

*Original Research*

# The Effect of Organic Compost Dosage on Changes in Soil Dissolved Organic Matter Components Analyzed with Three-dimensional Fluorescence Spectroscopy and Ultraviolet Spectroscopy

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## Abstract

The application of organic compost in farmland is a major agronomic measure to increase soil organic matter content, enhance soil nutrients, and regulate soil microbial activity and other soil micro components. The influence of organic compost dosage on soil DOM components was studied. In this study, greenhouse pot experiments were conducted with 7 cattle manure organic compost dosage ratios of 0%, 5%, 10%, 20%, 40%, 60%, and 80%. Three-dimensional fluorescence spectroscopy with fluorescence regional integrated volume integration (3DEEM-FRI) and ultraviolet spectroscopy (UV) were used to analyze the effect of organic compost dosage on soil DOM components and structural characteristics. The results indicated that with the increase in organic compost dosage, the soil TOM content showed a linear increase, and the soil DOM content showed an exponential increase. The proportion of humic acid-like components in soil DOM showed a linearly increasing trend, and the proportion of tryptophan protein and fulvic acid components showed a linearly decreasing trend. The proportion of tyrosine protein and microbial metabolites, as well as lignin and quinone in DOM components, showed Exponential (Exp3P2) changes which first presented an increasing and then a decreasing trend, and reached a turning point at 20 g·kg<sup>-1</sup> of TOM. With the increase in the soil TOM content, the substituents on the benzene ring of soil DOM transformed from non-polar functional groups, such as fatty chains and esters, to polar functional groups such as hydroxyl, carboxyl, and carbonyl. The molecular condensation degree, molecular weight, and humification degree of the soil DOM showed a linearly increasing trend. The content of the hydrophobic organic component and the aromaticity of the soil DOM showed a linearly decreasing trend, and the proportion of biogenic sources showed an exponentially decreasing trend. The proportion of non-humified and humified substances in the soil

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DOM showed an initial increase and then a decreasing trend and reached the highest value at 35 g·kg<sup>-1</sup> of TOM. The organic compost dosage affected the proportion and molecular structure of the soil DOM components by regulating the biological effects of microorganisms and enzyme activity, as well as the conversion process from soil TOM to DOM.

**Keywords:** dissolved organic matter (DOM), soil, three-dimensional fluorescence spectra (3DEEM), organic compost

## Introduction

The use and research of organic compost have a long history, and extensive practice has shown that the application of organic compost in farmland can increase soil organic matter content, improve soil properties, promote the formation of soil aggregates, increase soil nutrients, and have a regulatory effect on soil microecology. Xia et al. [1] found that the application of cattle manure organic compost can increase the contents of soil total organic matter (TOM) and soil DOM and increase soil microbial biomass. DOM is an important active component of organic matter in soil ecosystems, playing a crucial role in a range of soil biochemical processes [2] and significantly impacting the cycling of soil microecological systems. The soil microecological carbon cycle is an important link in global carbon and nutrient cycles. Therefore, terrestrial DOM in farmland soil is crucial for global carbon and nutrient cycles, climate change, and human health [3]. The DOM structure contains oxygen-containing and aromatic functional groups, which can promote interactions between DOM and inorganic and organic matter, thereby significantly altering the biochemical and geochemical behavior of inorganic and organic matter in soil systems [4]. Therefore, the study of DOM in farmland soil has become a hot topic.

At present, to elucidate the structures and properties of the soil DOM at the microecological and molecular levels and further understand the ecological and environmental effects and transformation mechanisms of DOM, many techniques and methods were used to study DOM both domestically and internationally, including infrared spectroscopy (IR), nuclear magnetic resonance (NMR), mass spectrometry (MS), 3DEEM, and UV, etc. Compared with other methods, 3DEEM technology is very sensitive, easy to use, and does not damage the sample structure, and can quantitatively characterize the degree of humification of organic matter [5, 6]. The 3DEEM-FRI and UV were used to analyze the effect of organic compost dosage on soil DOM components and structural characteristics. The FRI can quantitatively analyze 3DEEM, determine DOM configuration and heterogeneity [7], and quantitatively distinguish subtle changes in 3DEEM to improve spectral analysis capabilities. 3DEEM can better reveal the impact mechanism of agricultural measures, such as fertilization, on soil microecology and provide micro evidence for the impact of human activities, such as

fertilization, on nutrient transformation mechanisms in farmland [6]. UV spectroscopic analysis, as an economical and rapid method, can provide molecular structure information of the soil DOM based on the specific spectra [8] and obtain a wealth of important information about DOM composition and transformation [9]. The combination of 3DEEM and UV is widely used to study the composition, sources, and biochemical cycles of DOM in soil, lake, and river sediments [10]. Therefore, the combined use of 3DEEM and UV to determine the spectral characteristics of soil DOM can more accurately reveal DOM composition and molecular structure changes, as well as the response mechanism to soil microorganisms and physicochemical properties, and provide microscopic evidence for the application of organic fertilizers to improve soil.

## Materials and Methods

### Experimental Treatment

The experiment was simulated indoors in a solar greenhouse, with the temperature maintained between 25°C and 35°C and the humidity maintained between 50% and 70%. The tested soil is sandy loam soil from Bashang in Zhangbei County, Zhangjiakou City, Hebei Province, and the physical and chemical properties of the tested soil are shown in Table 1. Organic compost (organic matter 14.70%, nitrogen (N) 0.42 g·kg<sup>-1</sup>, C:N 20:1, NH<sub>4</sub><sup>+</sup>-N 79.35 mg·kg<sup>-1</sup>, NO<sub>3</sub><sup>-</sup>-N 223.08 mg·kg<sup>-1</sup>, and phosphorus (P<sub>2</sub>O<sub>5</sub>) 0.22 g·kg<sup>-1</sup>) is made from cattle manure, which has been composted for 3 months. Seven treatments were designed based on the ratio of organic compost dosage to soil quality for this research: 0%, 5%, 10%, 20%, 40%, 60%, and 80%, respectively. The experiment selected a polyethylene pot with a diameter of 20 cm and a height of 25 cm as the culture pot. The organic compost and soil were mixed evenly and placed in a culture pot, and each treatment contained 4.5 kg of dry mixed soil and was repeated 3 times. Naked oats were planted, sown on April 20, 2023, harvested on September 10, 2023, and irrigated regularly. After harvesting, different treatment soils were collected, air dried, ground, sieved through a 100-mesh screen, and prepared for further analysis.

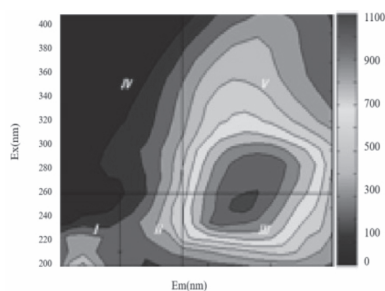
Table 1. Physical and chemical properties of the tested soil.

Soil	BD/(g·cm <sup>-3</sup> )	SMC	TN/(g·kg <sup>-1</sup> )	TP/(mg·kg <sup>-1</sup> )	TOM/(g·kg <sup>-1</sup> )	pH	NH <sub>4</sub> <sup>+</sup> -N/(mg·kg <sup>-1</sup> )	IP/(mg·kg <sup>-1</sup> )
Sandy loam	1.58	2.77%	0.86±0.05	314.33±3.67	14.56±0.90	7.40	11.26±4.20	17.11±0.83

