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Molecular changes of ferric oxide bound soil humus during the decomposition of maize straw

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Abstract

Background: The mechanism of organic carbon sequestration in different genesis of paddy soils has not been well understood. A 6-month incubation experiment, adding maize straw, was performed in three different genesis of paddy soils and an uncultivated marsh soil. Fourier transform infrared (FTIR) spectroscopy was used to investigate the characteristics of Fe/Al bound organic carbon (Fe/Al-OC) extracted with a mixed solution of 0.1 M NaOH and $Na_4P_2O_7$ from soils.

Results: The function groups of Fe/Al-OC such as phenol-C, amide-C, and C–O groups of polysaccharides were demonstrated by FTIR spectroscopy. Compared to control treatment, the proportion of phenol-C of Fe/Al-OC added maize straw increased with time, but the C–O groups of polysaccharides decreased.

Conclusions: These results indicate that polysaccharide groups of Fe/Al-OC mainly contribute to the sequestration of Fe/Al-OC during the primary decomposition stage of maize straw, and the phenol group was stable in different genesis of paddy soils. Both of the polysaccharides and phenol groups were important to C sequestration in paddy soils and marsh soil.

Keywords: Fourier transform infrared (FTIR) spectroscopy, Soil carbon sequestration, Paddy soil, Fe/Al bound organic carbon, Hydrophobic substance

Background

The carbon cycling in soils is a critical process of earth surface system closely related to global climate change, and the interest in agricultural soil as global carbon storage has increased in recent years [1, 2]. Organic matter (OM) mineralization in soil may be reduced through increasing organic carbon (OC) sequestration into hydrophobic domains in humic material of comfortable composition [3]. The content of OC was high in paddy soil than in dry land and the mineralization rate of OC was lower in anaerobic culture rather than aerobic culture in paddy soil [4].

In addition, the OC sequestration in China's rice paddies related to chemical binding of iron or aluminum [5]. A number of reports relate to the return of crop residue into field [6–11]. However, the mechanism of carbon sequestration of crop residue in different origin of paddy soils is not clear until now. The composition and nature of humus derived from different origin paddy soils are various [12]. Moreover, humified OM and the reduction of their microbial mineralization could protect the labile organic compounds effectively in soil [13]. The microbial and enzyme activities are seriously changed [4], and the formation of soil humus is considerably different during decomposition of crop residue [8]. With the decomposition of maize residue, the characteristic of soil humus is various [14, 15]. Not only Solid-state ^{13}C nuclear magnetic resonance (NMR) spectroscopy can be used to study soil organic matter (SOM) [16–18], but also Fourier transform infrared (FTIR) spectroscopy is also a useful technique in distinguishing the main chemical functional groups of SOM, such as carbohydrates, lignin, cellulose, fats, and/or lipids and proteinaceous compounds [19]. The direct information of functional groups in SOM fraction could be investigated by FTIR spectroscopy [20].

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Only a few studies on the mechanisms of SOM accumulation in representative paddy soils are documented [19, 21]. Are the characteristics of organic fractions of paddy soils and uncultivated marsh soil similar in the course of carbon sequestration? Which functional groups favor soil carbon sequestration in paddy soils?

The objective of this work is to investigate the characteristics of Fe/Al bound organic carbon (Fe/Al-OC) in different genesis of paddy soils by FTIR spectra.

Methods
Soils
Three representative paddy soils and an uncultivated marsh soil were used in this study. The paddy soils (Hydrgric Anthrosols) included: First, Soil TP, developed from clayey lacustrine deposits in the Tai-Lake plain, from Wu Jiang County, Jiangsu Province; second, Soil RP, derived from red earth in the rolling red soil area, from Jin Xian Country, Jiangxi Province; and third, Soil PP, formed on Jurassic purple shale and sandstone, from the Sichuan purple sandstone rolling area, Chongqing Municipality. Forth, Soil MS, a marsh soil Calcaric Fluvisol, from Shengjin Wetland, Chizhou City, Anhui Province. The marsh soil was used for comparison with paddy soil because it was the precedent soil for rice paddies in the lower reaches of Yangtze River valley. The soil samples were taken from topsoil at $0-15$ cm depth in early spring of 2008 and were three field replicates.

