previous studies of Arunachalam and Arunachalam (2000), Barbhuiya et al. (2004), and Mori et al. (2016) have reported seasonal variation in MBC in the sub-tropical broad-leaved forest, wet-tropical forest and tropical savannah, respectively. However, studies reporting the same in semi-arid forests are still lacking. Also, seasonal effect considering the association of soil respiration, and enzyme activity with soil MBC remain uncertain in semi-arid conditions. SOC is mainly recognized as a driving factor that regulates soil microbial growth (Wardle 1992). Soil respiration mainly depends on the concentration, composition, and rate of supply of carbon substrates in the soil solution to the microbial community which is responsible for soil respiration (Van Hees et al. 2005; Iqbal et al. 2010). Also, to evaluate the effect of management on soil respiration in forest ecosystems, soil microbial community and its dynamics should be given much attention (Qi and Yang 2017).

In arid and semi-arid ecosystems strong seasonal precipitation and cycles of drying and rewetting are predominant (Austin et al. 2004). The irregular rainfall pattern in the semi-arid region can create drying and wetting stresses in forest soils (Fierer and Schimel 2002). Hence in such ecosystems, soil microbes are highly sensitive to water impulse and precipitation which results in soil organic carbon decomposition initiation, thereby leading to a cascade of various responses (Carbone et al. 2011; Xu et al. 2018). Since the beginning of the twentieth century, changes in precipitation, temperature, and extreme climatic events have been observed (Milly et al. 2002; Peterson et al. 2002), simultaneously, rainfall event was also found to be less frequent and more extreme (Stocker 2014). Variability in monsoon rainfall is observed in the country (Mall et al. 2006) which will develop a threat for tropical soils in the Indian subcontinent (Bhattacharyya et al. 2000). As the future climate is predicted to be more extreme even in the case of the semi-arid condition, the data on such conditions would be more important. Thus, to predict the future of carbon cycle and their potential to sequester carbon in the soil in the changing world, we need to understand the seasonal changes in the processes that play important role in carbon cycling (i.e., soil respiration, enzyme activity, and soil MBC).

The study was conducted in the National Capital Territory (NCT) of Delhi which has a scattered forest cover and investigations on soil respiration, enzyme activity, and MBC are very limited. Since these studies are essential for understanding carbon cycling, hence, creating management policies to maximize the sequestration of carbon in the soil becomes necessary. The data gap in the semi-arid forest of India compelled us to investigate whether the soil respiration, enzyme activities, and MBC were influenced by seasonal changes in the semi-arid climate. Our study also discussed the relationship between SOC and the above factors to provide a theoretical basis for further understanding of the carbon cycling in semi-arid forests of India.

Materials and methods

Study site

Our study sites are Delhi Ridges which are fragmented forest patches and scattered into four ridges, viz., Southern Ridge, Central Ridge, South-Central Ridge, and Northern Ridge occupying an area of 6200, 864, 626, and 87 ha, respectively. Out of the total geographical area of 1483 km², the forest cover of the state is 192.41 km² which is 12.97% of the total geographical area of Delhi (FSI 2017). The state lies in the northern part of India between 28° 24′ 17″ – 28°53′ 00″ N latitudes and 76° 45′ 30″ – 77° 21′ 30″ E longitude. The Delhi Ridge forests are the northern extension of Aravalli hill range and occupy 77.77 km² of forest cover. The distribution of the four ridges (Tomar and Baishya 2019) is depicted
in Fig. 1. During June 2016–May 2017, the temperature recorded in the study area varied between 15.6 and 40.5 °C in January and May, respectively. The annual rainfall recorded was 1285.2 mm from June 2016–May 2017 (Fig. 2a). The highest rainfall was observed in July and August. The basic soil characteristics of each Delhi Ridge are represented in Table 1.

The dominated species in the Delhi Ridge is Prosopis juliflora which is an exotic tree species introduced by the British regime in 1877 to enhance the vegetation of the region. According to Champion and Seth (1968), Ridge forests are classified as tropical thorn forests. Climate is semi-arid and soil texture is sandy loam. The soil pH ranged between 6.31 and 7.51. The state witnesses four seasons, namely, pre-monsoon (March to May), monsoon (June to Aug.), post-monsoon (Sept. to Nov.), and Winter (Dec. to Feb.) (Tomar and Baishya 2019).

