Soil CO2 and CH4 concentration dynamics and their relationships with soil physicochemical properties, soil enzyme activity, and root biomass under shallow groundwater level

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Abstract

Soil CO2 and CH4 concentrations are crucial determinants of crop physiology and soil environment. This study aimed to investigate the dynamics of soil CO2 and CH4 concentrations and their correlations with soil nutrient content, enzymatic activities and root biomass at shallow groundwater levels. Lysimeter experiments were conducted at five groundwater depths (20, 40, 50, 60, and 80 cm) and three fertilizer application rates (low, 75%; normal, 100%; high, 125%). Soil CO2 and CH4 concentrations, physicochemical properties, and enzymatic activities were determined in the three growth stages of winter wheat crop, and plant biomass was measured post-harvest. Groundwater depth significantly (P [?] 0.001) affected CO2 and CH4 concentrations and root parameters, and their critical values appeared at the groundwater depth of 50–60 cm. Soil water content presented quadratic function relation with CO2 concentration, and exhibited the linear correlation with CH4 concentration. As an aerobic respiration product, soil CO2 concentration showed significant positive correlations with organic matter and total N levels, urease, phosphatase and sucrase activities, and root biomass in winter wheat. Soil CH4 concentration depending on anaerobic microbial activity showed significant correlations with soil nutrients, such as soil organic matter, total N, and available K. Fertilization significantly impacted root parameters (P [?] 0.001) and shoot biomass (P [?] 0.05) instead of CO2 and CH4 concentrations. In contrast, groundwater depth emerged as a crucial factor as it affected soil physicochemical properties, soil enzymatic activities, root respiration, and winter wheat growth at shallow groundwater levels.

Soil CO_2 and CH_4 concentration dynamics and their relationships with soil physicochemical properties, soil enzyme activity, and root biomass under shallow groundwater level

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HIGHLIGHTS:

1) Critical values focused at the groundwater depth of 50–60 cm.

2) Soil CO_2 and CH_4 concentrations were affected by groundwater depth rather than fertilization level.

3)Soil CO_2 concentration was significantly positively correlated with root biomass, soil nutrients and enzyme activities.

4) Soil CH_4 concentration was only correlated with soil moist and nutrients.

Abstract

Soil CO_2 and CH_4 concentrations are crucial determinants of crop physiology and soil environment. This study aimed to investigate the dynamics of soil CO_2 and CH_4concentrations and their correlations with soil nutrient content, enzymatic activities and root biomass at shallow groundwater levels. Lysimeter experiments were conducted at five groundwater depths (20, 40, 50, 60, and 80 cm) and three fertilizer application rates (low, 75%; normal, 100%; high, 125%). Soil CO_2 and CH_4 concentrations, physicochemical properties, and enzymatic activities were determined in the three growth stages of winter wheat crop, and plant biomass was measured post-harvest. Groundwater depth significantly (P [?] 0.001) affected CO_2 and CH_4 concentrations and root parameters, and their critical values appeared at the groundwater depth of 50–60 cm. Soil water content presented quadratic function relation with CO_2 concentration, and exhibited the linear correlation with CH_4 concentration. As an aerobic respiration product, soil CO_2 concentration showed significant positive correlations with organic matter and total N levels, urease, phosphatase and sucrase activities, and root biomass in winter wheat. Soil CH_4 concentration depending on anaerobic microbial activity showed significant correlations with soil nutrients, such as soil organic matter, total N, and available K. Fertilization significantly impacted root parameters (P [?] 0.001) and shoot biomass (P [?] 0.05) instead of CO_2 and CH_4 concentrations. In contrast, groundwater depth emerged as a crucial factor as it affected soil physicochemical properties, soil enzymatic activities, root respiration, and winter wheat growth at shallow groundwater levels.

KEYWORDS

groundwater depth, soil CO_2 concentration, soil CH_4concentration, root biomass, soil nutrient content

1 INTRODUCION

Soil carbon pool is the largest natural terrestrial carbon resource and is closely related to soil fertility and environmental quality (Buysse et al., 2016; Qu et al., 2018; Sun et al., 2018; Zhang et al., 2018a). Soil respiration is the primary output of the soil carbon pool (Lou et al., 2017; Tong et al., 2017), and it embodies root respiration and microbial respiration (root and mycorrhizosphere respiration, surface-litter, organic matter decomposition, and so on) (Acosta et al., 2018; Li et al., 2018). Microbial respiration represents microbial decomposition and transformation rate (Lou, Gu and Zhou, 2017; Qu, Kitaoka and Koike, 2018), whereas root respiration represents the root metabolism rate that is affected by photosynthesis, plant phenology, root biomass, carbohydrate content in plant shoots, and net primary production, etc. (Savage et al., 2013; Li et al., 2018). The C accumulation and C recycling in the soil are directly dependent on photosynthesis as photosynthate are the primary source of carbohydrates for root respiration or microbial respiration in root exudates (Savage, Davidson and Tang, 2013). Root biomass is a crucial C source in the soil. High root biomass is often associated with high root respiratory activity (Tomotsune et al., 2013), and it provides a high amount of substrate for microbial decomposition (Wertha and Kuzyakov, 2008). Briefly, soil respiration is influenced by factors, such as crop growth (root growth, photosynthetic efficiency, dry matter accumulation, etc.) and soil environmental factors (microbial activity, enzyme activity and nutrient content, etc.), and these factors, in turn, are significantly affected by soil water status and fertilizer application (Zhong et al., 2016; Li et al., 2018; Hu et al., 2019).

