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EFFECTS OF MAIZE-SOYBEAN INTERCROPPING ON SOIL RESPIRATION

JINGJING CUI, QUANGUO ZHANG[1], LINGLING YU[2], SIKANDER KHAN TANVEER[3], GUIYUAN ZHAO[1] AND QIANG GUO[*]

Tangshan Academy of Agricultural Science, Tangshan, Hebei, 063001, China

Keywords: Maize-soybean intercropping, Soil CO_2 emission rate, Carbon balance.

Abstract

Adopting appropriate cropping systems is an effective way to reduce greenhouse gas (GHG) emission, meanwhile GHG emission from farmland is a hot topic recently. In recent years, maize-soybean intercropping has been promoted in many regions in China. To explore the effects of maize-soybean intercropping on soil CO_2 emission and carbon balance, we established three planting methods, namely, maize monoculture (M), soybean monoculture (S), and maize-soybean intercropping (MS). Results showed that the cumulative emissions of soil CO_2 were M (21231 $kg \cdot hm^{-2}$) > S (19715 $kg \cdot hm^{-2}$) > MS (17321 $kg \cdot hm^{-2}$). The two-factor composite model of soil temperature and moisture content could explain the variation in soil CO_2 emission rate well, which reached 58.36-68.54%. Correlation analysis showed that the soil CO_2 emission rate was significantly correlated with peroxidase activity ($P < 0.01$). The soil carbon balance was favorable under different treatments, serving as a sink for atmospheric carbon. The soil carbon sequestration potential of M and MS treatments was significantly higher than that of S treatment ($P<0.05$), with increases of 37.11 and 34.02%, respectively. These results indicated that M and MS treatments had strong carbon sequestration potential. M treatment exhibited superior soil carbon balance compared with MS and S treatments, with increases of 29.80 and 359.31%, respectively. Therefore, maize treatment was a better planting method compared with soybean and maize-soybean treatments.

Introduction

About one-fifth of CO_2 emissions originate from soil, categorizing it as a principal greenhouse gas (GHG) source (Linquist *et al.* 2012). Soil respiration is essential for the CO_2 exchange between soil and atmosphere, accounting for more than two-thirds of total respiration in terrestrial ecosystems (Alam *et al.* 2024, Huang *et al.* 2025). Soil respiration is a complex biochemical process regulated by abiotic and (Zhong *et al.* 2016, Raquel *et al.* 2020, Propa *et al.* 2021). In addition, human factors, i,e., cultivation methods, fertilization management, planting patterns, and irrigation can affect soil respiration.

Intercropping affects the soil micro-environment and microbial activities by using excess water and nutrients in soil (Wang *et al.* 2024). Alterations in soil micro-environments and microbial activities result in modifications to soil carbon and nitrogen cycling, thereby influencing the generation and emission of GHGs from the soil. The total area of maize-soybean intercropping exceeded 20 million acres in China during 2023. However, there was a few research on the relationship between soil enzyme activity, soil moisture, soil temperature and soil CO_2 emission rate under maize-soybean intercropping mode. Therefore, this study investigated the changes in soil enzyme activity, soil moisture, soil temperature, and soil CO_2 emission rate under the maize-soybean intercropping mode. This work will provide a reference for elucidating the relationship between carbon sources and sinks, as well as the main influencing factors.

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[*]Author for correspondence: <guoqiang8081@163.com>. [1]Institute of Cereal and Oil Crops, Hebei Academy of Agriculture and Forestry Sciences, Shijiazhuang, Hebei, 050035, China. [2]Tangshan crop seed station,Tangshan, Hebei, 063001, China. [3]SAARC Agriculture Centre, Dhaka-1215, Bangladesh.

Materials and Methods

The experimental site is situated at the Tangshan Agricultural Science Research Institute farm (latitude, 39.1627 N; longitude, 118.5731 W; altitude, 12 m, Tangshan, Hebei, China), characterized by a warm temperate marine monsoon climate with annual rainfall of 500-800 mm. The experimental site consisted of sandy loam soil, and the basic physico-chemical properties are shown in Table 1.