Coarse organic matters (free roots and plant debris) and stones in surface samples (0–15 cm) of these soils were picked out with tweezers after sampling. The samples were sieved (<2 mm) and stored at 20 °C for 10 days before incubation. It was used to minimize the variations which were caused by microbial activity at different temperatures.

Maize residue
Maize residue was used as organic amendment to the soil. Maize straw was collected from a maize farm and air-dried at room temperature. The kernels and roots were removed. In addition, other parts of maize straw were cut to small pieces and passed through a 0.42 mm sieve and were oven-dried at 60 °C. The maize residue contained 438 and 6.81 g N kg^{-1} on dry base.

Laboratory incubation
The field triplicate soils were mixed before incubation in order to keep the same type of soils in a similar condition. The maize residues were used as C input to soils and mixed with soils in a percentage of 6 % (dry base). The C/N ratio of the residual/soils mixture was adjusted to 20 using a solution of $(NH_4)_2SO_4$. The mixtures were weight into plastic containers and the water holding capacity (WHC) is adjusted to 60 %. Then, they were incubated in

an incubator (LRH-250-S, Medicine Machinery Co. Ltd., Guangdong, China) under 25 °C in triplicate. The distilled water was added into samples per 48 h in order to keep the WHC at 60 %. In addition, the control treatments without maize residuals were also incubated at the same time. Incubations were carried out for 30, 90, and 180 days.

Extraction of iron/aluminum bound fractions
The Ca bound organic carbon (Ca-OC) and iron/aluminum bound organic carbon (Fe/Al-OC) were extracted using 0.5 M Na_2SO_4 and a mixed solution of 0.1 M NaOH and $Na_4P_2O_7$ following the outline of Xu and Yuan [22]. The samples were extracted by shaking for 1 h with a 1:10 w/v of 0.5 M Na_2SO_4 solution and then the mixture was remained for 24 h. The mixture was centrifuged approximately at 1780g for 15 min at 20 °C in order to obtain the Ca-OC. The supernatants were filtered (0.22 μm) in a 250 mL plastic bottle after centrifuging, and the residue was washed with a 1 % Na_2SO_4 solution. The procedure above was repeated in triplicate, and the supernatants were combined. Then, it was centrifuged at approximately 7150g for 20 min at 20 °C and filtered (0.22 μm) into a volumetric flask.

The residue was washed with distilled water and centrifuged at 7150g for 20 min at 20 °C in triplicates after extracting Ca-OC. The residue was subsequently extracted by shaking for 1 h with a 1:10 w/v of 0.1 mol/L NaOH and $Na_4P_2O_7$ (1:1 v/v, pH = 13.0) solution and then the mixture was remained for 24 h. The mixture was centrifuged at approximately 1780g for 15 min at 20 °C in order to obtain the Fe/Al-OC. After centrifuging, the supernatants were transferred into a 250 mL plastic bottle by filtering (0.22 μm). The residue was the humin fraction. The steps above were also repeated in triplicate, and then the supernatants were transferred in the same bottle. All the supernatants were centrifuged at approximately 7150g for 20 min at 20 °C and filtered (0.22 μm) into a volumetric flask. The Fe/Al-OC was dialyzed against deionized water, and freeze-dried.

Soil analysis
Basic properties
Basic properties are provided in Table 1. Soil pH was measured with a composite electrode under a 1:2.5 soil to water ratio. Organic carbon was determined by digestion with 0.4 mol/L potassium dichromate and back titrating with 0.2 mol L^{-1} ferrous ammonium sulfate. Total *N* was measured by kjeldahl method. The free iron oxide (Fe$_d$) was extracted by a Dithionite–citrate system buffered with sodium bicarbonate (DCB) and was measured with atomic absorption spectrometer (AAS). Clay content was determined by pipette method. The cation exchange capacity (CEC) was measured by NH$_4$OAC buffer [23].