Soil moisture tends to be high in areas at the shallow groundwater level (Zhang et al., 2018b), and it directly or indirectly affects soil respiration. In heterotrophic respiration, CO_2 and CH_4 are the primary metabolites produced due to aerobic and anaerobic microbial decomposition, respectively (Cai et al., 2009). A high level of soil water content leads to waterlogging and reduced oxygen supply in the soil (Buysse, Flechard, Hamon

and Viaud, 2016), which inhibits aerobic microbial activity and enhances anaerobic microbial activity (Wang et al., 1996). Thus, CH_4 is replaced by CO_2 as the primary metabolite in soil. In autotrophic respiration, reduced O_2 supply to roots hampers root respiration and root growth by decreasing ATP production, energy-dependent nutrient uptake and nutrient transport (Aguilar et al., 2003). Indirect effects of soil moisture are due to anaerobic conditions resulting from high groundwater level. As demonstrated in the previous report, it retarded secretion of soil enzymes, such as urease and phosphatase, etc. by soil microbes and root cells (Pulford and Tabatabai, 1988). Regardless of soil temperature, decreased secretion of enzymes led to decreased enzymatic activity, which results in slower cycling of soil nutrients and CO_2 production rate than aerated soil (Cao et al., 2017; Zhang, Zhu, Zhou and Li, 2018b). Overall, shallow groundwater level profoundly impacted the soil respiration.

Fertilizer application regulates soil respiration by altering the soil nutrient content and enzymatic activities. Chemical fertilizer application influences soil respiration by increasing NPK availability, growth of crop roots, and microbial population in the soil (Creze and Madramootoo, 2019). Xue et al. (2014) showed that N (urea) and P (chemical fertilizer) increased the density, surface area, and dry biomass weight of crop roots as N and P play crucial roles in the allocation of photosynthates in shoots and roots. Subsequently, a higher root abundance and activity led to higher soil CO_2emission (Jurasinski et al., 2012; Sun et al., 2018). The application of urea and ammonium fertilizers increased the microbial activity leading to an increased SOC decomposition as ammonium is the preferred nitrogen source for soil microbes (Gong et al., 2011; Geisseler and Scow, 2014). Also, chemical fertilizer induced increase in root and microbial biomass increased the secretion of soil enzymes (Guan, 1986; Dick, 1997). Increased soil enzymatic activity effectively improves the recycling of soil nutrients and promotes the growth of soil root and microbes (Guan, 1986; Dick, 1997; Iovieno et al., 2009), improving the intensity of soil respiration. However, the correlation between soil enzymatic activity and soil respiration has been rarely explored.

In general, the fertilizer application could increase soil respiration by increasing soil fertility. However, contrasting outcomes were also reported. Zhu et al. (2016) reported that low concentration of N supplementation increased soil respiration, underground plant biomass content, and soil microbial biomass carbon, whereas high concentration of N supplementation mitigated these factors in the grassland. An appropriate fertilizer application rate is crucial in regulating soil environmental conditions for crop root and microbial growth, which are closely related to soil respiration.

Shallow groundwater level exists widely in Central China in which is one of the main regions for winter wheat production in China. In the winter wheat growing season, due to the high intensity of rainfall and poor drainage conditions (Ren et al., 2016), crop productivity is frequently affected due to abiotic stress. Shallow groundwater level alters the soil environmental conditions, such as physical, biological, and chemical properties, associated with the crop roots and microbial growth, and soil biochemical reactions, resulting in the differences of soil nutrient cycle and crop growth characteristics in the regions between shallow and normal groundwater level (Grimaldi et al., 2015). It is a well-known fact that appropriate drainage and fertilizer application decreases waterlogging and improves soil environmental conditions. However, the complex correlation between groundwater level, fertilization level, root growth, enzymatic activity, and soil respiration and their interaction mechanisms in shallow groundwater level soil, remains ambiguous.

In this study, we aimed to 1) determine the effects of groundwater depth and fertilizer level on soil CO_2 and CH_4concentrations in different stages of winter wheat growth in shallow groundwater level and 2) investigate the correlation of soil CO_2 and CH_4 concentrations with soil nutrient content, enzymatic activity, and root biomass, to better understand the influence of shallow groundwater level on soil environmental conditions.

2 MATERIALS AND MEHTODS

2.1 Experimental site and treatments

The experiment was performed at the Experimental Station of Yangtze University (Latitude, 30°21'N; Longitude, 112°09'E; Elevation, 31.8 m above sea level) in Jingzhou, Hubei, China. It is a subtropical humid monsoon region with a rainy spring and summer with mean annual precipitation and air temperature of

1100 mm and 16.7°C, respectively. The mean monthly rainfall increases from 29.6 mm in January to 159.9 mm in June. Groundwater depth in the experimental region is around 50 cm on average, total salinity in the groundwater is less than 1 g L^{-1}, and pH is 6.7–8.9. The soil is yellow brown paddy and loamy containing 22% clay (0–2 μm), 75% silt (2–50 μm), and 3% sand (50–2000 μm), respectively.

The experiments were conducted in micro-lysimeters that were 112 cm deep and 70 cm in diameter. The micro-lysimeters were evenly filled layer by layer with soil collected from a local farm field at the bulk density of 1.27 g cm^{-3}. The micro-lysimeter's groundwater level at the depths of 20, 40, 50, 60, and 80 cm in the soil surface was automatically controlled by using water inlet and outlet apparatus (Figure 1). Soil filled micro-lysimeters were employed to estimate the initial contents of organic matter, total N, available P, available K, and soil pH value (soil: water ratio of 1: 2.5), which were found to be 8.63 g kg^{-1}, 1.29 g kg^{-1}, 16.90 mg kg^{-1}, 153.76 mg kg^{-1}, and 7.8, respectively.