Table 1. Basic physical and chemical properties of the tested soil.

pH	Organic matter (g/kg)	Alkali hydrolyzed nitrogen (mg/kg)	Available phosphorus (mg/kg)	Available potassium (mg/kg)
7.01	15.6	36.4	15.9	177.4

A single-factor randomized block design was used in the experiment where each treatment was repeated three times. There were nine plots, each covering 40 m^2 (2 m × 20 m). On the basis of the practical experience of Cui *et al.* (2023), the treatment was administered as follows:

Maize monoculture (M): The spacing between rows was 40 cm. Plant spacing was 33 cm. Planting density was about 7.5×10^4 plants/hectare.

Soybean monoculture (S): The spacing between rows was 40 cm. Plant spacing was 33 cm. Planting density was about 1.2×10^5 plants/hectare.

Maize-soybean strip intercropping (MS): The strip width measured 200 cm. Two rows of maize and two rows of soybeans were cultivated in an intercropping system. The distance between two rows of maize and two rows of soybeans was 40 cm. The distance between maize belt and soybean belt was 60 cm. The sowing method and row spacing for maize and soybeans were identical to those used in monoculture.

The maize variety was Junhui 521, whereas the soybean variety was Jidou 17. The two varieties were widely used in maize soybean strip intercropping in Tangshan city in China. Maize and soybean were sown and harvested simultaneously in one year. The sowing times were June 15, 2023 and June 10, 2024, and the harvesting times were October 15, 2023 and October 10, 2024, respectively.

The soil CO_2 emission rate was measured by using a soil respiration measurement system (Li-COR 8100 A, USA) following the method described by Gao *et al.* (2008). During the measurement, the breathing chamber was placed on the PVC base, and CO_2 released from the soil was collected. In the monoculture community, three PVC bases were uniformly buried between rows. In the intercropping community, three PVC bases were buried evenly at the center of crop belts, and three PVC bases were buried evenly between belts.

After sowing, the soil CO_2 emission rate was measured every 7-10 days until the crops were harvested. The measurement time was from 8:00 AM to 12:00 PM. Simultaneously, soil temperature and volumetric moisture contents in the 5-cm soil layer near the PVC base were measured by P/N-8100-201 Omega Probe and Theta Probe type ML2x, respectively. The intercropping treatment used the weighted average of the CO_2 emission rate derived from the maize belt, soybean belt, and blank area of the inter belt as the CO_2 emission rate value.

The formula of Li *et al.* (2019) was used for calculating cumulative CO_2 emissions (*CE*):

$$CE = \sum_{i=1}^{n} \left[\frac{(F_{i+1} + F_i)}{2} \times (t_{i+1} - t_i) \times 60 \times 60 \times 24 \times 10^{-6} \times 44 \right] \times 10$$

In the formula, *CE* is the cumulative of soil CO_2 emissions ($kg\cdot hm^{-2}$); (F_{i+1}+F_I) is the sum of soil CO_2 emission rate between two consecutive measurements ($\mu mol\cdot m^{-2}\cdot s^{-1}$); ($t_{i+1}$ -t_i) is the time interval between two consecutive measurements (d); and n is the number of measurements in total.

Accumulated carbon emissions from soil respiration (*CCE*): $CCE=CE\times 0.27$

In the formula, *CCE* is accumulated carbon emissions from soil respiration (kg/hm^{-2}); *CE* is the cumulative of soil CO_2 emissions ($kg\cdot hm^{-2}$); and 0.27 is the proportion of C to CO_2 molecular weight.

Carbon balance (*NEPC*) (Karelin *et al*. 2024)

$NEPC=NPPC-RmC$; $NPPC=(NPPa+NPPr)\times 0.45$; $NPPr=NPPa/2.1$; $RmC=CCE\times 0.865$; $Cs=NPPC/CCE$

NEPC is the carbon balance of ecosystem ($kg\cdot hm^{-2}$), *NPPC* is the carbon sequestration of net primary productivity ($kg\cdot hm^{-2}$), *RmC* is the carbon release from heterotrophic respiration of soil microorganisms ($kg\cdot hm^{-2}$), *NPPa* is the aboveground biomass ($kg\cdot hm^{-2}$), and *NPPr* is the root biomass ($kg\cdot hm^{-2}$). The carbon content in the aboveground and root parts of crops was 0.45. The ratio of aboveground biomass to root biomass was 2.1. The conversion coefficient of soil heterotrophic respiration was 0.865. *Cs* is the carbon sequestration potential of soil ecosystem.