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Table 1 Physicochemical properties of the representative types of soils studied

Soil types	SOC (g kg^{-1})	Fe$_d^a$ (g kg^{-1})	Total N (g kg^{-1})	C/N	Clay (g kg^{-1})	CEC^b (Cmol kg^{-1})	pH (H$_2$O)	WHC^c (g kg^{-1})
TP	19.02	14.99	1.65	11.56	273.1	18.91	5.28	666
RP	21.79	15.95	1.89	11.54	198.0	8.83	4.61	708
PP	17.61	10.00	1.70	10.37	156.9	23.79	7.26	594
MS	17.88	15.05	1.74	10.29	97.0	14.01	7.33	573

The soils used in this study were three replicates and mixed before incubation in order to keep the same type of soils in a similar condition. *TP* Tai-Lake plain soil; *RP* red earth paddy soil; *PP* purple paddy soil; *MS* marsh soil

^{a} Free ferric oxide

^{b} Cation exchange capacity

^{c} Water holding capacity

The yields of Fe/Al-OC extracted from soil without incubation (Fig. 1a) and soils incubated for 30, 90, and 180 days (Fig. 1b) were calculated by the percentages of dry weight of Fe/Al-OC in the weight of soils used in this study.

FTIR spectroscopy analysis

Spectral characterization of Fe/Al-OC samples was performed with a Bruker 66v/s FTIR spectrometer in the mid IR area from 4000 to 400 cm^{-1}. Spectra were recorded with a resolution of 4 cm^{-1} and 128 scans per sample. The

Fig. 1 The yield of Fe/Al-OC extracted from soils without incubation (**a**) and soils incubated for 30, 90, and 180 days (**b**); *TP* Tai-Lake plain soil; *RP* red earth paddy soil; *PP* purple paddy soil; *MS* marsh soil; In figure **b** the results are mean ± SD (*n* = 3)