Winter wheat (*Triticum aestivum* L.) was sown on October 28, 2015, at a density of 210 plants m^{-2}, and harvested on May 4, 2016. During the wheat growth period, compound fertilizer containing N: P_2O_5: K_2O in the ratio of 14: 16: 15 and urea with 46% N content were applied thrice before sowing, at the seedling stage and the jointing stage, and the mass proportion of chemical fertilizer applied at the three stages of wheat growth was 7: 1: 2. In line with the local practices, the application rates of N, P, and K in the whole growth period of winter wheat were 180, 65, and 60 kg ha^{-1}, respectively, which were designated as the normal fertilization level treatment (NF) in this study. The rainfall and mean daily air temperature were 388 mm and 12.2°C, respectively, in the winter wheat growth period during 2015–2016. As per the weather records (1952–2016), this region received normal rainfall throughout the year with a drought index of 0.34 during the winter wheat growth period (Zhang, Zhu, Zhou and Li, 2018b). No supplementary irrigation was provided during the whole growth period of winter wheat.

Experimental treatments included five groundwater depths (20, 40, 50, 60, and 80 cm) and three fertilization application rates (low, normal, and high). The fertilizer application rates for the low fertilization level and the high fertilization level were 75% (75% NF) and 125% (125% NF) of the standard fertilizer application rate (NF), respectively. All experimental treatments in the study were replicated three times.

2.2 Observation indexes and measurement methods

Geothermometers set to16:00 were used for measuring soil temperature at the 5, 10, 15, and 20 cm depth per day (Figure 1). The soil was sampled at the depths of 0–10 and 10–20 cm during jointing (March), heading (April), and grain filling (May) stages of a wheat growth three times. Soil water content, organic matter content, pH value, soil nutrient content (N, P, and K), and the soil enzymatic activities (urease, alkaline phosphatase, sucrase) were estimated. To calculate soil water content, an electric oven set to 105°C was used, for calculating soil organic matter content, titration based wet combustion method was used, and for calculating soil pH value in 1: 2.5 soil-water extract, a pH meter was used (FG3-ELK, Mettler-Toledo International Trading Co., Ltd, Shanghai, China). Available soil P content was assayed spectrophotometrically (UV-5500PC Spectrophotometer, Shanghai Metash Instrument Co., Ltd., China), total soil N content was determined using an automatic Kjeldahl apparatus (K9840, Hanon Instrument, Jinan, China), and available K content was determined using a flame photometer (FP640, Shanghai INESA Scientific Instrument Co., Ltd., China). Urease, alkaline phosphatase, and sucrase activities were assayed as per Guan's methods (Guan, 1986). The released NH_4^+ was determined using 10% aqueous urea as substrate, incubated at 37°C for 24 h, and absorbance was measured spectrophotometrically at 578 nm wavelength. The urease activity was expressed as mg NH_4^+-N (g soil 24 h)$^{-1}$. For the determination of alkaline phosphatase activity, the disodium phenyl phosphate solution was used as a substrate and incubated at 37°C for 24 h. The resulting phenol formation was determined spectrophotometrically at 600 nm wavelength, and alkaline phosphatase activity was expressed as mg phenol (g soil 24 h)$^{-1}$. For determination of sucrase activity, sucrose solution was used as the substrate and incubated at 37°C for 24 h. After incubation, this solution was filtered, and the filtrate was boiled with 3 mL of 3, 5-dinitrosalicylic acid (DNS) in a water bath for 5 min. The absorbance of the reducing sugars was measured at 508 nm wavelength, and sucrase activity was expressed as mg of glucose (g soil 24 h)$^{-1}$.

A syringe suction device was used to sample soil at the 0–5, 5–10, 10–20, and 20–40 cm depth to collect soil gas. A rubber hose with small pinholes was inserted into the above-mentioned soil depth with the soil-filled lysimeter (Figure 1). Hose with 4 mm inner radius and 1 mm thick wall and upper port sealed with rubber plug to maintain tension after need puncture was used. The soil gas was sucked from the hose using a needle and injected into a special vacuum bottle. CO_2 and CH_4 concentrations were measured with a gas chromatograph (7890A, Agilent Technologies, Inc., Wilmington, USA) and expressed as mL/L and μL L^{-1}, respectively.

Plant height, tillering rate, and the number of leaves were calculated at various winter wheat growth stages. After harvesting, root traits were measured for the entire intact root system extracted from the micro-lysimeter and treated individually for each experimental replication. Cleaned fresh roots were scanned using EPSON perfection V700 Photo (Epson America, Inc. Long Beach, USA) and analyzed with WinRHIZO 2009 (Regent Instruments Inc. Quebec, CA), and root diameter and root length density were averaged and expressed as mm and cm cm^{-3}, respectively.

2.3 Statistical analysis

One-way analysis of variance (ANOVA) was used to calculate significant differences between different treatments using SPSS version 21.0 (SPSS Inc. Chicago, USA). A statistically significant ANOVA F -value was used to perform the least significant difference test (significance level of $P = 0.05$) for the separation of the means. Simple linear regression and curve estimation analyzed the correlation between the soil CO_2 and CH_4 concentrations and soil water content, root biomass, soil nutrient contents, soil enzymatic activities. For Pearson correlation analysis, $P = 0.05$ was considered as statistically significant.