In 2024, when the soil CO_2 emission rate was measured in the maize seedling stage, jointing stage, silk emergence stage, mid-filling stage, and maturity stage, soil samples from the 0-20 cm soil layer near the PVC base for each treatment were collected at the same time. These samples were brought immediately to the laboratory. After removal of the gravel, plant residues. and other unwanted materials, the samples were sieved with a 2 mm sieve. Thereafter, the samples were analyzed for the activities of different soil enzymes including urease, protease, sucrose, and peroxidase. The activities of soil urease, peroxidase, sucrase, and protease were measured according to the titration method described by Guan (1986).

All statistical analysis was performed with SPSS 17.0 software (SPSS Inc., Chicago, IL, USA). The Analysis of Variance was conducted using an ANOVA procedure, and the significant differences for all statistical tests were calculated at the level $P= 0.05$. Sigma Plot 12.0 (Aspire Software International, Ashburn, VA) was used for drawing map.

Results and Discussion

The change in soil CO_2 emission rates in each treatment exhibited a similar pattern, characterized by an initial increase followed by an overall decrease (Fig. 1). In 2023 and 2024, the soil CO_2 emission rates of M, MS, and S peaked in late August to early September, measuring 7.65, 6.91, and 7.11 $\mu mol\cdot m^{-2}\cdot s^{-1}$ and 7.69, 6.94, and 7.09 $\mu mol\cdot m^{-2}\cdot s^{-1}$, respectively. Subsequently, the soil CO_2 emission rates decreased gradually. As a result of the impact of rainfall, the soil CO_2 emission rate exhibited an upward trend, peaking again on September 17 (2023) and September 21 (2024).

Under different planting methods, the average cumulative soil CO_2 emissions for 2 years was M>S>MS (Fig. 2). The difference between M and MS treatments statistical significance was at $P<0.05$ level. M treatment recorded the highest yield, which reached 21,231 kg/hm^{-2}. Compared with S and MS treatments, M treatment resulted in increases of 7.69 (19,715 kg/hm^{-2}) and 22.57% (17,321 kg/hm^{-2}), respectively.

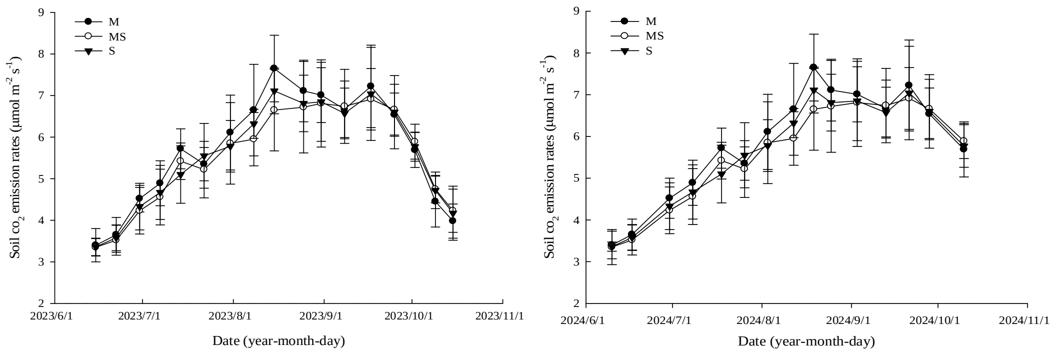

Fig. 1. Dynamic changes in soil CO_2 emission rate under different planting methods.