background of ambient air was submitted. The spectrum was obtained on pellets, adding 1 mg of lyophilized sample to 200 mg KBr and previously dried in an oven. The software used to analyze the data was OMNIC version 6.1 for Windows (Thermo Nicolet Instrument Corp. Madison, WI). Firstly, the mid-infrared spectroscopy was automatically baseline corrected and normalized. Then the area was integrated between the tangent of two trough points of the peak and the spectroscopy. The total area of peaks was calculated by adding each peak area. Finally, the percentage of each peak for total area of peaks was calculated.
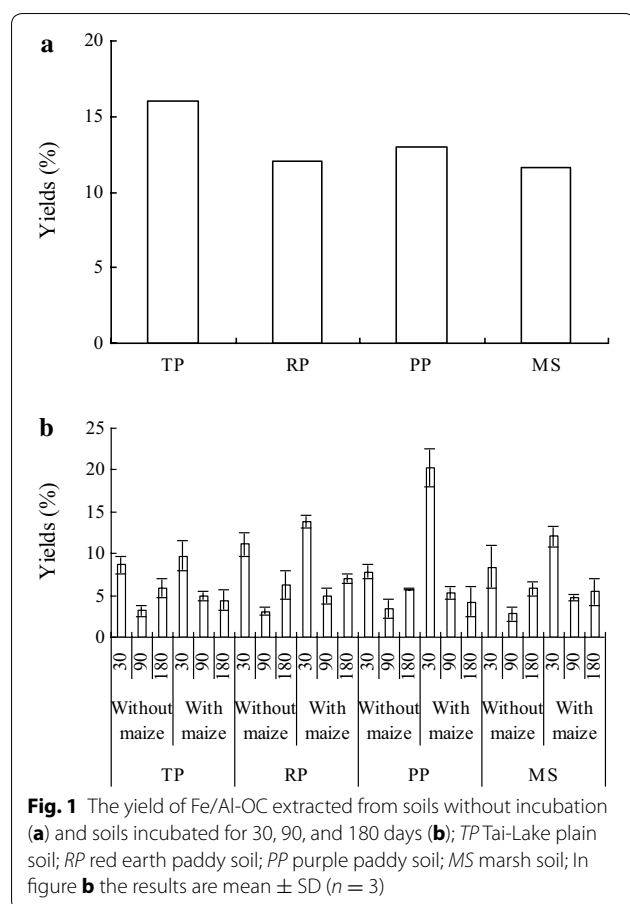
Results
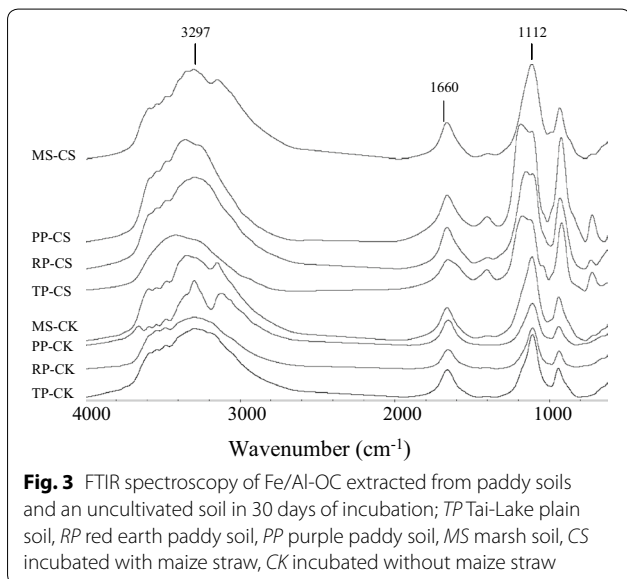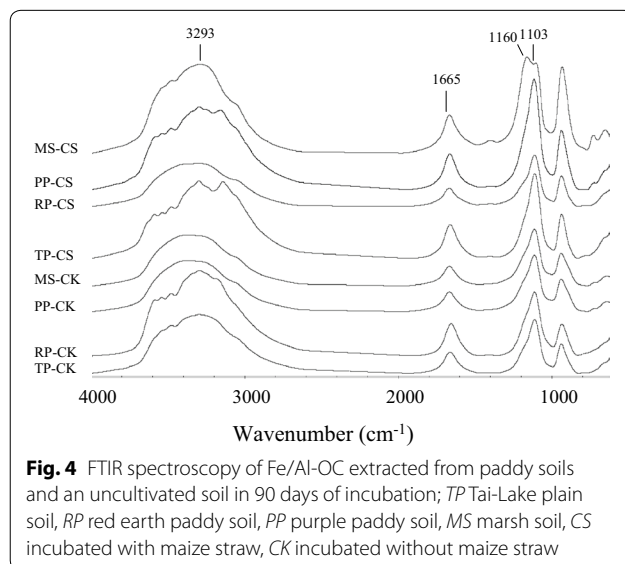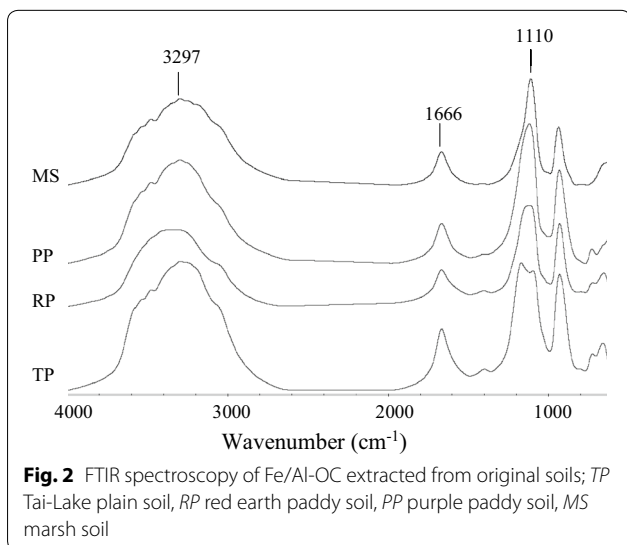
Organic carbon functional groups of Fe/Al-OC