The average values of soil CO_2 and CH\sout$_4$ concentrations and soil water content, organic matter content, soil nutrient content, soil enzymatic activities at 0–20 cm soil depth were used to analyze the correlations between soil CO_2 and CH_4 concentrations and the rest of the factors. The CO_2 and CH_4concentrations at 0–40 cm soil depth were averaged and employed when wheat root biomass was involved in the establishment of correlations.

3 RESULTS

3.1 Root parameters

As depicted in Figure 2, total root biomass, mean root diameter, and root length density first increased and later decreased with increasing groundwater depth. The maximum values of root biomass and root length density were recorded at the groundwater depth of 60 cm, and maximum values of mean root diameter were recorded at the groundwater depth of about 50 cm (Figure 2). It indicated that root diameter expansion was impacted more by the water shortage than root biomass accumulation and root extension. The root biomass and mean root diameter were positively correlated to the fertilization level (Figure 2A–B). Out of all the three fertilization levels tested, root length density in the normal fertilization level was 11.8–25.4% and 9.2–12.5% higher than that in the high and the low fertilization levels, respectively.

3.2 Shoot biomass and root-shoot ratio

Identical variation trends were observed in shoot and root biomass values pertaining to groundwater depth and fertilization levels (Figure 3A). Shoot biomass increased with the declining groundwater level until the groundwater depth of 60 cm, and then it decreased with the further increase of groundwater depth (Figure 3A). A higher fertilization level led to higher shoot biomass; however, it was not statistically significant (Figure 3A).

The root-shoot biomass ratio under low fertilization level was significantly correlated to the groundwater depth (Figure 3B). A linear correlation was observed under normal and high fertilization levels between root-shoot biomass ratio and groundwater depth till groundwater depth reached 60 cm; however, root-shoot biomass ratio and groundwater depth exhibited an inverse correlation at groundwater depth > 60 cm (Figure 3B). It suggested that the groundwater level (> 60 cm) impacted root growth more than shoot growth in

wheat plants (Figure 3B). Root-shoot biomass ratio decreased with increasing soil fertilization level (Figure 3B).

3.3 Soil CO_2 concentration

Variation in soil CO_2 concentration with soil depth during three winter wheat stages at the different groundwater depths and fertilization levels is depicted in Table 1. With the increasing duration of the wheat growth, average soil CO_2concentration for all sampling depths first increased and later decreased, and maximum values appeared during the heading stage of vigorous wheat growth (Table 1). It suggested an enhanced effect of root and microbial respiration on CO_2 emission during the vigorous wheat growth stage. Soil CO_2 concentration increased with soil depth due to atmospheric gas exchange (Table 1).

The CO_2 concentration in shallow ([?] 10 cm) soil depth first decreased and later increased with the increasing groundwater depth, and the minimum CO_2 concentration value appeared at the groundwater depth of 50–60 cm (Table 1). When the sampling depth was $>$ 10 cm, soil CO_2 concentration decreased with increasing groundwater depth (Table 1). The fertilization level did not show any significant effect on soil CO_2 concentration. Average values of soil CO_2 concentration at three different fertilization levels were 1.26–1.33, 2.55–2.63, and 1.48–1.59 mL/L during the jointing, heading and filling stages of the wheat growth period, respectively, under experimental conditions (Table 1).

3.4 Soil CH_4 concentration

Soil CH_4 concentration exhibited slight seasonal variations (Table 2) indicated by the lower values during the heading stage compared to other wheat growth stages. Soil CH_4concentration decreased with decreasing groundwater depth (Table 2). The significantly higher CH_4 concentration was observed at the sampling soil depth closer to the groundwater surface. However, soil CH_4 concentration did not change significantly with soil depth and fertilization levels. The differences of CH_4 concentration values at soil depths and fertilization levels were less than 7.3% and 13.7%, respectively.

4 DISCUSSION

4.1 Seasonal variations

Soil CO_2 concentration was highest at the heading stage of wheat growth (Table 1). The seasonal soil respiration variation is primarily caused by the soil temperature (Hu et al., 2019). Soil CO_2 is primarily derived from soil respiration that includes heterotrophic respiration and autotrophic respiration. Soil temperature exerts a significant effect on heterotrophic respiration as the microbes that contribute to heterotrophic respiration are highly sensitive to soil temperature (Zhong, Yan, Zong and Shangguan, 2016; Qu, Kitaoka and Koike, 2018). Besides, microorganisms led nutrient transformation, and biochemical processes are also dependent on the soil temperature (Guan, 1986; Qu, Kitaoka and Koike, 2018). Root exudation and root respiration rate, which serve as crucial drivers of autotrophic respiration, depends on plant growth and thus are closely associated with temperature (Liu et al., 2015; Tong, Li, Nolan and Yu, 2017). The correlations of heterotrophic and autotrophic respirations with temperature during the winter wheat growth period are linear and unimodal curve, respectively (Zhang et al., 2013). Autotrophic respiration was found to be more sensitive to change in temperature than heterotrophic respiration (Zhang, Lei and Yang, 2013). The synergistic effect of autotrophic and heterotrophic respiration resulted in the higher soil CO_2 concentration at 20°C of soil temperature, which was close to the average temperature during the heading growth stage of winter wheat (Zhang, Lei and Yang, 2013; Liu et al., 2015; Zhong, Yan, Zong and Shangguan, 2016; Tong, Li, Nolan and Yu, 2017).