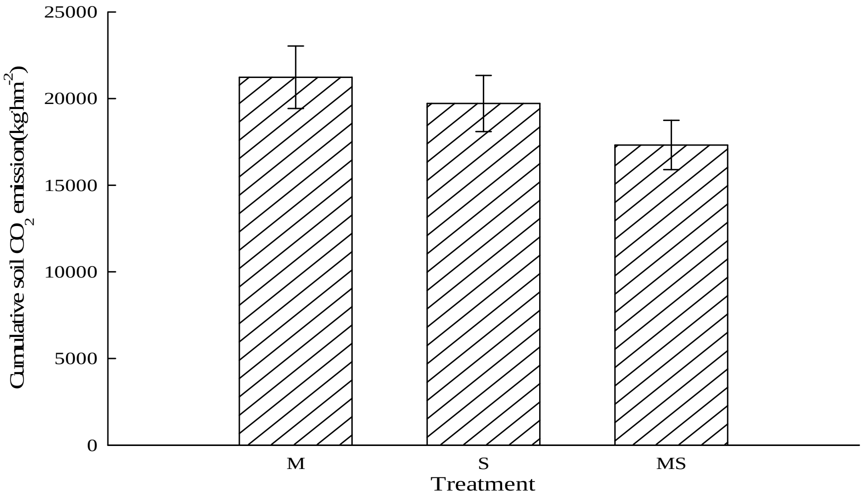

Fig. 2. Cumulative CO_2 emissions under different planting methods.

The relationship between 5 cm soil temperature and soil CO_2 emission rate was fitted using an exponential function (Fig. 3). The fitting equation between soil temperature and soil CO_2 emission rate reached a significant level ($P<0.05$). Under different treatments, soil temperature could explain the changes in soil CO_2 emission rate, which reached 48.67-60.65%. Among the different treatments tested, M treatment had the highest *R-squared* of 60.65%.

The relationship between 5-cm soil moisture and soil CO_2 emission rate was fitted using a non-linear function (Fig. 4). Under different treatments, soil moisture content could explain the changes in soil CO_2 emission rate, which reached 21.98-33.35%; these values were lower than those of soil temperature. S treatment had the highest *R-squared* of 33.35%. MS treatment had the lowest *R-squared* of 21.98%. When the content of soil moisture was low, the soil CO_2 emission rate increased with the increase in soil moisture content. When the soil moisture content exceeded a specific threshold, the soil CO_2 emission rate showed a decreasing trend.

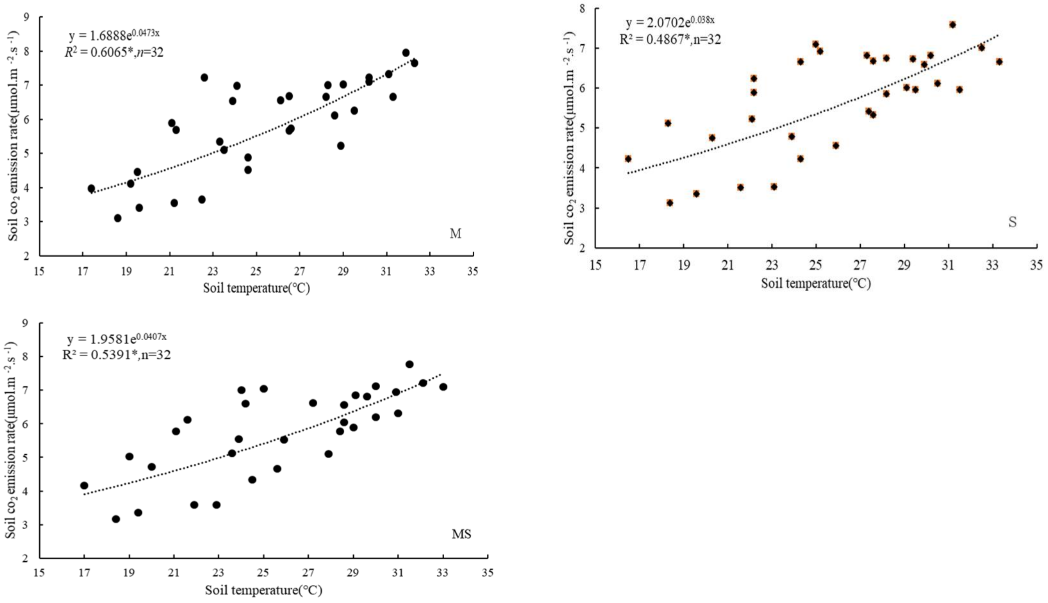

Fig. 3. Fitting model of soil temperature and soil CO_2 emission rate under different planting methods.