The spectral region was from 4000 to 400 cm^{-1}, and automatic baseline correction was conducted. The map profiles were created for peak heights at 3500–3200, 1666, 1160, and 1110 cm^{-1} (Figs. 2, 3, 4, 5). The broad intense band at 3500–3200 cm^{-1}, corresponded mainly to stretching vibrations of H-bonded hydroxyl (O–H) groups of phenol, as well as traces of amine (N–H) stretch [19, 24–26]. The broad band near 1666 cm^{-1} was assigned primarily to C=O of amide I [19, 27]. The absorption at 1160 cm^{-1} attributed to C–O band in both polyalcoholic and ether functional groups, such as those in oligo- and polysaccharide [3]. The strong band at about 1110 cm^{-1} could be attributed to C–O stretching vibrations of polysaccharides [19, 28, 29] and/or possibly contributed from phosphate groups [30].

In this study, the areas of absorbance peaks of functional groups are shown in Tables 2, 3, 4, 5. The functional groups of Fe/Al-OC were demonstrated in FTIR spectra including phenols, amide polymers, and polysaccharide (Figs. 2, 3, 4, 5).

Variation on the characteristics of Fe/Al-OC in different incubation stages

Compared to Fe/Al-OC extracted from paddy soils and marsh soil without maize straw, the relative contents of

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Fig. 2 FTIR spectroscopy of Fe/Al-OC extracted from original soils; *TP* Tai-Lake plain soil, *RP* red earth paddy soil, *PP* purple paddy soil, *MS* marsh soil

Fig. 4 FTIR spectroscopy of Fe/Al-OC extracted from paddy soils and an uncultivated soil in 90 days of incubation; *TP* Tai-Lake plain soil, *RP* red earth paddy soil, *PP* purple paddy soil, *MS* marsh soil, *CS* incubated with maize straw, *CK* incubated without maize straw

Fig. 3 FTIR spectroscopy of Fe/Al-OC extracted from paddy soils and an uncultivated soil in 30 days of incubation; *TP* Tai-Lake plain soil, *RP* red earth paddy soil, *PP* purple paddy soil, *MS* marsh soil, *CS* incubated with maize straw, *CK* incubated without maize straw

phenol-C of Fe/Al-OC extracted from soil TP, RP, PP, and MS with maize straw decreased by 12.70, 8.12, 8.53, and 1.58 % in 30 days of incubation, respectively. Moreover, the relative contents of polysaccharide-C of Fe/Al-OC extracted from soil TP, RP, PP, and MS increased by 11.49, 6.85, 9.40, and 1.01 %, respectively. In addition, the proportions of amide-C of Fe/Al-OC extracted from soil TP, RP, and MS increased by 1.21, 1.27, and 6.85 %, but decreased by 0.87 % in soil PP (Fig. 3; Table 3). These results showed that the relative content of polysaccharide groups increased in the primary stage of incubation with maize straw, and the amide groups were stable. However, the proportion of phenol groups of Fe/Al-OC extracted from paddy soils and marsh soil decreased.

Compared to soils without maize straw, the proportion of phenol-C of Fe/Al-OC extracted from soil TP, RP, PP, and MS with maize straw decreased by 2.18, 3.99, 3.40, and 2.71 % in the incubation of 90 days, respectively. The proportion of polysaccharide-C of Fe/Al-OC extracted from soil TP, RP, PP, and MS with addition of maize straw increased by 1.75, 4.05, 3.27, and 3.01 %, respectively. Moreover, the proportions of amide-C of Fe/Al-OC extracted from soil TP, RP, PP, and MS with maize straw increased by 0.43 % and 0.13 % in soil TP and PP, but decreased by 0.06 and 0.30 % in soil RP and MS (Fig. 4; Table 4). These results showed that the relative content of polysaccharide groups increased in 90 days of incubation. However, the increase of polysaccharide groups was lower in 90 days of incubation than in 30 days of incubation in paddy soil, while it was higher than in 30 days of incubation in marsh soil. Moreover, the amide groups were also stable. The proportion of phenol groups of Fe/Al-OC extracted from paddy soils and marsh soil decreased too. Compared with marsh soil, it decreased much in marsh soil more than in paddy soils.