Soil sampling
Soil sampling was done for 1 year between June 2016 and May 2017. The soil was collected using a steel soil auger after removing litter from the soil surface in pre-monsoon, monsoon, post-monsoon, and winter seasons in all four Ridges. Three replicates of five randomly collected sub-samples were collected and homogenized from each ridge in both soil depth, namely, 0–10 cm and 10–20 cm. Soil samples were collected from near the collars inserted for soil respiration study. Samples were taken to the laboratory in airtight and labeled sampling packets, plant litter, and woody debris were removed and stored at 4 °C for further analysis.

Soil physico-chemical and microbiological analysis
Soil moisture was determined within 48 h of soil collection. Ten grams of fresh soil was dried in an oven at 105 °C until constant weight. Soil moisture was determined following Allen et al. (1974). Soil temperature was measured using soil thermometer inserted till 10 cm depth in the soil. Soil organic carbon (SOC) was analyzed by digesting soil with a few drops of hydrochloric acid (HCl). The digested soil was oven dried and SOC content in the digested sample was determined using Liqui TOC II analyzer (Elementar Analysis systems GmbH, Germany). Soil respiration was measured at midday between 11:00 am and 2:00 pm local time, during each season throughout the study period, using a portable infrared gas analyzer (Q-Box SRILP Soil Respiration Package, Qubit Systems, Canada) equipped with soil respiration chamber (Model: G 180) having a diameter of 10.16 cm and a volume of 1.0 L. The collars were inserted 1.9 cm into the soil at each sampling point about two weeks before the first measurement. Litter was removed before the insertion of the chamber. All collars were left at the site for the entire study period. Soil respiration was expressed in μmol CO₂ m⁻² s⁻¹. Soil temperature and moisture were measured near each collar at the same time as soil respiration. Soil β-glucosidase activity (BA) indicates the state of organic matter and decomposition activity in the soil (Garcia
et al. 1994; Wang et al. 2014). It was assayed using p-nitrophenyl-β-D-glucoside (PNG) (Eivazi and Tabatabai 1988). One gram of field moist 2 mm sieved soils was taken in a 50 ml conical flask. To this 250 μl of toluene, 4 ml of modified universal buffer (pH 6) and 1 ml of 25 mM p-nitrophenyl-β-D-glucoside was added. The blank flask did not get a PNG solution. The flask was swirled, covered with stopper, and incubated at 37 °C for 1 h. Following incubation time 1 ml of 0.5 M CaCl₂ and 4 ml of 0.1 M THAM (tris-hydroxymethyl aminomethane), pH 12 was added. The contents were filtered using Whatman filter paper no. 2. Before filtering the blanks, 1 ml of PNG was added. Finally, the absorbance of p-nitrophenol (PNP) released was determined using a spectrophotometer at 400 nm. The concentration of PNP was estimated using a PNP standard calibration curve. Results were expressed as μg PNP g⁻¹ DW h⁻¹.

Soil dehydrogenase activity (DHA) reflects the metabolic activity in the soils (Wolinska and Stępniewska 2012). It was determined using 2,3,5-triphenyltetrazolium chloride (TTC) reduction assay (Casida 1977). Six grams of field moist 2 mm sieved soil was taken in a glass tube (15 × 150 mm). To this, 0.06 g of CaCO₃ was added along with 1 ml of 3% TTC solution. Blanks were also created for each soil sample where TTC was not added. The total volume of fluids was made to 3.5 ml (i.e., 3.5 ml autoclaved DI water in blanks and 2.5 ml in samples). All the tubes were sealed using parafilm, starred, and incubated at 37 °C for 24 h in dark. Following incubation time, soil samples were transferred with methanol into a 50-ml graduated cylinder through Whatman filter paper no. 5. The red methanolic extract of tri-phenyl formazan (TPF) was determined using a spectrophotometer at 485 nm against soil banks. The concentration of TPF was estimated using a TPF standard calibration curve. Results were expressed as μg TPF g⁻¹DW h⁻¹. Soil phenol oxidase activity (PO) indicates oxidation of complex organic compounds like lignin during decomposition (Luo and Gu 2015). It was determined using the ABTS assay (Floch et al. 2007). A total of 0.1 g field moist 2 mm sieved soil was taken in a 15-ml centrifuge tube and to this 10 ml of MUB solution (pH 2) in 200 μl of 0.1 M ABTS (2,2′-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid)) solution was added. The mixture was incubated at 30 °C for 5 min in a water bath following which the mixture was centrifuged at 12000 rpm for 2 min at 4 °C. The oxidation rate to ABTS⁺ was determined in the supernatant at 420 nm. Results were expressed as μmol ABTS⁺ g⁻¹ DW min⁻¹. Microbial biomass carbon (MBC) was determined by the fumigation-extraction method (Vance et al. 1987). Ten grams of field moist 2 mm sieved soil was fumigated with ethanol-free chloroform for 24 h in a vacuum desiccator and the other 10 g was not fumigated. Forty milliliter of 0.5 M K₂SO₄ solution