### Soil Analysis

Soil analysis was conducted based on the methods in the literature [6]. DOM fluorescence spectroscopic analysis of soil: took a certain amount of soil, extracted (20°C, 200 r·min<sup>-1</sup>, 24 h) by 1 mol·L<sup>-1</sup> KCl in a ratio of 1:10 (m/V, g·mL<sup>-1</sup>), centrifuged (5000 r·min<sup>-1</sup>, 15 min), then filtered with a 0.45 μm glass fiber membrane; DOM solution was obtained and diluted to 10 times to reduce the internal filter effect, the diluent was determined DOM and dissolved organic carbon (DOC). The DOM three-dimensional fluorescence spectrum was determined by a HITACHI F-7000 fluorescence spectrometer (F-7000, Hitachi High-Technology Corp, Tokyo, Japan). The fluorescence spectrophotometer was equipped with a 150 W Xenon arc lamp (Hitachi High-Technology Corp, Tokyo, Japan) as the light source and two grating monochromators coupled with a slit as the EEM wavelength selectors, with quartz cuvettes of 1 cm; scanning speed was set to 60,000 nm/min, and each measurement period was 2 min in duration. The 3DEEM parameters were as follows: Bandpass: excitation wavelength,  $\lambda_{ex} = 5$  nm, emission wavelength,  $\lambda_{em} = 5$  nm; Scanning speed: 2400 nm·min<sup>-1</sup>. Three-dimensional fluorescence spectrometric determination: excitation wavelength (Ex) 200-440 nm, emission wavelength (Em) 250-600 nm, MilliQ water was used as 3DEEM blanks. The fluorescence spectra of

the soil DOM were quantitatively analyzed by FRI [11] to determine DOM structure and heterogeneity, quantitatively distinguish subtle changes in EEM, and improve spectral analysis capabilities. By calculating the percentage of the specific region standard volume ( $\varphi_{(i,n)}$ ) and whole region standard volume ( $\varphi_{(T,n)}$ ) ( $P_{(i,n)} = \varphi_{(i,n)} / \varphi_{(T,n)} \times 100\%$ ), the content and relative content of fluorescent substances with specific structures within the corresponding region were determined. The three-dimensional fluorescence component regions of the soil DOM are shown in Fig. 1 [11]. In general, peaks at shorter excitation wavelengths (<250 nm) and shorter emission wavelengths (<380 nm) are related to simple aromatic proteins, such as tyrosine and tryptophan-like compounds (Regions I and II). Peaks at shorter excitation wavelengths (<250 nm) and longer emission wavelengths (>380 nm) are related to fulvic acid-like substances (Region III). Peaks at intermediate excitation wavelengths (250-400 nm) and shorter emission wavelengths (<380 nm) are associated with dissolved microbial metabolites (Region IV). Peaks at longer excitation wavelengths (>250 nm) and longer emission wavelengths (>380 nm) represent humic acid-like organic fractions (Region V) [6, 12]. The description and characterization significance of the 3DEEM characteristic parameters of DOM are shown in Table 2.



Region	Ex/Em (nm)	DOM component
Region I	220-250/280-330	Tyrosine protein
Region II	220-250/330-380	Tryptophan protein
Region III	220-250/380-550	Fulvic acid class
Region IV	250-400/280-380	Dissolved microbial metabolites
Region V	250-400/380-550	Humic acid class

Fig. 1. Three-dimensional fluorescence component regions of soil DOM.

Table 2. Description and characterization significance of three-dimensional fluorescence spectral characteristic parameters.

Fluorescence spectral parameters	Significance	Definition	Reference
Fluorescence Index ( <i>FI</i> )	Characterizes the humus sources of DOM	At $\lambda_{ex} = 370$ nm, the ratio of fluorescence intensity at emission wavelengths of 450 nm and 500 nm.	[13-15]
Humification Index ( <i>HIX</i> )	Characterizes the humification degree of DOM	At $\lambda_{ex} = 254$ nm, the ratio of fluorescence intensity integration values between emission wavelengths of 435-480 nm and 300-345 nm.	[13-15]
Biological Index ( <i>BIX</i> )	Characterizes the strength of DOM endogenous features	At $\lambda_{ex} = 310$ nm, the ratio of fluorescence intensity at emission wavelengths of 380 nm and 430 nm.	[13-15]

Table 3. Description and significance of UV characteristic parameters of DOM.

Absorbance	Significance	Definition	Reference
SUVA <sub>254</sub>	Characterizes the degree of DOM humification	Absorption coefficient of unit DOC concentration at a wavelength of 254 nm.	[16]
SUVA <sub>260</sub>	Characterizes the hydrophobic component content of DOM	Absorption coefficient of unit DOC concentration at a wavelength of 260 nm.	[16]
SUVA <sub>280</sub>	Characterizes the aromaticity of DOM	Absorption coefficient of unit DOC concentration at a wavelength of 280 nm.	
$A_{253}/A_{203}$	Reflects the molecular structure of DOM and the types of benzene ring substituents, positively correlated with the complexity of substituents.	The ratio of absorbance at 253 nm and 203 nm in UV-visible spectroscopy.	[16]
$A_{250}/A_{365}$	Characterizes the aromaticity and molecular weight of DOM	The ratio of absorbance at 250 nm and 365 nm in UV-visible spectroscopy.	[16]
$A_{240}/A_{420}$	Characterizes the molecular condensation degree of DOM (inversely proportional)	The ratio of absorbance at 240 nm and 420 nm in UV-visible spectroscopy.	
$S_R$	Characterizes the source composition and structural changes of DOM	The slope ratio of absorbance at 275-295 nm and 350-400 nm in the UV-visible spectrum.	[16]
$A_2/A_1$	Reflects the proportion of organic matter, such as lignin and quinone, in the early stage of humification	The ratio of integrated areas in the 460-480 nm and 260-280 nm wavelength bands.	[17]
$A_3/A_1$	Represents the ratio of non-humified and humified substances	The ratio of integrated areas in the 600-700 nm and 260-280 nm wavelength bands.	[17]
$A_3/A_2$	Indicates the aromaticity of organic matter	The ratio of integrated areas in the 600-700 nm and 460-480 nm wavelength bands.	[17]

The UV absorption spectra of the soil DOM were scanned using a dual-beam ultraviolet/visible spectrometer (TU-1900, Beijing Purkinje General Instrument Co., Ltd, Beijing, China). The sample processing method for UV measurement: take 2 g of the soil sample, mix the soil sample with distilled water in a ratio of 1:10, shake and extract continuously for 12 h, then centrifuge for 10 min at 4000 r·min<sup>-1</sup>, pass through a 0.45 μm filter membrane, and take the supernatant to measure DOM. The scanning wavelength range is from 190 nm to 1002 nm with a scanning wavelength interval of 2 nm. The UV spectral parameters were calculated according to the methods described in the literature [16, 17]. The description and significance of UV characteristic parameters of DOM are shown in Table 3.

DOC was measured using a TOC-5000A (Shimadzu, Japan). Microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN) were measured using the methods of Calbrix. MBC was determined with a multi N/C 3100 analyzer (Analytik Jena AG, Jena, Germany). MBN was analyzed with a FOSS Kjeldahl nitrogen analyzer (Model Kjeltac 2300, FOSS Analytical AB, Hoganas, Sweden) [18]. A dual-beam ultraviolet/visible spectrometer (TU-1900, Beijing Purkinje General Instrument Co., Ltd, Beijing, China), automatically controlled by computer, was used to measure soil sucrose by the 3,5-dinitrosalicylic acid colorimetric method, soil phosphatase by the colorimetric method of benzene disodium phosphate, and soil urease by indigophenol colorimetry [6]. Soil catalase was determined by potassium permanganate titration [6].

Soil physical and chemical properties were determined by conventional analysis [19].

### Data Analysis

Significance analysis of data differences is carried out using Excel 2007 and SPSS 17.0 software. The relationship model between soil organic matter and DOM components is performed with Origin 9 software. The fitting models include three types, namely, the linear model ( $Y = aX + b$ ), the exponential model ( $Y = ae^{bX}$ ) and the triple exponential smoothing curve model (Exp3P2,  $Y = a + bX + cX^2$ ), where  $Y$  is the spectral component parameter of soil DOM,  $X$  is the content of soil TOM, and  $a$ ,  $b$ , and  $c$  are model parameters. The three-dimensional fluorescence spectroscopy is processed by MATLAB 2007 software.

## Results

### The Effect of Organic Compost Dosage on the soil DOC content

Organic compost is the main source of soil TOM and an important source of DOM. The effect of organic compost dosage on TOM and DOC content is shown in Fig. 2. With the increase in organic compost dosage, the soil TOM content showed a linear increasing trend, and the soil DOC content showed an exponential increasing trend. The DOC content can reflect the soil

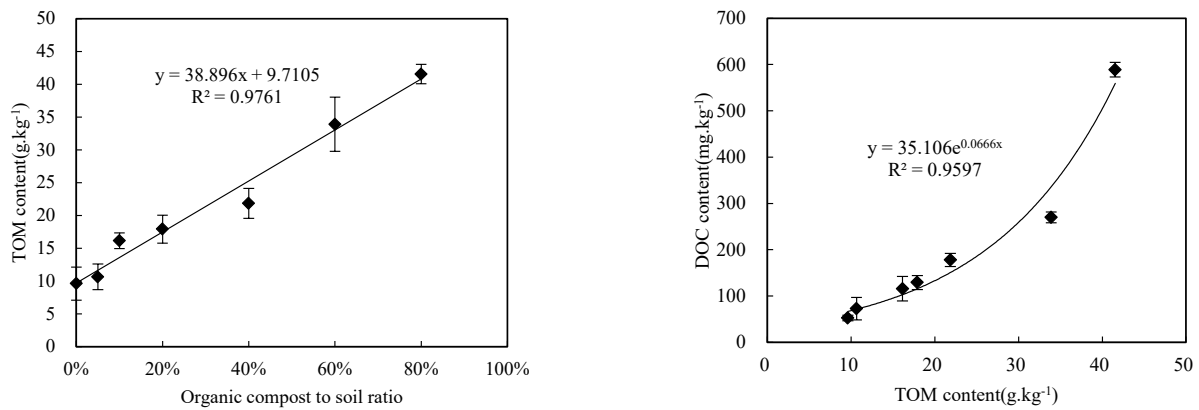


Fig. 2. Relationship between organic compost dosage and the content of soil TOM and DOC.