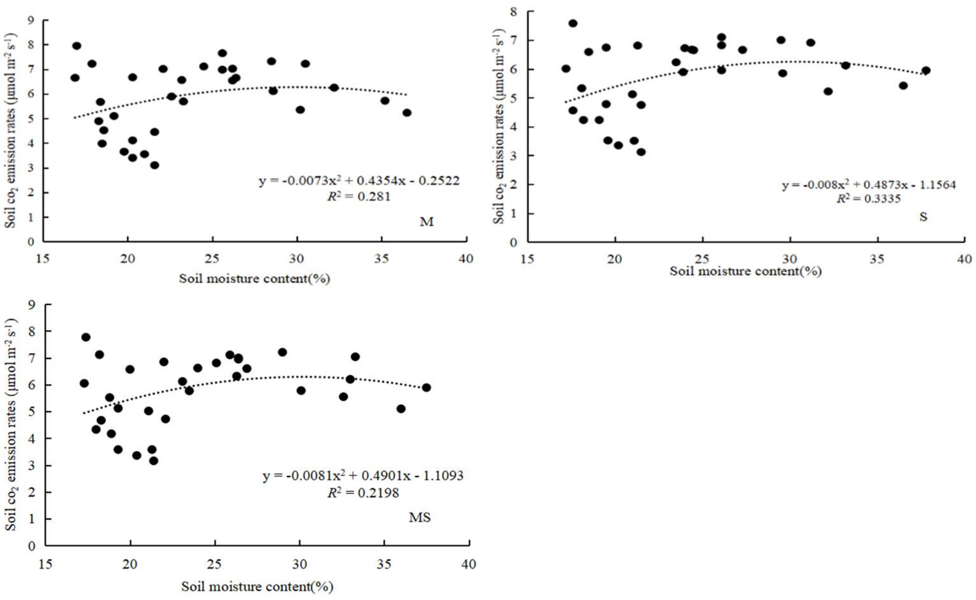

Fig. 4. Fitting model of soil moisture content and soil CO_2 emission rate under different planting methods.

A two-factor composite model was established, where soil moisture content (*W*) and soil temperature (*T*) were independent variables and soil CO_2 emission rate (Y) was dependent variable ($Y=f+aW+bT+cW^2+dT^2$, $P<0.05$) (Table 2). *R-squared* of the two-factor composite models for each treatment ranged from 0.5836 to 0.6854. Compared with the single-factor model, the two factor composite model demonstrated a better fit, which indicated that the two-factor composite model of soil moisture content and soil temperature could explain the changes in soil CO_2 emission rate well.

Table 2. Fitting parameters of water heat dual-factor composite model under different planting methods.

Treatment	*f*	*a*	*b*	*c*	*d*	R^2	*F*	*P*
M	−12.4447	0.6337	0.5654	−0.0076	−0.0105	0.6854	14.7027	<0.0001
S	−9.9382	0.4653	0.5653	−0.0054	−0.0103	0.5836	9.4592	<0.0001
MS	−8.4893	0.3780	0.5278	−0.0033	−0.0098	0.6177	10.9041	<0.0001

The fitting equation is $Y=f+aW+bT+cW^2+dT^2$; *Y*: Soil CO_2 emission rate; *W*: Soil moisture content; *T*: Soil temperature. a, b, c, and d are fitting coefficients.

The trends in soil protease activity were similar among different treatments. The maximum protease activity occurred in the maize seedling stage, whereas the minimum occurred in the mature stage. The trends in soil peroxidase and urease activity were consistent across different treatments. The maximum peroxidase and urease activity was observed at the maize jointing stage, whereas the minimum activity was recorded at the maturity stage (Fig. 5).

Correlation analysis showed a positive correlation between soil CO_2 emission rate and soil enzyme activity (Table 3). A significant correlation was found between soil CO_2 emission rate and soil urease activity ($P<0.05$), and a highly significant correlation was observed between soil CO_2 emission rate and peroxidase activity ($P<0.01$), indicating a close relationship between soil CO_2 emission rate and soil enzyme activity.

Table 3. Correlation analysis between soil CO_2 emission rate and soil enzyme activity.

Correlation analysis	Protease	Urease	Peroxidase	Sucrase
Soil CO_2 emission rate	0.45^{NS}	0.71*	0.84**	0.51^{NS}

NS= non-significant, * and ** indicate significance at 5 and 1%, respectively.