CH_4 concentration did not change obviously at different growth stages of winter wheat (Table 2). It might be due to the insignificant correlation between CH_4 concentration and soil temperature. As shown in previous studies, high-temperature stimulated the activity of methane-producing microbes, and CH_4 emission increased with temperature until the temperature reached 34.5°C (Cai, Xu and Ma, 2009). However, CH_4 oxidation also increased with soil temperature, and the optimum temperature for maximum CH_4 oxidation was found to be 20–30°C (Cai, Xu and Ma, 2009; Jassal et al., 2011). Furthermore, oxidation and transport

of CH_4, which are primarily influenced by soil gas diffusivity, serve as the crucial factors that influence CH_4 concentration (Cai, Xu and Ma, 2009)(Cai, 2009 #161;Cai, 2009 #2120). As per the outcomes of the current study, altered soil CH_4 concentration might be more dependent on gas diffusivity than soil temperature, in line with the findings by Jassal, Black, Roy and Ethier (2011).

4.2 Effects of groundwater depth

In this study, the statistical analysis of experimental data indicated that groundwater depth and root parameters were significantly correlated (P [?] 0.001) (Table 3). Higher root parameter values appeared at 50–60 cm of groundwater depth (Figure 2). The root system of winter wheat was mainly distributed at 10–35 cm soil depth (Hodgkinson et al., 2017). A shallow groundwater level creates a waterlogging environment, which affects wheat root growth and shoot biomass adversely (Celedonio et al., 2017), and inhibits wheat root respiration due to insufficient oxygen supply (Hodgkinson et al., 2017; Chen et al., 2018). On the other hand, deep groundwater level mitigates the absorption and utilization of groundwater by crops. Development of root system plasticity promoted root extension at higher soil depth and increased the water access from deeper soil layer with higher water content (Becker et al., 2015; Ali et al., 2018). Shoot biomass was insignificantly affected by the groundwater depth (Table 3). However, the root-shoot biomass ratio, which is an index for differential investment between above-ground and underground biomass, was significantly affected by groundwater depth (Table 3). A higher root-shoot biomass ratio was observed at the groundwater depth of 60 cm. It demonstrated that the adverse effect of the groundwater level on wheat root growth was higher than shoot growth at the groundwater level that was deeper than 60 cm.

The concentrations of soil CO_2 and CH_4were significantly correlated with groundwater depth (P [?] 0.001) (Table 3). Groundwater depth was correlated to soil water status and soil aeration conditions, and it substantially influenced production, emission, and accumulation of CO_2 and CH_4 by affecting soil microbial activity, enzymatic activity, nutrient cycling, and so on (Buysse, Flechard, Hamon and Viaud, 2016; Zhang et al., 2018a; Hu et al., 2019). Higher shallow groundwater levels increase soil water content significantly by reducing pore space for soil gas and creating anaerobic conditions (Wang and Lu, 2006). It adversely affected the growth of aerobic microorganisms, and enhanced anaerobic microorganisms' activity. However, a deeper groundwater level also inhibits the growth of aerobic microbes that relies on water for metabolism (Liu et al., 2015). Consequently, soil CO_2 concentration had a quadratic function relation (P [?] 0.001) with soil water content (Figure 4A), and soil CH_4 concentration (Figure 5A) were found to be linearly correlated to soil water content (P [?] 0.001).

Soil CO_2 production depends on crop root growth. Root respiration represents the metabolism of root cells and respiratory activity, and higher root biomass showed a higher potential in increasing autotrophic root respiration (Tomotsune, Yoshitake, Watanabe and Koizumi, 2013). Meanwhile, greater root residual input provided more C to rhizospheric microbes, which enhanced C decomposition and heterotrophic microbial respiration (Wertha and Kuzyakov, 2008). These findings were validated by the significant positive correlation (P [?] 0.001) between the soil CO_2 concentration and root biomass (Figure 6). However, CH_4 concentration did not show a significant correlation with root biomass as CH_4 is not the product of root cell metabolism. Therefore, CH_4 concentration was not significantly correlated to the growth status of crop root system.

Organic matter and total N in soil were found to be closely correlated with CO_2 and CH_4 production. It was further validated by the positive correlations between CO_2 (Figure 4B–C), CH_4 (Figure 5B–C), and organic matter, total N. Increased organic matter content increased CO_2 and CH_4 emissions. Soil CO_2 and CH_4 from soil respiration are derived from microbial decomposition of soil organic matter (Illeris et al., 2003; Wertha and Kuzyakov, 2008; Li et al., 2013). A higher soil organic matter resulted in a higher microbial population in the soil. However, in this study, the correlations of CO_2 and CH_4 concentrations with organic matter and total N contents might be due to the effect of groundwater depth. Previous studies have demonstrated that the levels of organic matter and total N, the crucial soil nutrient, were lowest at groundwater depth of 60 cm, and the enzymatic activities were highest due to the suitable soil moisture (Zhang, Zhu, Zhou and Li, 2018b). The CO_2 and CH_4 concentrations in soil should increase due to the fast nutrient cycling but decreased near groundwater depth of 60 cm (Liu et al., 2015; Zhang, Zhu, Zhou and Li, 2018b). It might

be due to the improved soil structure and the enhanced soil aeration conditions (Wang and Lu, 2006). High level of soil water content can deteriorate soil structure and make the soil denser to trap CO_2 and CH_4, resulting in a high CO_2 and CH_4 concentrations in the soil (Yang et al., 2013).

Soil P and K, crucial soil elements involved in protein synthesis, cation-anion balance, enzyme activation, and so on, are influenced by the groundwater depth (Kering et al., 2012). However, soil P and K did not show an apparent effect on soil CO_2 concentration under the experimental conditions (data not shown). Nevertheless, available K content showed a positive linear correlation (P [?] 0.001) with CH_4 concentration (Figure 5D). It might be due to K^+ led inhibition of CH_4absorption in soil. The K^+ concentration in soil solution increased osmotic pressure in methane-oxidizing microbial cells, inhibiting CH_4 oxidation, and increasing the CH_4 concentration in soil (Cai, Xu and Ma, 2009).