DOM content. The results showed that when the organic compost dosage was smaller, the soil DOM content increased slowly with the increase in dosage. When the organic compost dosage was larger, the soil DOM content increased exponentially with the increase in dosage.

#### The Effect of Organic Compost Dosage on the Three-dimensional Fluorescence Spectral Characteristics of Soil DOM

The effect of organic compost dosage on the 3DEEM components of soil DOM is shown in Fig. 3. With the increase in soil TOM content, the soil DOM in Region I and Region IV showed Exponential (Exp3P2) changes ( $R^2 = 0.4834$ ,  $R^2 = 0.7548$ ), which presented an initial increase followed by a decrease. The soil DOM in Region II and Region III showed a linear decreasing trend. The soil DOM in Region V showed a linear increasing trend. Therefore, the proportion of tyrosine protein and microbial metabolites in soil DOM showed an initial increase and then a decreasing trend. The proportions of tryptophan protein and fulvic acid showed a downward trend, and the proportion of humic acid showed an increasing trend. The results indicated that with the increase in organic compost dosage, the soil TOM content increased, and TOM was transformed into DOM. At the same time, the proportion of small-molecule DOM components was reduced by enhanced physical and chemical processes such as microbial degradation and TOM adsorption.

With increasing organic compost dosage, the changes in the soil DOM three-dimensional fluorescence parameters are shown in Fig. 4. Research has shown that  $FI > 1.9$  indicates that DOM mainly comes from microbial metabolites in the soil, while  $FI < 1.4$  indicates that DOM mainly comes from external inputs [13-15]. With the increase in BIX, the endogenous characteristics of DOM are enhanced, and the terrestrial characteristics are reduced [13-15].  $HIX < 4$  indicates a weak degree of DOM humification, while when HIX reaches 10-16, it indicates that DOM has significant

humus characteristics [13-15]. With the increase in the soil TOM content, the FI and BIX of soil DOM show a trend of first rising and then falling, and the HIX of soil DOM shows an upward trend. According to the fluorescence spectral parameters, when the soil TOM content is low ( $< 20 \text{ g.kg}^{-1}$ ), with the increase in the soil TOM content, the source of soil DOM shifts from exogenous to biogenic sources, and the biogenic source gradually increases. When the soil TOM content is high ( $> 20 \text{ g.kg}^{-1}$ ), with the increase in the soil TOM content, the source of soil DOM shifts from biogenic sources to exogenous sources, and the terrigenous source gradually increases, and the humification degree of soil DOM shows an increasing trend.

#### The Effect of Organic Compost Dosage on the UV Spectral Characteristics of Soil DOM

The effect of organic compost dosage on the UV spectral parameters ( $A_{253}/A_{203}$ ,  $A_{240}/A_{420}$ , and  $A_{250}/A_{365}$ ) of soil DOM is shown in Fig. 5. When the  $A_{253}/A_{203}$  ratio of DOM was low, the substituents on the aromatic ring structure were mainly non-polar functional groups, such as fatty chain photosynthetic esters. When the  $A_{253}/A_{203}$  ratio was high, the types of substituents on the benzene ring structure were mainly polar functional groups such as hydroxyl, carboxyl, and carbonyl [16]. The  $A_{240}/A_{420}$  was inversely proportional to the condensation degree of DOM molecules [16]. When  $A_{250}/A_{365} < 3.5$ , the content of humic acid in DOM was greater than that of fulvic acid, and organic matter with relatively high molecular weight was predominant; When  $A_{250}/A_{365} > 3.5$ , the content of humic acid in DOM was lower than that of fulvic acid, and organic matter with relatively small molecular weight was mainly present [16]. With the increase in soil TOM content, the  $A_{253}/A_{203}$  of soil DOM showed an exponential increase, and the  $A_{240}/A_{420}$  and  $A_{250}/A_{365}$  showed a linear downward trend. This indicates that the substituents on the benzene ring of soil DOM were transformed from nonpolar functional groups, such as fatty chains and esters, to polar functional groups such as hydroxyl, carboxyl,

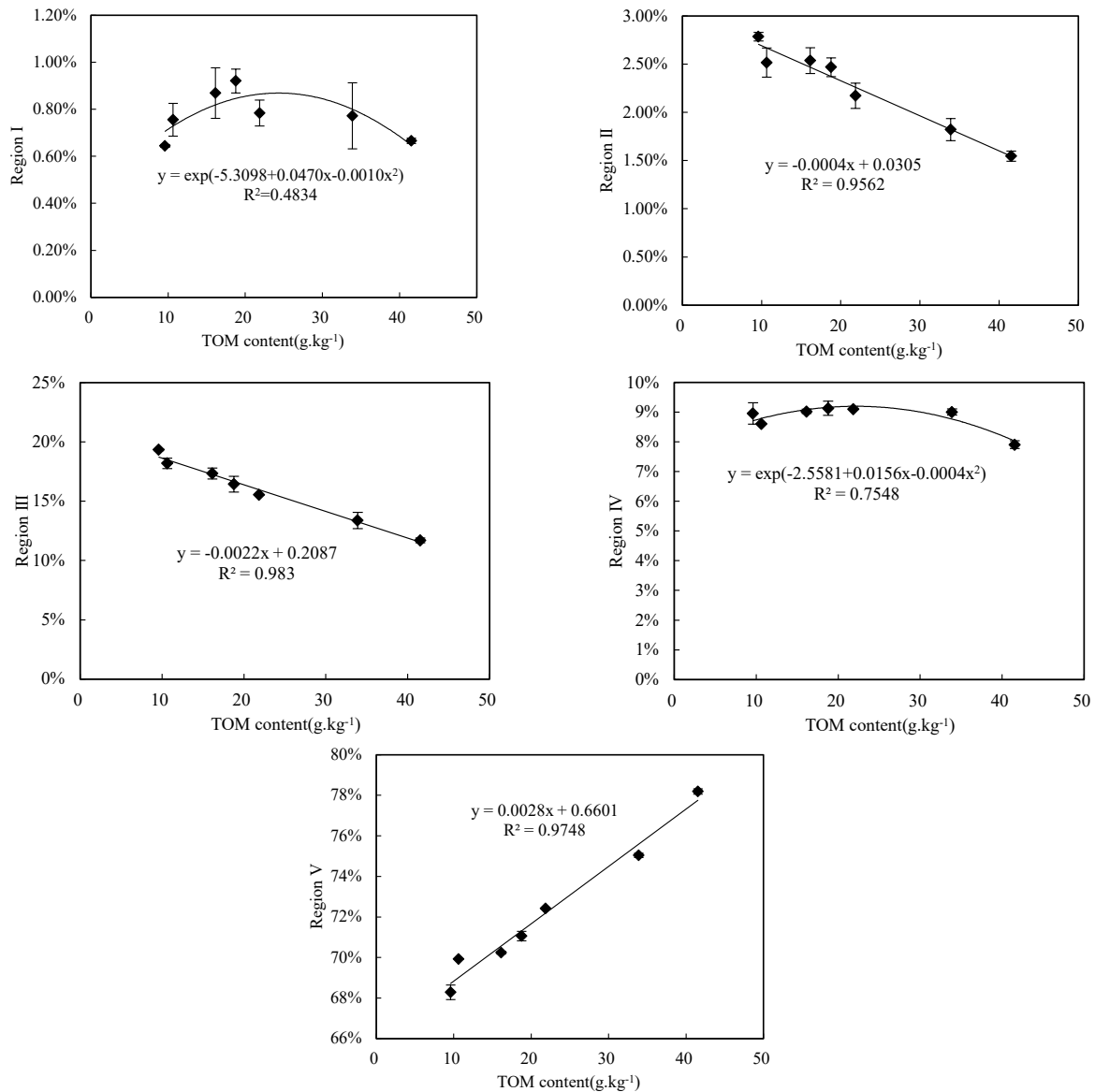


Fig. 3. Relationship between organic compost dosage and the three-dimensional fluorescence spectrum components of soil DOM.

and carbonyl groups. The molecular condensation degree of soil DOM was increased. The molecular weight of DOM increased. The content of humic acid was greater than that of fulvic acid, and the proportion of humic acid was increased.