The carbon sequestration of net primary productivity (*NPPC*) in M, S, and MS treatments was 7,610, 5,181, and 6,088 kg/hm^{-2}, respectively (Table 4). M treatment was higher than S and MS treatments, which reached 47 and 25%, respectively. Significant differences were observed in carbon release from heterotrophic respiration (*R*m*C*) under different treatments ($P<0.05$), with the ranking of carbon release being M>S>MS. The carbon balance (*NEPC*) for each treatment was positive, which indicated that all treatments acted as absorption sinks for atmospheric CO_2. Compared with S and MS, the carbon balance values of M treatment increased by 359.31 and 29.80%, respectively.

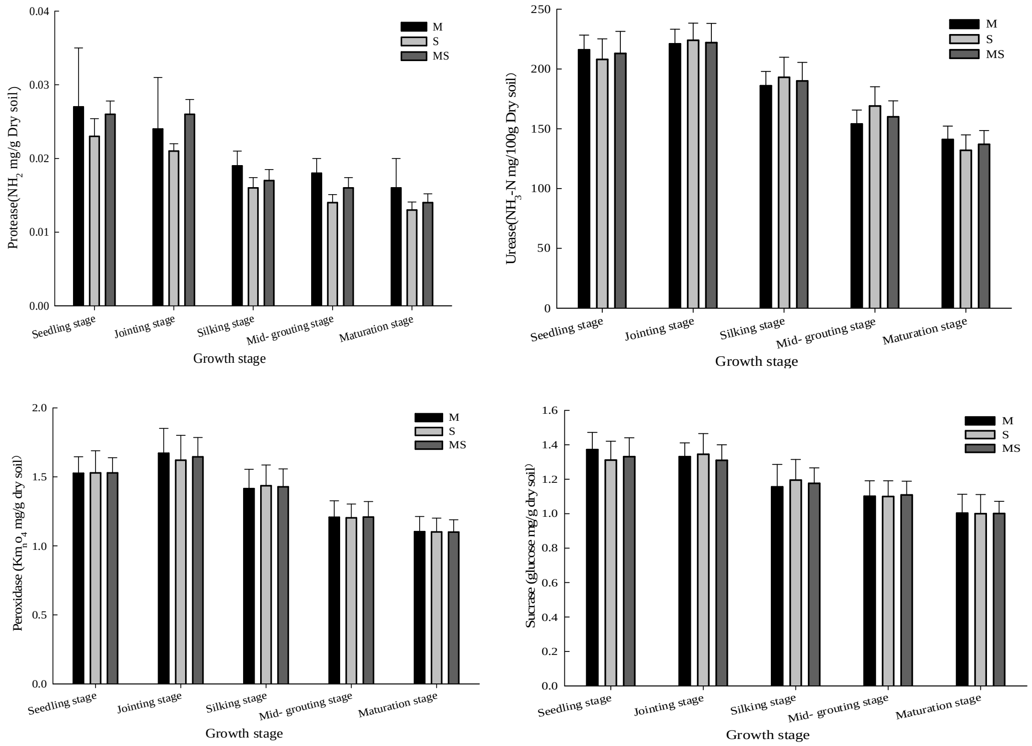

Fig. 5. Dynamic changes of soil enzyme activity under different planting methods.

Table 4. Carbon balance of crop growth season under different planting methods.

Treatment	NPPa kg/hm^{-2}	NPPr kg/hm^{-2}	NPPC kg/hm^{-2}	CCE kg/hm^{-2}	RmC kg/hm^{-2}	NEPC kg/hm^{-2}	Cs
M	11456.47± 120.56 a	5455.46± 57.41 a	7610.37± 80.09 a	5732.40± 486.01 a	4958.53± 420.40 a	2651.84± 340.31 a	1.33± 0.16 a
S	7800.67± 89.89 c	3714.60± 42.80 c	5181.87± 59.71 c	5323.15± 434.87 a	4604.52± 376.16 b	577.35± 31.45 c	0.97± 0.14 b
MS	9165.92± 110.05 b	4364.58± 52.40 b	6088.59 ± 73.10 b	4676.93± 367.67 b	4045.54± 318.03 c	2043.05± 244.93 b	1.30± 0.20 a

Different lowercase letters following data in the same column indicate significant differences between treatments ($P<0.05$).

