

生物炭与丛枝菌根真菌联用对土壤有机碳含量及稳定性的影响

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引用本文:

韩爽, 梁媛, 张德善, 等. 生物炭与丛枝菌根真菌联用对土壤有机碳含量及稳定性的影响[J]. 农业环境科学学报, 2026, 415(1): 127-138.

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生物炭与丛枝菌根真菌联用对土壤有机碳含量及稳定性的影响

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摘要:为探究生物炭和丛枝菌根真菌(AMF)联用对稻田土壤有机碳的固存效果和稳定性机制,本文通过室内培养实验,探究干湿交替条件下空白处理(CK)、单独添加生物炭(G_{BC})、单独接种AMF(G_{AMF})和二者联用(G_{BA})对土壤有机碳(SOC)组分、稳定性及铁碳结合机制的影响。结果表明: G_{BA} 处理显著提高了SOC、矿物结合态有机碳(MAOC)和铁结合有机碳(Fe-OC)。与CK相比, G_{BA} 使SOC提高了504.23%,MAOC提高了1306.80%。 G_{BA} 的颗粒态有机碳(POC)/SOC较CK显著降低了87.10%,但MAOC/SOC较CK提高了134.56%,达到92.17%,说明二者联用促进了POC向稳定的MAOC的转化。通过FTIR分析发现, G_{BA} 的芳香类碳较CK提高了30.57%。 G_{BA} 中Fe-OC的含量较CK提高了1397.51%,且OC/Fe摩尔比(2.72)表明 G_{BA} 中Fe-OC的产生主要通过吸附和共沉淀机制协同促进。XPS结果进一步揭示, G_{BA} 中 Fe^{3+} 占比最高(29.61%),其高电荷密度可强化矿物和有机碳络合,提高土壤碳稳定性。相关分析显示,AMF侵染率与MAOC和Fe-OC均表现为显著正相关($P<0.05$),生物炭添加提高了AMF的侵染率,协同促进了碳铁结合。研究表明,生物炭与AMF联用可显著提高稻田SOC含量和稳定性。

关键词:生物炭;丛枝菌根真菌;矿物结合态有机碳;铁结合态有机碳;稻田土壤

中图分类号:S153.6 文献标志码:A 文章编号:1672-2043(2026)01-0127-12 doi:10.11654/jaes.2025-0094

Effects of combined application of biochar and arbuscular mycorrhizal fungi on soil organic carbon content and stability

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Abstract: In order to explore the sequestration effect and stability mechanism of biochar and arbuscular mycorrhizal fungi (AMF) on soil organic carbon in paddy field, this paper explores the effects of blank treatment (CK), biochar addition alone (G_{BC}), arbuscular mycorrhizal fungi inoculation alone (G_{AMF}), and their combination (G_{BA}) on soil organic carbon (SOC) composition, stability, and iron-carbon binding mechanism of SOC under alternating dry-wet conditions through indoor incubation experiments. The results indicate that G_{BA} treatment significantly increased SOC, mineral-associated organic carbon (MAOC), and Fe-bound organic carbon (Fe-OC). Compared to CK, G_{BA} increased SOC by 504.23% and MAOC by 1306.80%. Compared to CK, G_{BA} significantly reduced particulate organic carbon (POC)/SOC by 87.10%, but MAOC/SOC increased by 134.56%, reaching 92.17%, suggesting that the combination of biochar and AMF promoted the conversion of POC to stable MAOC. FTIR analysis revealed that the aromatic carbon in G_{BA} increased by 30.57% compared to CK. The

收稿日期:2025-01-25 录用日期:2025-04-14

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基金项目:2022姑苏乡土人才项目(苏市农人[2023]4号);苏州市关键核心技术攻关(社会发展)项目(2023SS06);江苏省研究生科研与实践创新计划项目(KYCX23_3346)

Project supported: 2022 Local Talent Project of Gusu (Suzhou Farmer [2023] No. 4); Suzhou Key Core Technology Research and Development (Social Development) Project (2023SS06); Postgraduate Research & Practice Innovation Project of Jiangsu Province (KYCX23_3346)

content of Fe-OC in G_{BA} increased by 1 397.51% compared to CK, and the OC/Fe molar ratio (2.72) indicated that the production of Fe-OC in C_{BA} was primarily promoted through adsorption and co-precipitation mechanisms. XPS results further revealed that G_{BA} had the highest proportion of Fe^{3+} (29.61%), and its high charge density enhanced the complexation of minerals and organic carbon, thereby improving soil carbon stability. Correlation analysis showed that MAOC and Fe-OC were significantly positively correlated with AMF colonization rate ($P < 0.05$), and biochar addition increased the AMF colonization rate, synergistically promoting carbon-iron binding. The results showed that the combination of biochar and AMF significantly enhances the content and stability of SOC in paddy soils.