The effects of organic compost dosage on the UV spectral parameters ( $A_3/A_1$ ,  $A_3/A_2$ , and  $A_2/A_1$ ) of soil DOM are shown in Fig. 6. As the soil TOM content increased, the  $A_2/A_1$  of soil DOM showed a first decreasing and then increasing Exponential (Exp3P2) change ( $R^2 = 0.6059$ ). The  $A_3/A_1$  and  $A_3/A_2$  of soil DOM showed the first increasing and then decreasing Exponential (Exp3P2) changes ( $R^2 = 0.7333$ ,  $R^2 = 0.7586$ ). It showed that the proportion of lignin and quinone in soil DOM first decreased and then increased, and reached a turning point at a TOM of 20 g.kg<sup>-1</sup>. The proportion of non-humified and humified substances and the aromaticity of soil DOM first increased and then decreased, and reached their highest value at a TOM of 35 g.kg<sup>-1</sup>.

The effect of organic compost dosage on the UV spectral parameters ( $SUVA$  and  $S_R$ ) of soil DOM is shown in Fig. 7. The  $SUVA_{254}$  of DOM is positively correlated with the relative molecular weight and humification degree of organic matter [16]. The  $SUVA_{260}$  was positively correlated with the content of hydrophobic organic components in DOM [16]. The  $SUVA_{280}$  is positively correlated with the aromaticity of DOM. When  $S_R < 1$ , the main source of DOM was an exogenous source, while when  $S_R > 1$ , the main source of DOM was a biogenic source [16]. With the increase in soil TOM content, the  $SUVA_{254}$ ,  $SUVA_{260}$ ,  $SUVA_{280}$ , and  $S_R$  of soil DOM showed an exponential downward trend. The UV spectral parameters showed that the relative molecular weight and humification degree of soil DOM, the content of hydrophobic organic components, and the proportion of aromaticity and biogenic sources of soil DOM all showed a decreasing trend.

### The Effect of Organic Compost Dosage on the Microorganisms and Enzyme Activity of Soil

The effect of organic compost dosage on the soil microbial biomass, such as MBC, MBN, and microbial biomass phosphorus (MBP), is shown in Fig. 8. With the increase in soil TOM content, the content of the soil MBC and MBN showed Exponential (Exp3P2) changes ( $R^2 = 0.4871$ ,  $R^2 = 4922$ ), which presented an initial increase followed by a decrease. The content of the soil MBP showed an increase in Exponential (Exp3P2) change ( $R^2 = 9942$ ). It showed that when the soil TOM content was low ( $<20 \text{ g}\cdot\text{kg}^{-1}$ ), the contents of the soil MBC and MBN showed an upward trend; when the soil TOM was high ( $>20 \text{ g}\cdot\text{kg}^{-1}$ ), the contents of the soil MBC and MBN showed a downward trend, and with the

increase in the soil TOM content, the content of the soil MBP showed an upward trend.

The effect of organic compost dosage on soil urease activity (URE) and soil sucrase activity (SUC) is shown in Fig. 9. With the increase in soil TOM content, the soil URE showed an increasing Exponential (Exp3P2) change ( $R^2 = 6176$ ) and reached the maximum value at TOM of  $20 \text{ g}\cdot\text{kg}^{-1}$  and then stabilized. The soil SUC showed an initially increasing and then decreasing Exponential (Exp3P2) change ( $R^2 = 0.8381$ ) and reached the maximum value at a TOM of  $20 \text{ g}\cdot\text{kg}^{-1}$ . It showed that with the increase in soil organic compost dosage, when the soil TOM content was low ( $<20 \text{ g}\cdot\text{kg}^{-1}$ ), the content of the soil URE and SUC showed the increasing trends, and when the soil TOM content

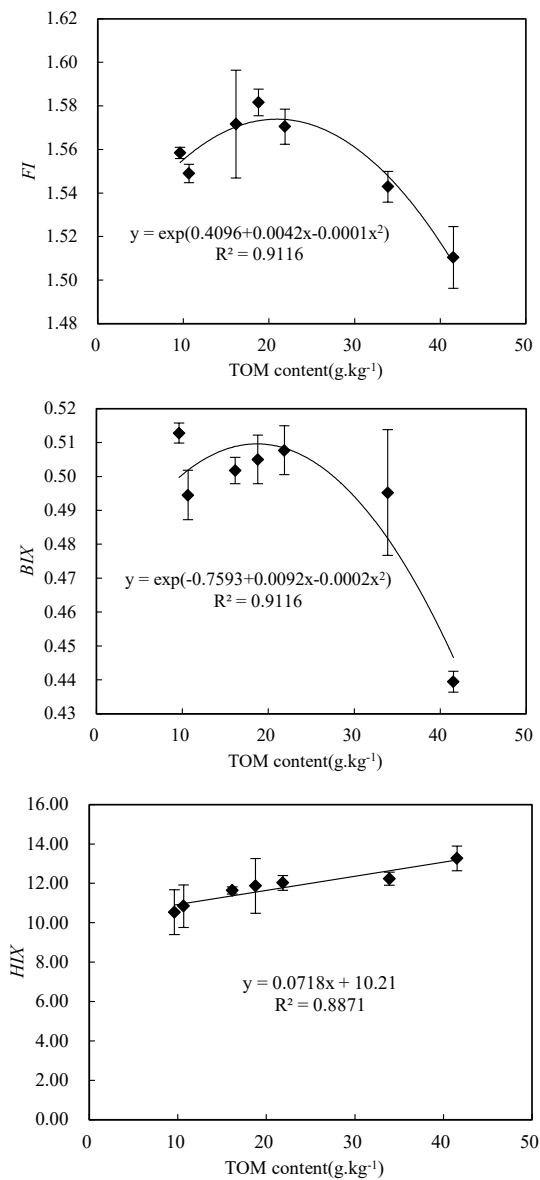


Fig. 4. Relationship between organic compost dosage and the three-dimensional fluorescence spectral parameters of soil DOM.

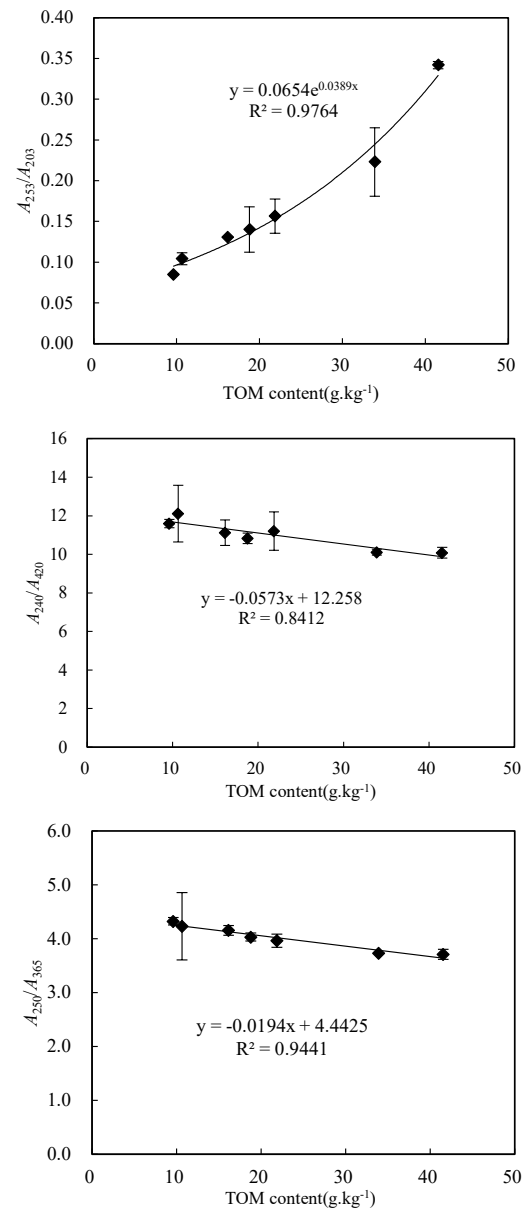


Fig. 5. Relationship between organic compost dosage and UV spectral parameters ( $A_{253}/A_{203}$ ,  $A_{240}/A_{420}$ , and  $A_{250}/A_{365}$ ) of soil DOM.