Table 1 Basic soil physicochemical properties (mean ± standard deviation) in the study area

| Ridge            | pH       | Bulk density (g cm⁻³) | TC (%)   | TN (%)   |
|------------------|----------|-----------------------|----------|----------|
| Southern Ridge   | 6.98 (0.02) | 1.07 (0.41)           | 0.65 (0.20) | 0.06 (0.06) |
| Central Ridge    | 6.91 (0.04) | 1.11 (0.31)           | 1.56 (0.25) | 0.12 (0.07) |
| South-Central Ridge | 6.94 (0.02) | 1.06 (0.49)           | 1.09 (0.17) | 0.11 (0.00) |
| Northern Ridge   | 6.85 (0.04) | 1.11 (0.32)           | 1.20 (0.34) | 0.11 (0.05) |

TC total carbon; TN total nitrogen
was used to extract both fumigated and non-fumigated samples. Samples were shaken for 1 h in a rotary shaker. The extract was filtered using Whatman filter paper no. 42. The carbon content in the filtrate was measured using Liqui TOC II analyzer. MBC was calculated as MBC = 2.22Ec, where Ec is organic carbon from fumigated soil—organic carbon extracted from non-fumigated soils (Wu et al. 1990). MBC was expressed as μg C g⁻¹.

Statistical analysis

Data were analyzed by two-way ANOVA at which season (monsoon, post-monsoon, winter, and post-monsoon) and soil depth (0–10 cm and 10–20 cm) were selected as factors. To determine pair-wise differences by post hoc tests, the data were submitted to one-way ANOVA for each season. Tukey’s HSD post hoc test was applied. A p < 0.01 or 0.05 level of significance was adopted throughout. Pearson’s correlation analysis was also carried out by including all the soil microbiological and physical variables measured in the study. The correlation pattern was further examined by principal component analysis (PCA). All the statistical analysis was done using IBM SPSS 16, statistical software.

Results

Physical and chemical variables

Soil moisture and temperature both showed significant seasonal variation during the study period (p < 0.01). Soil temperature ranged from 14.4 to 34 °C (Fig. 2b). It was observed highest in pre-monsoon and lowest in winter. Soil moisture was highest in monsoon and lowest in winter. It ranged from 3.07 to 17.12% and 2.97 to 12.97% under 0–10 cm and 10–20 cm depth, respectively in all the Delhi Ridges (Fig. 3a, b). The seasonality of soil moisture coincided with the seasonal pattern of precipitation. Significant seasonal variations were also obtained (p < 0.05) in SOC (Table 2), which was highest in winter and lowest in monsoon. SOC varied within the range of 0.84 to 2.04% and 0.26 to 1.74% in 0–10 and 10–20 cm depth, respectively (Fig. 3c, d). It also showed variations across depths and observed higher values in 0–10 cm depth (Table 2) in all the Delhi Ridges.

Seasonal variation in soil respiration

The highest soil respiration was observed in the monsoon season when the precipitation was at a peak (Fig. 4). Soil respiration showed significant seasonal variation (F value: 136.26 and p < 0.01) and showed maximum values in monsoon season in all the Delhi Ridges. It ranged from 0.57 to 3.31 μmol CO₂ m⁻² s⁻¹. Minimum soil respiration occurred in the winter season as the precipitation is lowest and soil temperature and soil moisture are minimum during the winter season.