Soil enzyme activity involves in soil nutrient cycling. However, only phosphatase was affected by the groundwater depth, while all the three enzymes (urease, phosphatase and sucrase) were significantly affected by the fertilization level (Zhang, Zhu, Zhou and Li, 2018b). The correlation between the CO_2 and CH_4concentrations with soil enzymatic activities is discussed below.

4.3 Effects of fertilization level

Currently, chemical fertilizer plays a vital role in meeting the increasing demand for staple grain. Appropriate nitrogen fertilizer application promotes photosynthesis, a strong root system for higher nutrient absorption (Olmo et al., 2015; Hirte et al., 2018), thus increasing dry matter accumulation (Jiang et al., 2008). In this study, fertilizer application resulted in increased wheat root biomass (P [?] 0.001), mean root diameter (P [?] 0.001), and shoot biomass (P [?] 0.01) due to higher nutrients in soil (Table 3) (Ali et al., 2018; Hirte, Leifeld, Abiven and Mayer, 2018). However, excessive N application not only increased the resource wastage and non-point source pollution, but reduced crop root length density, adversely impacting plant biomass and grain yield (Chen et al., 2018). Also, fertility deficit certainly decreased photosynthetic activity and crop efficiency, hindering crop and root system growth (Chen et al., 2016). These two aspects might explain the increased root length density in treatment involving normal fertilization application as compared to the other two fertilization treatments under the experimental conditions (Figure 2C).

Fertilizers, especially N fertilizers, significantly affected soil respiration (Zhu et al., 2016; Creze and Madramootoo, 2019). However, as per the current study, the fertilizer application rate affected the soil CO_2 and CH_4 concentrations insignificantly (Table 3). Nitrogen supplementation inhibits microbial heterotrophic respiration in soil by suppressing soil microbial biomass but stimulate root respiration (Wang et al., 2016). However, as per the previous report, insignificant changes in heterotrophic and soil respiration after N fertilization application did not affect microbial biomass significantly (Liu et al., 2015; Zhong, Yan, Zong and Shangguan, 2016). Thus, the precise mechanism for the effect of N fertilizer application on soil respiration needs further investigation.

Increased total N content led to a decreased C/N ratio and improved soil CO_2 flux (Bellingrath-Kimura et al., 2015; Pires et al., 2017). Furthermore, soil N/P ratio also influenced autotrophic respiration and microbial activity. A balanced N/P ratio increased root biomass accumulation and soil CO_2 concentration (Bellingrath-Kimura et al., 2015; Pires et al., 2017; Sun et al., 2018). Nitrogen fertilizer application enhanced the root respiration rate by increasing the availability of soil nutrients, the N content in root, and photosynthate allocation below the ground (root biomass) (Sun et al., 2018). This maybe partially resulted in the significant correlation (P [?] 0.001) of soil CO_2 concentration with total N content (Figure 4B).

In addition to soil nutrients, the activities of soil enzymes, i.e., urease (P [?] 0.001), phosphatase (P [?] 0.01), and sucrase (P [?] 0.001), were significantly influenced by fertilization levels (Zhang, Zhu, Zhou and Li, 2018b) and linearly correlated to soil CO_2concentration (Figure 7). Urease, phosphatase, and sucrase in the soil are mainly secreted by aerobic microbes in the soil and root cells (Guan, 1986; Wang and Lu, 2006). It might be the reason that CH_4 concentration was not correlated with the soil enzymatic activities. Soil enzymatic activity can be used as an index of microbial activity for expressing the soil respiration intensity (Iovieno, Morra, Leone, Pagano and Alfani, 2009; You et al., 2018). Catalysis of soil enzymes could

accelerate the microbial decomposition of soil organic matter (Iovieno, Morra, Leone, Pagano and Alfani, 2009; Xiao et al., 2016). Also, high soil nutrient cycling rate increased the plant organic matter accumulation and root growth, which in turn increased the soil root respiration. Thus, the close correlations between soil CO_2 concentration and soil enzymatic activities might be the outcome of the synergistic effects of fertilizer level on soil nutrition content, microbial activity, and crop root growth. However, the contribution of soil enzymatic activities to the CO_2 concentration could be so small that CO_2 concentration was not affected by the fertilization level (Table 3).

5 CONCLUSIONS

Groundwater depth altered soil moisture and thus significantly affected root parameters, root-shoot biomass ratio, and the concentrations of soil CO_2 and CH_4. The highest root parameters and root-shoot biomass ratio were observed at the groundwater depth of 50–60 cm. For CO_2 and CH_4concentrations, the critical values of gas concentration appeared at the groundwater depth of 50–60 cm. The significant correlations of nutrient contents with CO_2 and CH_4 concentrations and the positive correlation between CO_2 concentration with root biomass validated the effect of groundwater depth mediated by soil moisture content and aeration condition.

Fertilization level significantly affected the root parameters and shoot biomass, and the appropriate application of fertilizer promoted the growth of crop roots and biomass matter accumulation. Fertilization levels affected root parameters, soil nutrient, and enzymatic activity, which were closely related to soil CO_2 concentration. The outcomes of the study showed that CO_2 and CH_4 concentrations were independent of fertilization levels.

In conclusion, crop growth was significantly affected by fertilization and groundwater depth, and soil respiration was significantly affected only by groundwater depth. It suggested 50–60 cm as the optimal groundwater depth for better soil respiration, root growth, matter accumulation, and distribution in winter wheat crop.