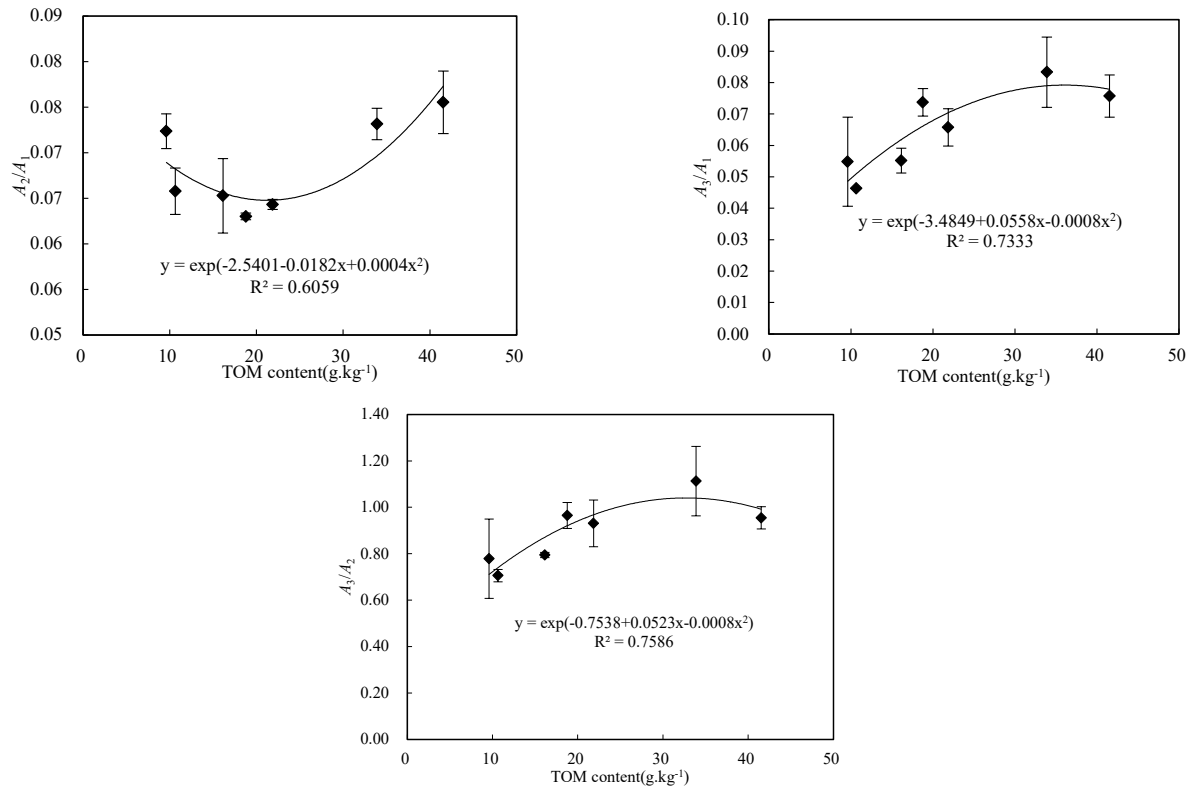


Fig. 6. Relationship between organic compost dosage and UV spectral parameters ( $A_3/A_1$ ,  $A_3/A_2$ , and  $A_2/A_1$ ) of soil DOM.

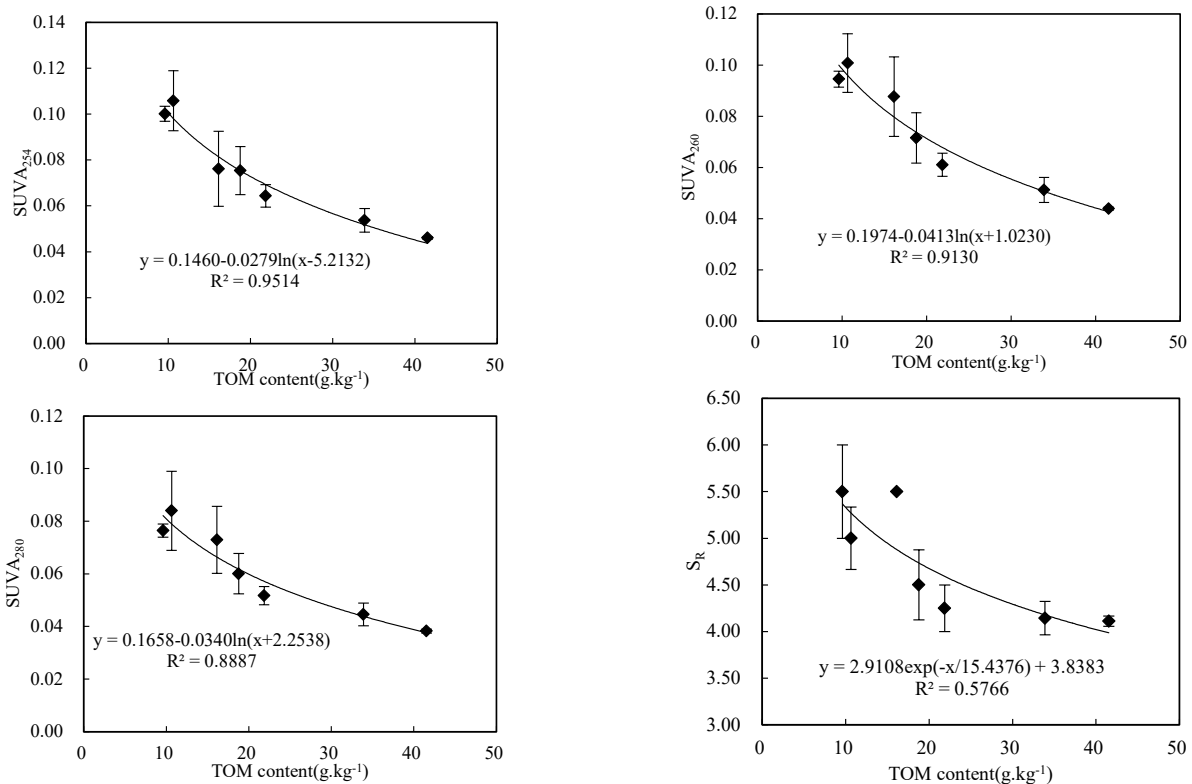


Fig. 7. Relationship between organic compost dosage and the UV spectral parameters ( $SUVA$  and  $S_r$ ) of soil DOM.

was high (>20 g·kg<sup>-1</sup>), the content of soil SUC showed a decreasing trend, and the content of soil URE tended to stabilize.

### Discussion

#### The Effect of Organic Compost Dosage on the Content of Soil DOM

Organic compost is an important source of soil organic matter in farmland and has a significant impact on the content and composition of soil TOM. Organic matter plays a key role in maintaining ecosystem health and function, and is an important source of nutrition

and energy for plant growth and soil organisms [20]. The experimental results showed that the content of soil TOM increased in a linear trend with the increase in organic compost dosage. Xia et al. [1] found that the application of cow manure organic fertilizer can increase the content of the soil organic carbon and water-soluble organic carbon, then increase the soil microbial biomass. With the increase in soil TOM content, the content of soil DOM showed an exponential increase. Liu et al. [21] found a positive correlation between TOM and DOM. When the application rate of organic fertilizer was at a low level, with the increase in organic fertilizer input, the content of the soil TOM and the soil microbial biomass increased (Fig. 8). The DOM converted from soil TOM was easily absorbed and utilized by soil microorganisms to convert into CO<sub>2</sub> [22]. The soil microbial residues were converted to DOM [20]. Therefore, under the dual action of microorganisms, the content of DOM increased slowly with the increase in organic fertilizer input. At this time, soil DOM was mainly accumulated through microbial synthesis. The proportion of highly active small-molecule DOM was relatively high. The proportion of soluble microbial metabolites in DOM was increased. When the input of organic fertilizer was at a high level, with the increase in organic fertilizer application, the content of soil TOM was increased, and a large amount of TOM was converted into DOM.