Microbiological variables

Seasons and depth significantly affected all the enzymatic activities except β-glucosidase, which is affected by depth only. Variations in seasons and depth significantly influenced (p < 0.05) dehydrogenase and phenol oxidase (Table 2). The interaction between season and depth was influential for dehydrogenase (p < 0.05). β-glucosidase ranged from 29.14 to 212.59 μg PNP g⁻¹ DW h⁻¹ and 11.15 to 123.48 μg PNP g⁻¹ DW h⁻¹ in 0–10 cm and 10–20 cm depth, respectively (Fig. 5a, b). Dehydrogenase showed higher values in monsoon. It varied from 0.26 to 16.47 μg TPF g⁻¹ DW h⁻¹ and 0.11 to 8.95 μg TPF g⁻¹ DW h⁻¹ in 0–10 cm and 10–20 cm depth, respectively (Fig. 5c, d). Similarly, phenol oxidase showed higher values in post-monsoon and monsoon season and lower in pre-monsoon and winter seasons. It ranged from 4108.60 to 10187.55 μmol ABTS⁺ g⁻¹ DW min⁻¹ and 4102.95 to 7393.87 μmol ABTS⁺ g⁻¹ DW min⁻¹ in 0–10 cm and 10–20 cm depth, respectively (Fig. 5e, f).

Soil MBC showed clear significant (p < 0.05) seasonal variation during the study period (Table 2). Soil MBC varied with seasons and ranged from 49.88 to 484.52 μg C g⁻¹ and 17.08 to 358 μg C g⁻¹ in 0–10 and 10–20 cm depth, respectively (Fig. 6a, b) in all the Delhi Ridges. MBC was highest in monsoon season and lowest in the winter season. MBC also showed significant (p < 0.05) variation among the two depths and was observed higher in 0–10 cm than in 10–20 cm depth (Table 2).

Correlation analysis

A positive significant correlation was observed between MBC and other studied soil microbiological and physical variables (dehydrogenase, phenol oxidase, soil respiration, soil temperature, and soil moisture). MBC was observed to be negatively correlated with SOC (Table 3). Soil respiration showed a positive correlation with MBC, dehydrogenase, phenol oxidase, soil temperature, and soil moisture. β-glucosidase showed a significant positive correlation with SOC. However, it was not correlated with other microbiological and physical factors (Table 3). Dehydrogenase showed a positive significant correlation with respiration, MBC, and soil moisture, while a negative correlation was observed with SOC (Table 3). Phenol oxidase showed a positive correlation with MBC, soil respiration, soil temperature, and soil moisture, but a negative correlation was observed with SOC and β-glucosidase. In 10–20 cm depth, correlation patterns were found to be similar except for a negative correlation and no correlation of β-glucosidase and phenol oxidase was observed with soil temperature (Table 3).
The principal component analysis (PCA) was carried out on all the studied soil microbiological and physical variables as factors and the analysis revealed that two principal components with eigenvalue more than one are responsible for the variance observed in the studied factors. The component contributing to maximum variance becomes the first PC (principal component) and others become the second PC. In 0–10 cm depth, the PCA axis F1 (Fig. 7a) accounted for 56.17% of total variation among the studied soil parameters and the loadings values are 0.961 (soil respiration), 0.903 (soil moisture), 0.849 (MBC), −0.736 (SOC), 0.724 (soil temperature), 0.716 (DHA), 0.643 (PO), and −0.203 (BG) while PCA axis F2 accounted for 22.25% variation with loading values: 0.868 (BG), 0.618 (SOC), 0.583 (DHA), 0.337 (MBC), −0.366 (PO), 0.287 (soil moisture), −0.223 (soil temperature), and 0.067 (soil respiration). Similarly, in

**Table 2** Results of two-way ANOVA (season and depth) for microbiological variables

|        | Df  | F value |
|--------|-----|---------|
|        | BA  | DHA     | PO    | MBC  | SOC  |
| Season | 3   | 2.08    | 57.38* | 18.61** | 92.80** | 11.33** |
| Depth  | 1   | 26.30** | 15.38* | 4.01*  | 58.55** | 44.56** |
| Se × dp| 3   | 0.23    | 6.44** | 0.77  | 5.79** | 0.46   |

*df* degree of freedom; *Se* season; *dp* depth; *se × dp* interaction season and depth; BA β-glucosidase activity; DHA dehydrogenase activity; PO phenol oxidase activity; MBC microbial biomass carbon; SOC soil organic carbon

*n* = 12

* p < 0.05

** p < 0.01

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10–20 cm depth, the PCA axis F1 (Fig. 7b) accounted for 54.17% of total variation among the studied soil parameters and the loadings values are 0.960 (soil respiration), 0.879 (soil moisture), 0.869 (MBC), 0.709 (DHA), 0.699 (soil temperature), −0.638 (SOC), 0.597 (PO), and −0.358 (BG) while PCA axis F2 accounted for 22.63% variation with loading values: 0.817 (BG), and 0.642 (SOC), 0.610 (DHA), 0.364 (MBC), −0.324 (PO), 0.243 (soil moisture), −0.227 (soil temperature), 0.096 (soil respiration).