Keywords: biochar; arbuscular mycorrhizal fungi; mineral-associated organic carbon; Fe-bound organic carbon; paddy field soil

土壤有机碳(Soil organic carbon, SOC)是土壤碳库的重要组成部分,其持久性一般受其化学顽固性、土壤团聚和矿物保护的控制^[1]。将SOC分为矿物结合态有机碳(Mineral-associated organic carbon, MAOC)和颗粒态有机碳(Particulate organic carbon, POC)可以更准确地了解SOC的固定和分解^[2]。MAOC是土壤中稳定的有机碳,决定了土壤碳的组成、容量和循环,影响SOC的积累和稳定^[3]。土壤中的矿物质通过阳离子桥接、配体交换、氢键、离子交换和范德华力与SOC小分子结合,形成稳定的MAOC^[4],其形成受外源碳种类和土壤微生物等因素的影响^[5]。自然界中大约21%的SOC与活性铁矿物直接结合,形成铁结合有机碳(Fe-bound organic carbon, Fe-OC),保护SOC免于微生物的降解^[6-7],这已被证实是碳固存的关键机制^[8-9]。铁氧化物和有机化合物之间的生物地球化学循环是影响SOC矿化和固存的关键因素之一^[10]。

生物炭施入土壤后不仅可作为碳源提高SOC含量,而且生物炭孔洞的物理保护和自身较高的芳香碳组分提高了SOC稳定性,另外,生物炭的表面官能团促进了低结晶性的矿物与土壤有机质之间的相互作用,这有利于MAOC积累,间接增强了SOC稳定性,这些作用均有利于土壤碳封存^[15, 11-12]。丛枝菌根真菌(Arbuscular mycorrhizal fungi, AMF)是土壤中广泛存在的一种真菌^[13],通过定殖在植物根系内部而与植物形成共生系统^[14],不仅可以促进植物对土壤养分的吸收,提高植物生物量,而且还可以通过菌丝缠绕和释放球囊霉素类相关蛋白(Glomalin-related soil proteins, GRSPs)与土壤颗粒通过物理黏连或化学吸附,提高土壤团聚体数量和稳定性,间接促进土壤碳封存^[15-16]。

单独施加生物炭和接种AMF均有利于SOC的固存已被证实。有学者尝试将生物炭和AMF联用研究二者的协同效应:Barna等^[17]的研究发现生物炭施入种植辣椒的土壤后,AMF的根系侵染率和GRSPs分泌被显著抑制,导致菌丝和胶结作用减少,无法形成菌丝网络与土壤颗粒结合,这可能削弱了土壤团聚,

不利于土壤碳封存。但Mason等^[18]发现生物炭能降低AMF的呼吸作用,减弱其对小麦种植土壤中的碳矿化程度,有利于土壤碳封存。故目前对生物炭和AMF联用对SOC的提升效果存在争议,二者协同对土壤碳总量和碳稳定性的影响,以及其协同固碳关键机制仍不明确。

我国是世界上最大的水稻生产国,2023年水稻种植面积占全国农田总面积的24.33%^[19-20],所以稻田土壤的固碳效果及作用机制对全球碳固存具有重要作用^[21]。稻田土壤长期淹水和干湿交替的种植环境会改变土壤通气条件,土壤始终处于氧化还原的循环过程,这不仅影响根际土壤中的铁形态变化,还影响了好氧微生物的活性,进而对SOC稳定性产生影响^[19, 22-23]。如李明远等^[24]发现干湿交替能显著影响稻田土壤铁氧化物的形态转化,增加无定形氧化铁(Fe_0)含量,而是否可能通过铁-有机碳络合增强SOC固存需要进一步探究。然而,生物炭和AMF联用对稻田干湿交替环境中土壤铁形态及其对碳稳定性的影响及关键作用机制仍不清楚。因此,本实验在干湿交替条件下,探究单独生物炭和AMF或二者联用对水稻生长、稻田SOC组分和稳定性的影响,研究铁氧化物介导的碳固定机制,为稻田SOC的封存提供一定的理论和技术支持。