In this study, soil respiration rate was significantly correlated with soil temperature ($P<0.05$, Fig. 3), which indicated that the seasonal variations in soil respiration rate were mainly caused by changes in soil temperature during this experiment. The optimal temperature promotes crop root growth. The activity of microbial and extracellular enzymes increased, accelerating the consumption and decomposition of soil carbon substrates and leading to an increase in soil respiration rate. However, during the late periods of crop growth, the physiological metabolic reactivity of soil microorganisms and roots weakened gradually. Therefore, the soil respiration rate also decreased gradually (Liang *et al.* 2021).

The effect of soil moisture on soil CO_2 emissions is complex. Xu and Qi (2001) found that a soil moisture content of around 20% is a critical value. Beyond this threshold, the impact of soil moisture on soil CO_2 emissions shifts from a positive correlation to a negative correlation, which was consistent with the results of this study. By contrast, some studies have found that the correlation between soil CO_2 emissions and soil moisture content is not significant (Dong *et al.* 2017). The minimal variation in soil moisture likely diminished its effect on soil CO_2 emissions. When soil temperature or soil moisture is at extreme levels, another factor may emerge as the primary influence on soil CO_2 emissions. Therefore, the single-factor model ignores the interdependence among various factors, rendering it inadequate for accurately depicting variations in soil CO_2 emission rates. The study demonstrated a significant correlation involving soil temperature, soil moisture, and soil CO_2 emissions (Guan *et al.* 2021). This study found that the dual-factor composite model of soil moisture and soil temperature could effectively elucidate variations in soil CO_2 emission rate compared with the single-factor model.

The rate of soil CO_2 emissions is regulated not only by soil temperature and moisture but also by soil enzyme activity (Li *et al.* 2019). This study found a significant association between peroxidase activity and soil CO_2 emission rate ($P<0.01$). In the biological process of soil respiration, the demand for hydrogen peroxide surpasses that of other soil enzymes. The presence of peroxide in soil and organisms may mitigate hazardous effects during the metabolic processes of organisms in soil (Wang *et al.* 2024). In addition, peroxide activity is associated with soil microbial activity, which can reflect the intensity of such activity. The activity of peroxide is also related to soil physical and chemical properties, as well as the microbial population, thereby serving as an indicator of soil respiration intensity.

Research shows that intercropping can reduce CO_2 emissions. CO_2 emissions under sugarcane-soybean intercropping decreased by 35.58% compared with those under sugarcane monoculture (Zhang *et al.* 2013). The CO_2 emissions under wheat-*Isatis* intercropping were reduced by 29.3% compared with those under wheat monoculture (Wu *et al.* 2017). Compared with maize monoculture, CO_2 emissions under wheat-maize and pea-maize intercropping were reduced by 32.0 and 38.0%, respectively (Qin *et al.* 2013). Studies have also indicated that intercropping does not mitigate soil CO_2 emissions. In sugarcane–soybean intercropping, the CO_2 emissions increased significantly compared with those in sugarcane monoculture (Guan *et al.* 2016). Gui *et al* (2024) found that intercropping exerts a minimal effect on carbon sequestration in agricultural ecosystems, which may even reduce carbon sequestration. The above research conclusions were inconsistent with the results of the present study. In this study, the cumulative soil CO_2 emissions under M treatment was significantly higher than those under MS treatment ($P<0.01$). The dual-factor composite model of soil temperature and moisture effectively elucidated the changes in soil CO_2 emission rate, accounting for 58.36%–68.54%. The activity of soil peroxidase significantly influenced the soil CO_2 emission rate (R-squared = 0.84, $P<0.01$). M treatment had a high ecosystem carbon balance value of 2651.84 $kg \cdot hm^{-2}$. M treatment showed a strong carbon sink function and soil carbon sequestration potential (1.33). The *NEPC* value with M treatment was significantly higher than that of S and MS treatments ($P<0.05$). M treatment exhibited high *NPPa* and *NPPr*, which enhanced its carbon sequestration of net primary productivity.

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