Discussion

Our study indicated that studied soil microbiological activity and MBC showed a clear seasonal pattern in all the Delhi Ridges. Soil respiration showed clear seasonal variation and was highest in monsoon season and lowest in winter season. The seasonal pattern of soil respiration was as per previous findings of Tang et al. (2006) and Meena et al. (2020), suggesting that seasonal changes in soil respiration are correlated with soil moisture and soil temperature. Lou et al. (2004) showed a correlation of soil respiration, soil moisture, and MBC. Additionally, we observed the correlation of soil respiration with the above factors as well as with enzyme activity (dehydrogenase and phenol oxidase). High soil respiration during monsoon is attributed due to an increase in microbial activity during the rewetting of soil after a period of drying (Salazar et al. 2018). The microbial activity utilizes C from SOC and releases it in the atmosphere in the form of CO₂ resulting in an increase in C emission from soil to atmosphere, thereby decreasing the soil C storage (Wang et al. 2013). Soil temperature and moisture directly or indirectly affect respiration-related enzyme activity by affecting the supply of the substrates (Kishimoto-Mo et al. 2015).

Soil enzymes have a major role in biochemical processes occurring in the soil environment as they are the core mediators of organic matter decomposition, nutrient cycling, and energy transfer (Shao et al. 2015). In the present study, dehydrogenase and phenol oxidase activity showed significant seasonal variation but β-glucosidase activity did not show any variation concerning the seasons. Every enzyme is characterized by its substrate and ability to catalyze specific biochemical reactions (Song et al. 2012). β-glucosidase activity catalyzes the hydrolysis of β-glucosides, thereby producing glucose and thus the enzyme complex is involved in the decomposition of plant remains (Hayano and Tubaki 1985). According to Baldrian et al. (2010), enzyme activities in forest soils show seasonal variation. However, in our study β-glucosidase activity did not show seasonal variations and correlation with soil moisture. Similar results were also obtained by Hedo et al. (2015) in semi-arid dry forest strands. The activity of β-glucosidase is likely to be controlled by organic matter in the soil and the varying inputs of the litter with a season do not affect the activity of this enzyme (Wick et al. 2002). We obtained a positive significant correlation \((r = 0.64, p < 0.01)\) of β-glucosidase with SOC (Table 3), as the enzyme mainly participates in mineralization and cycling of carbohydrates in the soil (Wick et al. 2002). It is involved in the hydrolytic conversion of cellulose as a fraction of soil organic matter pool rather than total carbon (Wick et al. 2002). Positive correlations of β-glucosidase with organic C was also reported by Eivazi and Tabatabai (1990) and Wick et al. (2002). The activity of dehydrogenases is fundamental part of enzymes system occurring in living organisms because of the involvement of several dehydrogenases in the respiratory pathway (Wolinska and Stepniewska 2012). Dehydrogenase activity is intracellular and the enzyme complex participates in the transfer of electrons (Nannipieri et al. 1990). Dehydrogenase activity serves as an indicator of microbial activity in such semi-arid soils (Ros et al. 2003), this can be observed from a positive correlation between MBC and dehydrogenase activity in our study, as when the number of microbes increases, production of dehydrogenase enzyme also increases. With significant seasonal variation, dehydrogenase activity was found higher in monsoon season suggesting that soil moisture has a significant role in the production of dehydrogenase enzyme. The survival and the activity of soil microorganisms is known to be impacted by the availability of water (Uhlirova et al. 2005). Low water availability results in lowering of intracellular water potential, thereby reducing hydration, and inhibiting microbial activity.
Fig. 5 Seasonal variations in β-glucosidase activity (μg PNP g⁻¹ DW h⁻¹) (a) 0–10 cm depth, (b) 10–20 cm depth; dehydrogenase activity (μg TPF g⁻¹ DW h⁻¹) (c) 0–10 cm depth, (d) 10–20 cm depth and phenol oxidase activity (μmol ABTS⁺ g⁻¹ DW min⁻¹) (e) 0–10 cm depth, (f) 10–20 cm depth under all seasons in all the Delhi Ridges. Bars indicate standard error of the mean. Different letters represent significant differences among different seasons in all the Delhi Ridges (p < 0.05; Tukey post hoc test).
(Wall and Heiskanen 2003). Microbial communities suffer starvation during periods of moisture limitation; hence, the drought stress is considered as the most common environmental stress for soil microorganisms (Wolinska and Stepniewska 2012). As the activities of dehydrogenases in different forest ecosystems are involved in C cycling (Salazar et al. 2011), thus, dehydrogenases are associated with microbial biomass and affects the decomposition of organic matter which is reflected by high soil respiration (Zhang et al. 2010). Hence, we observed a positive significant correlation between dehydrogenase activity and soil respiration.