Compared to soils without maize straw, the proportions of phenol-C of Fe/Al-OC extracted from soil TP, RP, PP, and MS with maize straw increased by 4.71, 0.34, 1.22, and 2.70 % in the incubation of 180 days, respectively. In addition the proportion of polysaccharide-C of Fe/Al-OC extracted from soil TP, RP, PP, and MS decreased by 3.27, 0.56, 1.80, and 2.77 %, respectively. Relative contents of amide-C of Fe/Al-OC extracted from soil TP decreased by 1.44 %, but it increased by 0.22, 0.59, and 0.07 % in Fe/Al-OC extracted from soil RP, PP, and MS, respectively (Fig. 5; Table 5).
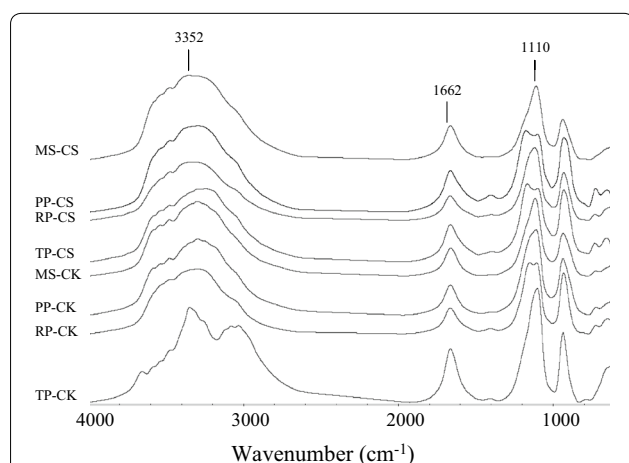
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Fig. 5 FTIR spectroscopy of Fe/Al-OC extracted from paddy soils and an uncultivated soil in 180 days of incubation; *TP* Tai-Lake plain soil, *RP* red earth paddy soil, *PP* purple paddy soil, *MS* marsh soil, *CS* incubated with maize straw, *CK* incubated without maize straw

Table 2 Relative areas of the main absorption peaks of IR spectrum of Fe/Al-OC extracted from original soils (%) (semi-quantitative)

Soil types	Phenol-C	Amide-C	Polysaccharide-C
TP	77.51	6.39	16.10
RP	74.43	6.07	19.50
PP	74.31	5.78	19.91
MS	78.16	6.22	15.62

Table 3 Relative areas of main peaks of IR spectrum of Fe/Al-OC extracted from paddy soils and marsh soil after 30 days of incubation (%) (semi-quantitative)

Soil types	Addition of maize straw (%)	Phenol-C	Amide-C	Polysaccharide-C
TP	0	81.08	7.18	11.73
RP	0	82.79	6.44	10.77
PP	0	80.48	8.03	11.49
MS	0	79.47	7.09	13.44
TP	6	68.38	8.39	23.23
RP	6	74.67	7.71	17.62
PP	6	71.94	7.17	20.89
MS	6	77.89	7.66	14.45

Table 4 Relative areas of main peaks of IR spectrum of Fe/Al-OC extracted from paddy soils and marsh soil after 90 days of incubation (%) (semi-quantitative)

Soil types	Addition of maize straw (%)	Phenol-C	Amide-C	Polysaccharide-C
TP	0	80.21	6.42	13.37
RP	0	81.52	6.58	11.90
PP	0	78.57	6.51	14.92
MS	0	78.65	6.52	14.83
TP	6	78.03	6.85	15.12
RP	6	77.53	6.53	15.94
PP	6	75.17	6.64	18.18
MS	6	75.95	6.22	17.84

Table 5 Relative areas of main peaks of IR spectrum of Fe/Al-OC extracted from paddy soils and marsh soil after 180 days of incubation (%) (semi-quantitative)