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CONFLICTS OF INTERESTS

The authors have no financial or personal conflicts of interest to declare.

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SD /cm	GD /cm	75%NF	75%NF	75%NF	NF	NF	NF	125%NF	125%NF
		Jointing	Heading	Filling	Jointing	Heading	Filling	Jointing	Heading
0–5	20	0.95±0.02 ab	0.81±0.01 a	0.93±0.00 a	1.30±0.02 a	1.12±0.10 a	1.02±0.09 a	1.67±0.11 a	0.88±0.11 a
	40	1.16±0.09 cd	0.81±0.11 a	0.79±0.07 a	1.37±0.11 a	0.64±0.04 b	0.89±0.07 ab	2.00±0.13 a	0.65±0.26 a
	50	1.06±0.04 bc	0.74±0.00 ab	0.74±0.06 a	0.77±0.02 b	0.61±0.01 b	0.74±0.03 b	1.09±0.10 b	0.59±0.04 a
	60	1.25±0.00 d	0.62±0.05 b	0.78±0.08 a	1.00±0.14 c	0.66±0.08 b	0.90±0.05 ab	1.22±0.18 bc	0.91±0.38 a
	80	0.82±0.01 a	0.73±0.03 ab	0.79±0.03 a	1.36±0.02 a	1.19±0.08 a	0.95±0.07 ab	1.56±0.17 ac	1.08±0.23 a
5–10	20	1.37±0.09 ab	1.61±0.09 ab	1.43±0.16 a	1.40±0.05 a	2.46±0.14 a	1.48±0.11 a	1.24±0.04 ab	1.41±0.05 a
	40	1.48±0.14 a	1.87±0.14 a	1.34±0.05 a	1.30±0.39 a	1.67±0.04 b	1.47±0.06 a	1.27±0.11 ab	0.70±0.04 b
	50	1.32±0.05 ab	1.43±0.09 bc	0.92±0.07 b	1.31±0.09 a	1.23±0.06 c	1.04±0.02 b	1.47±0.04 a	0.91±0.12 b
	60	1.02±0.02 c	1.02±0.09 d	0.90±0.08 b	1.23±0.02 a	1.14±0.12 c	1.01±0.09 b	1.17±0.00 bc	0.88±0.11 b
	80	1.15±0.04 bc	1.22±0.09 cd	1.00±0.03 b	1.35±0.15 a	1.81±0.43 a	1.11±0.08 b	1.38±0.19 c	1.57±0.05 a
10–20	40	1.86±0.14 a	4.11±0.28 a	2.47±0.05 a	2.02±0.06 a	4.43±0.35 a	2.18±0.18 ab	1.32±0.04 a	4.44±0.10 a
	50	1.58±0.15 ab	3.22±0.24 b	1.88±0.07 b	1.66±0.02 b	3.04±0.55 b	1.80±0.11 ab	1.21±0.15 a	4.09±0.18 ab
	60	1.30±0.13 bc	2.48±0.05 b	1.33±0.09 bc	0.92±0.09 c	3.05±0.53 b	1.69±0.00 b	1.22±0.01 a	3.48±0.16 b
	80	0.78±0.05 c	2.85±0.24 b	1.67±0.25 c	0.62±0.04 d	3.19±0.23 b	1.57±0.02 b	0.58±0.03 b	3.55±0.16 bc
20–40	50	2.12±0.02 a	6.84±0.38 a	2.90±0.63 a	2.10±0.30 a	6.74±0.68 a	3.52±0.09 a	1.82±0.24 a	6.50±0.43 a
	60	1.35±0.12 b	6.70±0.23 a	2.67±0.41 a	1.00±0.11 b	6.18±0.61 a	2.82±0.17 b	1.40±0.21 a	6.11±0.31 a
	80	0.83±0.07 c	6.28±0.06 a	2.56±0.30 a	0.73±0.04 c	5.58±0.07 a	2.79±0.00 b	0.93±0.19 b	5.57±0.32 a

Table 1 Variation of soil CO_2 concentrations (mL L^{-1}) averaged for with soil depth (SD) at the jointing, heading, and filling stages of wheat growth period under the fertilization levels of 75% (75% NF), 100% (NF) and 125% (125% NF) of the normal fertilizer application rate and the groundwater depths (GD) of 20, 40, 50, 60, and 80 cm. The values shown are the means of three replicates ± standard deviations. Within a treatment, the means followed by different lowercase letters are significantly different at$P$ [?] 0.05 as determined by the LSD test

SD /cm	GD /cm	75%NF	75%NF	75%NF	NF	NF	NF	125%NF	125%NF
		Jointing	Heading	Filling	Jointing	Heading	Filling	Jointing	Heading
0–5	20	2.43±0.05 a	2.24±0.06 a	2.63±0.04 a	2.40±0.07 a	2.41±0.10 a	2.39±0.00 a	2.49±0.03 bc	2.34±0.04 a
	40	2.41±0.11 a	2.20±0.03 ab	2.37±0.12 b	2.34±0.05 ab	2.17±0.01 b	2.31±0.06 ab	2.42±0.06 bc	2.18±0.01 bc