**Table 3** Pearson's correlation matrix between soil parameters at 0–10 cm and 10–20 cm depth

|          | RESP | BA  | DHA | PO  | MBC | SOC  | ST  | SM  |
|----------|------|-----|-----|-----|-----|------|-----|-----|
| **0–10 cm** |      |     |     |     |     |      |     |     |
| RESP     | 1    | 0.13| 0.74**| 0.66**| 0.79**| −0.66**| 0.61**| 0.88**|
| BA       | 1    | 0.25| −0.38**| 0.10  | 0.64**| −0.18  | −0.00 |     |
| DHA      | 1    | 0.26| 0.73**| −0.17 | 0.24  | 0.84** |     |     |
| PO       | 1    | 0.33*| 0.59**| −0.29**| 0.48**|       |     |     |
| MBC      | 1    | 0.30*| −0.40**| 0.63**|       |     |     |     |
| SOC      | 1    | −0.76**| 0.46**|       |     |     |     |     |
| ST       | 1    | 0.52**|     |     |     |     |     |     |
| SM       | 1    | 0.52**|     |     |     |     |     |     |
| **10–20 cm** |      |     |     |     |     |      |     |     |
| RESP     | 1    | −0.26| 0.72**| 0.51**| 0.86**| −0.57**| 0.61**| 0.87**|
| BA       | 1    | 0.17| −0.47**| −0.00 | 0.65**| −0.29**| −0.20 |     |
| DHA      | 1    | 0.30*| 0.80**| −0.06 | 0.23  | 0.75** |     |     |
| PO       | 1    | 0.35*| −0.46**| 0.22  | 0.42**|       |     |     |
| MBC      | 1    | 0.31*| 0.56**| 0.77**|       |     |     |     |
| SOC      | 1    | 0.63**| −0.33*|       |     |     |     |     |
| ST       | 1    | 0.53**|     |     |     |     |     |     |
| SM       | 1    | 0.52**|     |     |     |     |     |     |

RESP soil respiration; BA β-glucosidase activity; DHA dehydrogenase activity; PO phenol oxidase activity; MBC microbial biomass carbon; SOC soil organic carbon; ST soil temperature; SM soil moisture