Soil types	Addition of maize straw (%)	Phenol-C	Amide-C	Polysaccharide-C
TP	0	71.84	9.09	19.06
RP	0	74.60	6.56	18.84
PP	0	76.08	6.59	17.34
MS	0	77.89	6.82	15.29
TP	6	76.55	7.66	15.80
RP	6	74.94	6.78	18.28
PP	6	77.30	7.17	15.53
MS	6	80.59	6.89	12.52

polysaccharide C–O stretching or to vibrations of a SiO_2-related impurity in humic substances [31]. However, it was noteworthy that the extraction itself was known as leading to incorporations of phosphate and/or pyrophosphate in the extracted organic matter. Francioso et al. [30] attributed the absorption bands at 1100 to 1000 cm^{-1} mainly to phosphate groups as a result of high concentration of total P in the humic substances extracted by NaOH plus pyrophosphate. Rulmont et al. [32] showed that the strongest absorbance band of orthophosphate was near 1050 cm^{-1}. In this study, the extraction condition was the same, thus the effect of phosphate could be ignored.

Stability of phenol and polysaccharides groups of Fe/Al-OC

In this study, the results of FTIR spectroscopy showed that the proportion of polysaccharide increased in the primary stage, and then it decreased due to the rapid depletion without new carbon input. This was in line with the differences of the amounts of Fe/Al-OC. It

Discussion

Effect of phosphate and/or pyrophosphate on the bands of FTIR spectroscopy

The assignment of the absorption bands at about 1100 cm^{-1} was frequently assigned to alcoholic and

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indicated that the carbohydrate was quick in response to the C changes of Fe/Al-OC. However, the phenol-C of Fe/Al-OC was protected well. Conversely, the C–O groups of polysaccharides of Fe/Al-OC did not remain steadily during the decomposition of maize straw. A number of studies showed that the reaction between humus substances and iron oxides favored carbon sequestration in paddy soils [33, 34, 35]. Xie and Shang [36] observed that the functional quinone–phenol groups presented in humic substances might contribute to the complexed Fe(III) reduction. Moreover, the Fe(III)/Fe(II) redox couple in relation to oxidation and reduction of iron and humic substances had been described in detail [37–40].

Possibly, the Fe/Al-OC was protected by phenol groups against further degradation. In this study, the results indicated that the phenol-C was more stable than polysaccharide-C. Rumpel et al. [41] considered that microbial-derived polysaccharides were most likely stabilized preferentially by mineral interactions compared to plant-derived polysaccharides. Because the polysaccharides were mainly derived from maize straw in this study, and the polysaccharides were not stable and decreased with time.

Selective protection of hydrophilic organic matter

The selective preservation of aliphatic-C in humic acids had been demonstrated [3]. In this study, the absorbance peaks at about 2920 and 2850 cm^{-1} were not found. Possibly, the aliphatic-C was decreased during the purification process.

Proteins were showed by the band at 1666 cm^{-1} [42]. It was considered as one of the typical hydrophilic natural organic matters except polysaccharides and other monomeric biopolymer such as amino sugars [43]. The recalcitrance of proteins in humic substances was achieved by encapsulation into hydrophobic domains of humic substances. Proteinaceous moieties might be physically encapsulated within the aliphatic moieties of humic acids [44]. Protein groups of Fe/Al-OC did not show distinct changes for the investigated samples. Because of overall negative charge, acidic regions and hydrophobic mechanisms, proteins could establish interaction with cations [27].

Conclusions

With the help of FTIR spectroscopy, we were able to effectively identify the variation on the functional groups representing molecular structure of Fe/Al-OC during the decomposition of maize straw in soil. The results of FTIR spectroscopy showed that proportion of polysaccharide-C decreased with time, but the phenol-C tended to increase with time. These results indicate that polysaccharide groups of Fe/Al-OC mainly contribute to the

sequestration of Fe/Al-OC during the primary decomposition stage of maize straw, and the phenol group was stable in different genesis of paddy soils. Both of the polysaccharides and phenol groups were important to C sequestration in paddy soils and marsh soil.

Authors' contributions

XS carried out the molecular genetic studies, participated in the sequence alignment and drafted the manuscript. LL and Jufeng Zheng participated in drafting the manuscript. XZ and JZ carried out the molecular genetic studies. QH participated in drafting the manuscript. GP conceived of this study, participated in its design and help draft the manuscript. All authors read and approved the final manuscript.

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