SD /cm	GD /cm	75%NF	75%NF	75%NF	NF	NF	NF	125%NF	125%NF
	50	2.36±0.08 a	2.16±0.03 ab	2.26±0.04 b	2.33±0.05 ab	2.17±0.01 b	2.29±0.05 b	2.34±0.07 a	2.15±0.05 bc
	60	2.31±0.01 a	2.14±0.05 b	2.26±0.01 b	2.29±0.00 b	2.16±0.05 b	2.28±0.01 b	2.31±0.09 ab	2.12±0.02 b
	80	2.32±0.09 a	2.15±0.02 b	2.27±0.01 b	2.28±0.02 b	2.12±0.04 b	2.27±0.03 b	2.30±0.01 c	2.17±0.02 bc
5–10	20	2.44±0.07 ab	2.24±0.06 a	2.51±0.12 a	2.46±0.06 a	2.44±0.03 a	2.30±0.09 a	2.41±0.04 a	2.44±0.03 a
	40	2.43±0.05 a	2.17±0.01 b	2.33±0.13 a	2.32±0.05 b	2.18±0.03 b	2.30±0.03 a	2.41±0.06 a	2.14±0.01 bc
	50	2.37±0.08 b	2.16±0.01 b	2.29±0.03 a	2.32±0.03 a	2.17±0.02 b	2.28±0.01 a	2.31±0.03 a	2.17±0.02 b
	60	2.32±0.04 ab	2.14±0.01 b	2.25±0.05 b	2.31±0.00 a	2.14±0.01 b	2.25±0.01 a	2.31±0.01 b	2.12±0.01 c
	80	2.30±0.01 b	2.15±0.02 b	2.22±0.02 a	2.31±0.05 a	2.16±0.02 b	2.22±0.04 a	2.26±0.04 b	2.16±0.02 b
10–20	40	2.47±0.02 a	2.19±0.02 a	2.45±0.07 a	2.34±0.09 a	2.19±0.04 a	2.27±0.01 a	2.37±0.05 a	2.18±0.03 a
	50	2.42±0.11 a	2.15±0.03 b	2.28±0.04 a	2.30±0.06 a	2.17±0.02 a	2.27±0.02 a	2.38±0.04 a	2.16±0.02 ab
	60	2.32±0.06 ab	2.13±0.01 b	2.27±0.06 b	2.40±0.00 a	2.16±0.01 ab	2.31±0.03 a	2.44±0.05 a	2.11±0.02 b
	80	2.30±0.02 b	2.14±0.03 b	2.26±0.03 a	2.31±0.02 a	2.12±0.02 b	2.30±0.02 a	2.28±0.05 b	2.17±0.02 a
20–40	50	2.43±0.06 ab	2.15±0.04 a	2.37±0.03 a	2.45±0.04 a	2.14±0.08 a	2.24±0.04 a	2.45±0.04 a	2.12±0.02 a
	60	2.39±0.01 a	2.13±0.03 a	2.24±0.01 a	2.33±0.04 b	2.13±0.01 a	2.25±0.02 a	2.39±0.02 b	2.14±0.04 a
	80	2.30±0.09 b	2.10±0.01 a	2.21±0.04 b	2.33±0.06 b	2.14±0.03 a	2.26±0.01 a	2.30±0.00 c	2.17±0.04 a

Table 2 Variation of soil CH_4 concentrations (μL L^{-1}) averaged for with soil depth (SD) at the jointing, heading, and filling stages of wheat growth period under the fertilization levels of 75% (75% NF), 100% (NF) and 125% (125% NF) of the normal fertilizer application rate and the groundwater depths (GD) of 20, 40, 50, 60, and 80 cm. The values shown are the means of three replicates ± standard deviations. Within a treatment, the means followed by different lowercase letters are significantly different at$P$ [?] 0.05 as determined by the LSD test

Table 3 ANOVA statistical significance of between-subject effects and within-subject effects for groundwater depth (GD) and fertilization levels (FL) on soil CO_2 and CH_4 concentrations, root parameters, shoot biomass, and root-shoot ratio. *, **, and *** indicate statistical significance respectively at 0.05, 0.01, and 0.001 probability levels; NS indicates no significance at 0.05 statistical level

Items	Soil CO_2 concentration	Soil CH_4 concentration	Root biomass	Root diameter	Root length density	Shoo
GD	***	***	***	***	***	NS
FL	NS	NS	***	***	***	**
GD×FL	NS	NS	NS	NS	NS	NS

FIGURE 1 Experimental set-up for soil temperature monitoring and soil gas sampling in a lysimeter

FIGURE 2 Variations of root biomass (A), mean root diameter (B), and root length density (C) with groundwater depth at 75% (75% NF), 100% (100% NF), and 125% (125% NF) of normal fertilizer application rate

FIGURE 3 Variations of shoot biomass (A) and root-shoot biomass ratio (B) at 75% (75% NF), 100% (100% NF), and 125% (125% NF) of normal fertilizer application rate

FIGURE 4 Relationships between soil CO_2concentration soil water content with (A), organic matter content (B), and total N concentration (C) averaged in the soil depth of 0–20 cm during growing season for all tested groundwater levels and fertilization levels. *** indicate statistical significance at the 0.001 probability level

FIGURE 5 Relationships between soil CH_4concentration soil water content with (A), organic matter content (B), total N (C) and available K (D) concentrations averaged in the soil depth of 0–20 cm during growing season for all tested groundwater levels and fertilization levels. *, ** and *** indicate statistical significance respectively at the 0.05, 0.01 and 0.001 probability levels

FIGURE 6 Relationship between the averaged soil CO_2 concentration in the depth of 0–40 cm and root biomass for all tested groundwater depths and fertilization levels. *** indicate statistical significance at the 0.001 probability level

FIGURE 7 Relationships between soil CO_2concentration with activities of urease (A), phophatase (B), and sucrase (C) averaged in the soil depth of 0–20 cm during growing season for all tested groundwater levels and fertilization levels. ** and *** indicate statistical significance respectively at the 0.01 and 0.001 probability levels