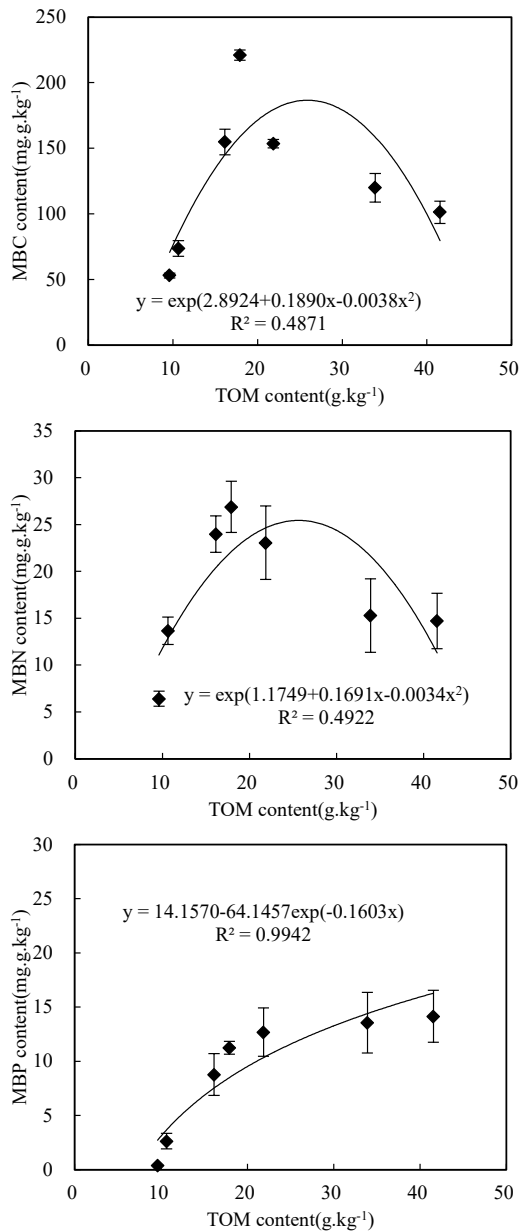


Fig. 8. Relationship between organic compost dosage and the contents of soil MBC, MBN, and MBP.

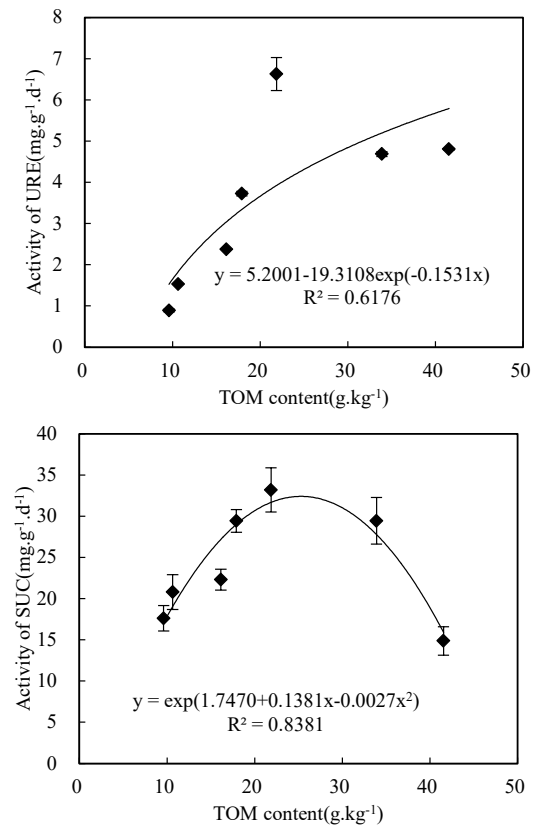


Fig. 9. Relationship between organic compost dosage and the content of soil urease activity (URE) and soil sucrase activity (SUC).

At the same time, the soil microbial biomass was decreased, and the degradation effect of soil microorganisms on DOM decreased (Fig. 8). Therefore, the content of the soil DOM showed a rapid increase trend with the increase in organic compost dosage. At this time, the proportion of humic acids with low activity in soil DOM was high, and the proportion of soluble microbial metabolites decreased. Microorganisms drove the cycle of DOM in soil from easily degradable to a difficult-to-degrade state [23]. It can be seen that increasing the application of organic compost has improved soil nutrient supply capacity, enhanced soil fertility, and increased crop yield.

### The Effect of Organic Compost Dosage on the Components of Soil DOM

The organic matter applied to soil by organic compost can be converted into the soil DOM, while large organic mineral aggregates can adsorb and encapsulate DOM, and reduce its activity. At the same time, soil organic matter can degrade DOM by increasing the activity of soil organisms and crop roots [20]. Studies have shown that microorganisms may be the main source of organic carbon, and that microbial necrotic clumps contribute up to 50-80% to SOC [20]. The experimental results showed that with the increase in organic compost dosage, the soil TOM content increased, the proportion of humic acid-like components in soil DOM showed an increasing trend, the proportion of tryptophan protein and fulvic acid components showed a decreasing trend, the content of humic acid with weaker activity was greater than that of fulvic acid with stronger activity and the proportion increased, the humification degree of soil DOM increased, and the hydrophobic organic component content of the soil DOM decreased. The experimental results also indicated that with the increase in organic compost dosage, soil microorganisms mainly degraded small molecules such as highly active proteins and fulvic acid in soil DOM, as well as hydrophobic DOM components. The correlation analysis (Table 4) showed that Region II (tryptophan protein) and Region III (fulvic acid class) were significantly negatively correlated with MBP and URE.

With the increase in organic compost dosage, the soil TOM content increased, and the proportion of tyrosine proteins and microbial metabolites in soil DOM components, and the fluorescence parameters of FI and BIX, all showed the trend of first increasing and then decreasing. The highest value was reached when the content of soil TOM was 20 g·kg<sup>-1</sup>. Pan et al. [24] found that after the application of organic fertilizer, the BIX value of soil DOM first increased and then decreased with the prolongation of time. When the soil TOM content was low (<20 g·kg<sup>-1</sup>), the content of the soil URE and SUC showed an upward trend, and when the soil TOM content was high (>20 g·kg<sup>-1</sup>), the content of the soil SUC showed a downward trend, and the content of the soil URE tended to stabilize. When

the TOM content in the soil was low (<20 g·kg<sup>-1</sup>), as the content of soil TOM increased, the source of soil DOM transformed from exogenous to biogenic sources. The proportion of DOM derived from microbial metabolic yield and protein-like components increased. The proportion of lignin and quinone in the soil DOM decreased, and the biogenic sources of the soil DOM gradually increased. The main reasons were the increase in the soil MBC and MBN content (Fig. 8) and the enhancement of the soil enzyme activity (Fig. 9) during this stage. The soil TOM was utilized and degraded by microorganisms into small-molecule DOM, leading to an increase in soil microbial-derived DOM. Correlation analysis showed that Region I (tyrosine protein) was highly significantly positively correlated with MBC, and Region IV (dissolved microbial metabolites) was significantly positively correlated with SUC. When the content of soil TOM was high (>20 g·kg<sup>-1</sup>), with the increase in soil TOM content, a large amount of organic matter was decomposed and produced thermochemical reactions [25, 26], and affected the metabolic activity of thermophilic microorganisms [27], decreased the biomass of soil microorganisms (Fig. 8), decreased the enzyme activity (Fig. 9), weakened the biological effect on the conversion from soil TOM to DOM. The DOM in soil mainly came from the DOM in exogenous organic fertilizers, indicating that the exogenous and terrestrial sources gradually increased, and the proportion of lignin and quinone in the soil DOM increased (Fig. 10). Therefore, excessive application of organic compost could inhibit the fertilizer efficiency of soil organic matter. In production, organic compost dosage should be applied in moderation to control soil TOM at around 20%.

### The Effect of Organic Compost Dosage on the Molecular Structure of Soil DOM

DOM came from organic compost, was a mixture of organic compounds with different crown energy groups and molecular weights [28], and contained chemical functional groups such as -SH, -NH<sub>2</sub>, -COOH, -OH, and benzene rings. The molecular weight ranged from several hundred daltons to tens of thousands of daltons, and the materials were mainly composed of organic acids, carboxylic acids, amino acids, carbohydrates, and humic acids [29]. A large number of studies have shown that important information about the chemical structure of DOM can be obtained by detailed analysis of UV-visible spectra [9], which provides a good indication of the microscopic molecular structure of DOM components [30]. With the increase in soil TOM content, the substituents on the benzene ring of soil DOM transformed from nonpolar functional groups, such as fatty chains and esters, to polar functional groups such as hydroxyl, carboxyl, and carbonyl groups. The degree of molecular condensation of the soil DOM increased, and the molecular weight of the soil DOM increased. The research results indicated that with the increase