*p < 0.05; significant correlation

**p < 0.01; n = 48; significant correlation**
Phenol oxidase is considered as an indicator of the breakdown of recalcitrant carbon pool, thereby contributing to heterotrophic soil respiration (Sun et al. 2018). Phenol oxidase is an oxidative enzyme having a key function in lignin decomposition (Grandy et al. 2008). Phenol oxidase activity in the present study fluctuated significantly among the seasons and was found higher in monsoon and post-monsoon seasons. The results are as per the previous study by Zhou and Zhang (2014) who suggested that seasonal variations in oxidative enzyme activity are influenced by soil water content and soil temperature, but it is mainly attributed to seasonal changes in soil moisture. High temperature during pre-monsoon was not able to induce high oxidative enzyme activity probably due to low moisture content. But, after rainfall events during monsoon, the activity of phenol oxidase increases with an increase in soil moisture. Correlation analysis in our study also indicated that phenol oxidase activity is dependent more on soil moisture than soil temperature. The water limitation may lead to a reduction in enzyme production because of moisture stresses, which include restricted nutrient uptake, mycelium growth, cell proliferation, substratum penetration, and cell desiccation (in extreme cases) (Toberman et al. 2008). As seasonal variation in soil moisture and temperature affects phenol oxidase activity, which is responsible for C transformation; hence, lignin degradation and C mineralization also vary with soil moisture and temperature (Zhou and Zhang 2014). This can be observed from significant positive correlation of phenol oxidase with soil respiration observed in our study. MBC showed the highest value in monsoon season and lowest in the winter season. It is quite evident that in arid and semi-arid regions, water controls most of the biological processes occurring in soil (Collins et al. 2008). The range of MBC reported in the present study was 49.8 to 484.52 μg g⁻¹ which falls approximately in reported range (61 to 2000 μg g⁻¹) by Vance et al. (1987), Henrot and Robertson (1994) for various temperate and tropical forest soils. A similar seasonal pattern in MBC was reported by Yang et al. (2010) and MBC was reported higher in summer when rainfall was higher. In the present study, the seasonality in MBC was found significantly correlated with soil moisture (R² = 0.82; p < 0.01) in 0–10 cm and (R² = 0.77; p < 0.01) in 10–20 cm (Table 3). Similar results were observed in a pine plantation in the subtropical zone by Chen et al. (2003). A meta-analysis of global MBC across terrestrial ecosystems by Xu et al. (2013) showed 16.7 Pg C in the 0–30 cm of soil. MBC depends greatly on SOC and its availability for microbial activity. In arid and semi-arid conditions, wetting and rewetting cycles were pre-dominant and major part of soil microbial biomass become dormant during the dry period (Lopez-Sangil et al. 2018). However, the rewetting results in an enhancement in MBC as SOC accumulation occurred during the dry period (Xu et al. 2018). In the present study, SOC showed the highest value during the winter season and lowest during the monsoon season. A study by García-Oliva et al. (2003) reported an accumulation of organic matter during the dry period which enhances microbial activity and biomass in the wet season. Our study also presented similar
results as the accumulation of SOC was observed during the winter season (dry period) which later increased MBC after soil moisture and temperature become optimum for microbial growth. We found a significant negative correlation of MBC with SOC. SOC is mainly decomposed by soil micro-organisms and reduction in microbial activity results in a decrease in MBC, thereby decreased SOC decomposition rate results in the accumulation of SOC (Shao et al. 2015).

Conclusion
Seasonality prevailing in Delhi Ridge had a significant influence on MBC and enzymatic activities. All the studied microbiological processes (except β-glucosidase activity) were found higher in the monsoon season because of optimum moisture and temperature during the period. This can also be observed by a significant positive correlation of MBC and enzymatic activity (dehydrogenase and phenol oxidase) with soil moisture, which is considered an important factor controlling soil processes occurring in semi-arid conditions. The enzymes considered in our study were involved in C cycle; thus, it is important in understanding the C dynamics in such semi-arid forests. The study also focused on correlation among enzyme activities and soil respiration and a significant positive correlation of enzymatic activities (dehydrogenase and phenol oxidase) suggested that they play a significant role in soil respiration. This study enlarges the knowledge about different microbiological processes and their dynamics which are important in understanding their role in C cycling and C dynamics in semi-arid ecosystems. Although semi-arid land plays a small role in carbon dynamics, globally, it represents one-third of terrestrial habitat and thus sensitive to large-scale environmental changes.

Abbreviations
ABTS: 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid); BA: β-glucosidase activity; C: Carbon; DHA: Dehydrogenase activity; FSI: Forest Survey of India; HCl: Hydrochloric acid; MBC: Microbial biomass carbon; NCT: National capital territory; PC: Principal component; PCA: Principal component analysis; PNG: p-Nitrophenyl-β-D-glucoside; PNP: p-Nitrophenol; PO: Phenol oxidase activity; RESP: Soil respiration; SM: Soil moisture; SOC: Soil organic carbon; ST: Soil temperature; THAM: Tris-hydroxymethyl aminomethane; TPF: Tri-phenyl formazan; TTC: 2,3,5-triphenyltetrazolium chloride

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Authors’ contributions
RB designed the experiment and methodology, scrutinized the whole data, and helped in writing the manuscript. UT performed the experiment, collected and analyzed the data, and led to the writing of the manuscript.

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Availability of data and materials
The datasets analyzed during the current study are not publicly available but are available from the corresponding author on reasonable request.

Ethics approval and consent to participate
No existing ethics and consent of interests.

Consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

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