in organic compost dosage, soil microorganisms mainly degraded non-polar crown energy groups of the soil DOM. According to the correlation analysis (Table 4), the  $A_{240}/A_{420}$ ,  $A_{250}/A_{365}$  showed an extremely significant negative correlation with MBP. With the increase in the soil TOM content, the proportion and aromaticity of non-humified and humified substances in the soil DOM first increased and then slightly decreased. The relative molecular weight of the soil DOM, the proportion of hydrophobic organic component content and non-hydrophobic organic components, and the proportion of aromaticity and biogenic sources of the soil DOM showed a decreasing trend and tended to stabilize at  $35 \text{ g}\cdot\text{kg}^{-1}$  of the soil TOM. With the increase in the soil TOM content, a large number of stable aggregates were formed [31]. DOM mineralization increased the condensation degree and molecular weight of the soil DOM and reduced the content of hydrophobic organic components and the aromaticity of the soil DOM [32]. According to the correlation analysis, the  $S_{254}$ ,  $S_{260}$ , and  $S_{280}$  of the soil were significantly negatively correlated with the MBP, the  $S_R$  was significantly negatively correlated with the MBP, the  $A_3/A_1$  was extremely significantly positively correlated with the MBP, the  $A_3/A_2$  was significantly positively correlated with the MBP, and the  $A_2/A_1$  was significantly negatively correlated with the MBN. They indicated that with the increase in soil microbial activity, the humification degree, aromaticity, and hydrophobic component content of the soil DOM decreased, and the proportion of non-humified and humified substances increased. Compared with the results of the 3DEEM spectral parameters, the trend of changes in the DOM humification degree reflected by the UV spectral parameters was opposite, mainly because the humic

substances characterized by the UV spectral parameters were small-molecule humic substances containing more conjugated double bonds and aromatic ring structures. The humic substances characterized by the 3DEEM spectral parameters not only contained the humic substances characterized by UV spectroscopy, but also contained humic substances with functional groups such as aromatic compounds characterized by visible light, so the degree of humification in UV spectra decreased with the increase in TOM. Therefore, with the increase in the soil TOM content, the input of organic fertilizer made the soil TOM reach a certain threshold, the ratio of the soil DOM functional group structure tended to stabilize, and under the conditions of this experiment, the TOM threshold was  $35 \text{ g}\cdot\text{kg}^{-1}$ . The research also found that with the increase in soil TOM content, the contents of the soil MBP and URE showed an Exponential (Exp3P2) upward trend and tended to stabilize when TOM reached the threshold of  $35 \text{ g}\cdot\text{kg}^{-1}$ . At this time, the activities of the soil MBP and SUC tended to stabilize, which meant that the degradation and transformation of OM by soil microorganisms tended to stabilize, and the functional group structure ratio of the soil DOM tended to stabilize (Fig. 10).

#### The Interaction between Soil DOM, Microorganisms, and Enzyme Activity

Microorganisms regulate carbon and energy fluxes through DOM conversion, further drive biogeochemical cycles [33], and play a central role in soil ecosystems. The input of DOM stimulates microbial activity and leads to the accelerated decomposition of stable soil organic matter (SOM). The changes in organic compost dosage induce physiological and enzymatic changes

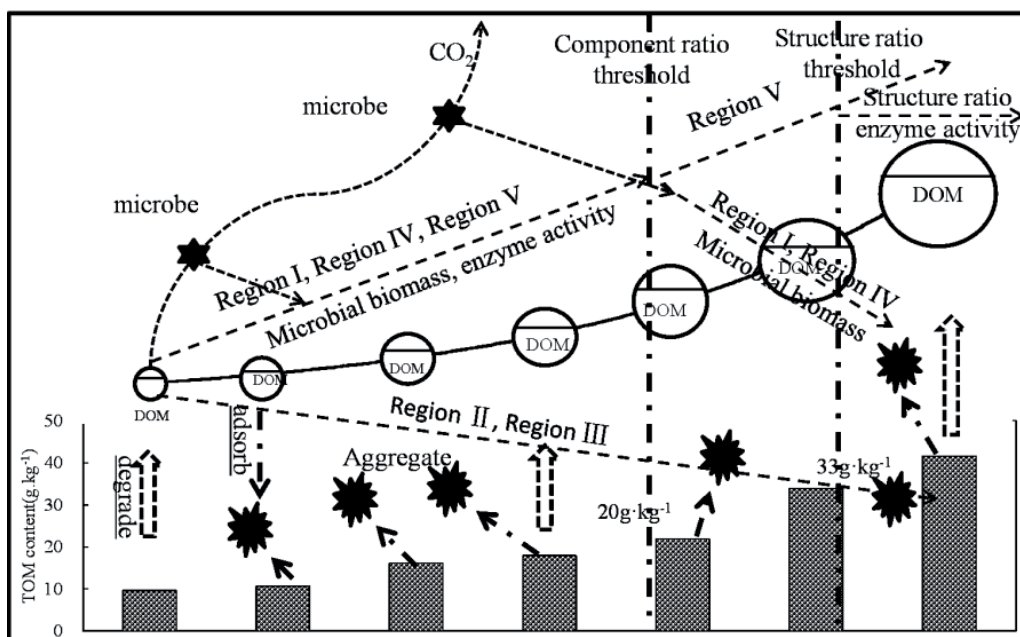


Fig. 10. Effect of soil TOM on DOM components.

Table 4. Correlation between soil DOM components and microbial and enzyme activities.

correlation	DOM	Region I	Region II	Region III	Region IV	Region V	FI	BIX	HIX	$A_{252}/A_{260}$	$A_{264}/A_{280}$	$A_{252}/A_{365}$	SUVA <sub>254</sub>	SUVA <sub>360</sub>	SUVA <sub>280</sub>	$S_r$	$A_2/A_1$	$A_3/A_1$	$A_4/A_1$	$A_5/A_2$	URE	SUC	MBC	MBP	MBN
TOM	0.939**	-0.207	-0.980**	-0.990**	-0.556	0.988**	-0.701	-0.777*	0.937**	0.973**	-0.911**	-0.960**	-0.931**	-0.942**	-0.936**	-0.799*	0.605	0.820*	0.765*	0.682	-0.024	0.093	0.828*	0.046	
DOM	1.000	-0.354	-0.924**	-0.920**	-0.781*	0.959**	-0.812*	-0.929	0.905**	0.991**	-0.786*	-0.844**	-0.816*	-0.832*	-0.830*	-0.683	0.642	0.639	0.532	0.544	-0.297	-0.038	0.684	-0.057	
Region I		1.000	0.232	0.114	0.583	-0.224	0.713	0.400	0.036	-0.279	-0.021	0.033	-0.045	0.072	0.094	-0.007	-0.791*	0.102	0.153	0.156	0.633	0.887**	0.310	0.898**	
Region II			1.000	0.986**	0.590	-0.992**	0.731*	0.784*	-0.913**	-0.957**	0.823*	0.963**	0.887**	0.916**	0.905**	0.847**	-0.555	-0.755*	-0.715*	-0.716*	-0.011	-0.040	-0.808*	-0.031	
Region III				1.000	0.524	-0.990**	0.645	0.757*	-0.959**	-0.960**	0.891**	0.985**	0.939**	0.953**	0.857**	0.857**	-0.497	-0.826*	-0.779*	-0.779*	-0.069	-0.069	-0.184	-0.878**	
Region IV					1.000	-0.636	0.900**	0.941**	-0.495	-0.717*	0.280	0.386	0.285	0.324	0.320	0.281	-0.608	-0.085	0.057	-0.032	0.729*	0.442	-0.137	0.385	
Region V						1.000	-0.741*	-0.833*	0.940**	0.984**	-0.847**	-0.955**	-0.893**	-0.917**	-0.908**	-0.824*	0.566	0.760*	0.697	0.684	-0.056	0.068	0.808*	0.047	
FI							1.000	0.877**	-0.514	-0.783*	0.447	0.552	0.412	0.472	0.470	0.397	-0.842**	-0.284	-0.192	-0.115	0.610	0.577	-0.207	0.564	
BIX								1.000	-0.750*	-0.897**	0.564	0.643	0.574	0.582	0.575	0.485	-0.600	-0.368	-0.232	-0.273	0.545	0.188	-0.449	0.158	
HIX									1.000	0.933**	-0.874**	-0.928**	-0.952**	-0.926**	-0.917**	-0.791*	0.338	0.778*	0.710*	0.767*	0.075	0.361	0.922**	0.346	
$A_{230nm}/A_{280nm}$										1.000	-0.842**	-0.902**	-0.864**	-0.875**	-0.870**	-0.732*	0.623	0.710*	0.619	-0.820*	-0.206	0.024	0.749*	-0.002	
$A_{230nm}/A_{320nm}$											1.000	0.907**	0.932**	0.909**	0.910**	0.687	-0.518	-0.929**	-0.878**	-0.609	-0.102	-0.321	-0.845**	-0.190	
$A_{230nm}/A_{365nm}$												1.000	0.945**	0.967**	0.956**	0.892**	-0.446	-0.888**	-0.866**	-0.786*	-0.211	-0.251	-0.906**	-0.195	
SUVA <sub>254nm}</sub>													1.000	0.965**	0.961**	0.777*	-0.374	-0.863**	-0.837**	-0.820*	-0.228	-0.372	-0.949**	-0.328	
SUVA <sub>260nm}</sub>														1.000	0.999**	0.888**	-0.434	-0.911**	-0.881**	-0.849**	-0.245	-0.295	-0.912**	-0.210	
SUVA <sub>280nm}</sub>															1.000	0.876**	-0.451	-0.912**	-0.880**	-0.841**	-0.233	-0.283	-0.899**	-0.191	
$S_r$																1.000	-0.219	-0.814*	-0.810*	-0.408	-0.408	-0.307	-0.823*	-0.239	
$A_2/A_1$																	1.000	0.380	0.307	-0.012	-0.570	-0.626	0.088	-0.727*	
$A_3/A_1$																		1.000	0.980**	0.699	0.359	0.418	0.838**	0.234	
$A_4/A_2$																			1.000	0.736*	0.501	0.428	0.833*	0.262	
URE																				1.000	0.592	0.475	0.886**	0.482	
SUC																					1.000	0.663	0.442	0.634	
MBC																						1.000	0.597	0.948**	
MBP																							1.000	0.572	

Note: n = 7; \* denotes a significant correlation when P&lt;0.05; \*\* denotes a highly significant correlation when P&lt;0.01.

in microbial communities, thereby regulating SOM decomposition through different mechanisms [34]. The DOM supporting microbial metabolism is composed of biologically unstable DOM (BLDOM) and recalcitrant DOM (RDOM). RDOM contributes to long-term carbon sequestration, and the humic acid class is the main component of RDOM. In this study, the different organic compost dosages added resulted in an exponential increase in the DOM content of the soil. The “hunger game” hypothesis was used to explain the changes of microorganisms under different nutritional conditions [35]. The hypothesis divides soil microorganisms into K-strategists and r-strategists. K-strategists tend to utilize stubborn organic carbon and exhibit lower reproductive rates, and r-strategists tend to use unstable organic carbon and have higher reproductive rates [36, 37]. In the nutrient-poor stage with less organic compost dosage, bacteria with low ribosomal RNA gene operon (*rrn*) copy numbers are favored. These K-strategists are characterized by slow growth and efficient resource utilization and rely on cooperation among species to mitigate resource scarcity. Therefore, during this stage, the soil MBC, MBN, MBP, and enzyme activity showed an upward trend, but the DOM content increased slowly. In the nutrient-rich stage with high organic compost dosage, r-strategists with high *rrn* copy numbers and fast growth rates proliferate by rapidly exploiting abundant resources, leading to intensified competition among species. During this stage, the soil MBC, MBN, MBP, and enzyme activity showed a decreasing trend, but the DOM content increased exponentially. The increases in DOM content were mainly due to the increase in the proportion of humic acid classes and the reduction in the proportions of unstable DOM (such as tyrosine protein, tryptophan protein, fulvic acid class, and dissolved microbial metabolites). The correlation analysis (Table 4) showed a significant negative correlation between tryptophan protein, fulvic acid class, and MBP. At the same time, the relative molecular weight and humification degree of DOM, the content of hydrophobic organic components, the aromaticity of DOM, and the proportion of biogenic sources all decreased during this stage. The stability of soil DOM was enhanced. Therefore, increasing the application of organic compost has promoted the fixation of soil carbon.

### Conclusions

With the increase in organic compost dosage, the soil TOM content increased linearly. With the increase in soil TOM content, the DOM content increased exponentially, and the proportion of humic acid-like components in the soil DOM showed a linear increasing trend. The proportion of tryptophan protein and fulvic

acid components showed a linear decreasing trend. The proportion of tyrosine protein and microbial metabolites in the soil DOM components, as well as the soil DOM fluorescence parameters *FI* and *BIX*, and the proportion of lignin and quinone, all showed initial increases followed by decreases in Exponential (Exp3P2) changes, and a turning point occurred at 20 g·kg<sup>-1</sup> of TOM. With the increase in the soil TOM content, the substituents on the benzene ring of the soil DOM transformed from non-polar functional groups, such as fatty chains and esters, to polar functional groups such as hydroxyl, carboxyl, and carbonyl groups. The molecular condensation degree of soil DOM increased, the molecular weight of DOM increased, the degree of soil DOM humification increased, and the hydrophobic organic component content and the aromaticity of DOM decreased. With the increase in the soil TOM content, the relative molecular weight and humification degree of DOM, the content of hydrophobic organic components, and the proportion of aromatic and biogenic sources of DOM showed an exponential downward trend. The proportion and aromaticity of non-humified and humified substances of DOM showed an initial increasing and then decreasing trend and reached the highest value at 35 g·kg<sup>-1</sup> of TOM. With the increase in organic compost dosage, the carbon and nitrogen content of microbial biomass and sucrase activity (SUC) in farmland soil showed patterns of initial increases and then decreases in Exponential (Exp3P2) changes. The interaction between soil DOM and microorganisms conformed to the “Hunger Games” hypothesis. When the soil TOM content was below 20 g·kg<sup>-1</sup>, with the increase in TOM content, soil microbial activity was enhanced, and the degradation of DOM by soil microorganisms and the DOM content of microbial metabolites increased. The soil DOM content increased slowly, the proportion of small-molecule active DOM components increased, and the biogenic and aromatic properties of DOM were enhanced. When the soil TOM content was high and above 20 g·kg<sup>-1</sup>, with the increase in TOM content, soil microbial activity decreased and the effect of microorganisms on DOM was weakened, soil DOM was enhanced by organic particulate matter, and DOM content rapidly increased, the proportion of macromolecular inert humic acid DOM components increased, DOM terrigenous properties were enhanced, and the polar functional groups in the substituents on the DOM benzene ring increased. The input of organic fertilizer affected the structure of soil DOM components by regulating soil microbial activity and organic particulate matter. In production, organic compost dosage should be applied in moderation to control soil TOM at around 20%.

### List of Abbreviations and Their Full Names

Abbreviations	Full names	Abbreviations	Full names
$A_{253}/A_{203}$	Absorbance at 253 nm/absorbance at 203 nm	$-\text{NH}_2^-$	Amino
$A_{250}/A_{365}$	Absorbance at 250 nm/absorbance at 365 nm	$\text{NH}_4^+-\text{N}$	Ammonia nitrogen
$A_{240}/A_{420}$	Absorbance at 240 nm/absorbance at 420 nm	NMR	Nuclear magnetic resonance
BD	Soil bulk density	$\text{NO}_3^--\text{N}$	Nitrate nitrogen
BLDOM	Biologically unstable dissolved organic matter	-OH	Hydroxyl
<i>BIX</i>	Biological index	$\text{P}_2\text{O}_5$	Phosphorus pentoxide
C	Carbon	RDOM	Recalcitrant dissolved organic matter
-COOH	Carbonyl	Regions I	Tyrosine-like compounds
3DEEM-FRI	Three-dimensional fluorescence spectroscopy with fluorescence regional integrated volume integration	Regions II	Tryptophan-like compounds
3DEEM	Three-dimensional fluorescence spectra	Regions III	Fulvic acid-like substances
DOM	Dissolved organic matter	Regions IV	Dissolved microbial metabolites
DOC	Dissolved organic carbon	Regions V	Humic acid-like organic fractions
EEM	Fluorescence spectrum	RNA	Ribonucleic acid
Em	Emission wavelength	-SH	Sulfur based
Ex	Excitation wavelength	SOC	Soil organic matter
Exp3P2	Triple exponential smoothing curve model	$S_R$	Slope ratio
<i>FI</i>	Fluorescence index	SUC	Soil sucrase activity
FRI	Fluorescence regional integrated volume integration	$\text{SUVA}_{254}$	Specific ultraviolet absorbance at 254 nm
HITACHI	Hitachi high-technology corp	$\text{SUVA}_{260}$	Specific ultraviolet absorbance at 260 nm
<i>HIX</i>	Humification index	$\text{SUVA}_{280}$	Specific ultraviolet absorbance at 260 nm
IP	Inorganic phosphorus	TN	Total nitrogen
IR	Including infrared spectroscopy	TOM	Total organic matter
KCl	Potassium chloride	TP	Total phosphorus
MBC	Microbial biomass carbon	URE	Soil urease activity
MBN	Microbial biomass nitrogen	UV	Ultraviolet spectroscopy
MBP	Microbial biomass phosphorus	$\varphi_{(i,n)}$	Specific region standard volume
MS	Mass spectrometry	$\varphi_{(T,n)}$	Whole region standard volume
N	Nitrogen		

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### Conflict of Interest

The author declares no conflict of interest.

### Data Availability Statement

The data that support the findings of this study are included in the manuscript, and further inquiries can be forwarded to the corresponding author.

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