1 材料与方法

1.1 供试材料

供试土壤来自安徽省巢湖市某稻田的无污染表层土壤(0~20 cm),土样自然风干后过2 mm筛保存备用,初始pH值为7.65, SOC为 $4.08 \text{ g} \cdot \text{kg}^{-1}$ 。生物炭原料为水稻秸秆,自然风干后在马弗炉 $500 \text{ }^\circ\text{C}$ 条件下限氧热解2 h后冷却待用,其pH值为9.49, SOC为 $450.6 \text{ g} \cdot \text{kg}^{-1}$,比表面积为 $120.08 \text{ m}^2 \cdot \text{g}^{-1}$ 。供试水稻品种为“徽两优丝苗”杂交水稻,水稻种子在实验室育苗至分蘖期移栽。本实验中所有土壤理化分析及微生物实验土样均采自根际土壤。

供试AMF菌种为根内球囊霉(*Glomus intraradices*, GI),由江苏大学环境生态研究所提供。AMF菌种扩散周期为4个月,宿主为高粱。所用的河沙(2 mm)和沸石(2 mm)经过高温高压蒸汽灭菌(120 °C,120 min),按1:1(m/m)完全混合均匀,作为AMF菌种培养基质。接种菌剂由孢子、菌丝、根段和基质组成。

1.2 盆栽实验

借鉴前人研究,分别设置添加1%(m/m)生物炭(G_{BC})、1%(m/m)丛枝菌根真菌接种菌剂(G_{AMF})、1%(m/m)生物炭和1%(m/m)丛枝菌根真菌接种菌剂复合物(G_{BA})以及不添加生物炭和丛枝菌根真菌的土壤(CK),共4组处理,每个处理设置3个重复,共12盆。未接菌处理组加入等质量灭活菌剂及10 mL过10 μm 滤膜的菌剂滤液。将供试土壤装入塑料花盆中,土壤及外源添加物共0.8 kg。

水稻种子于孔穴盘育苗至分蘖期后,移栽于上述各处理中,维持土壤淹水层为2 cm,自然落干后补充去离子水至2 cm淹水层,培养90 d后,分段收获水稻茎、叶和根系,并进行根际土壤取样,用于土壤理化性质、SOC组分和铁氧化物的测定。

1.3 指标及测定方法

1.3.1 土壤基本理化性质、菌根侵染率和植物生物量的测定

取1 g新鲜根样品(烘干后质量可忽略不计)切成2~4 cm的段,使用Phillips等^[25]的根系染色方法,即使用台盼蓝染色后在显微镜下覆盖观察。侵染率按照Brundrett等^[26]的方程式计算:

$$\text{总侵染率} = (\text{总交叉点数} - \text{无侵染交叉点数}) / \text{总交叉点数} \times 100\% \quad (1)$$

使用烘箱干燥法测量样品土壤含水量,土壤pH值采用pH计测量,土水比为1:2.5(m/V)。土壤阳离子交换量(CEC)通过三氯化六氨合钴浸提-分光光度法测定^[27]。所有新鲜水稻样品用蒸馏水洗净后,分地上部分和地下部分分别烘干至恒质量后称量取平均值,得到水稻生物量。

采用傅里叶红外光谱仪(Fourier transform infrared spectrometer, FTIR, Thermo6700, 美国)溴化钾压片法表征各处理官能团,测试波数4 000~500 cm^{-1} ,分辨率4 cm^{-1} 。使用X射线光电子能谱仪(X-ray photoelectron spectroscopy, XPS, Thermo Scientific K-Alpha, 美国)采集培养前后不同处理土壤的Fe 2p谱图。

1.3.2 SOC组分的测定

POC(>0.053 mm)和MAOC(<0.053 mm)通过六

偏磷酸钠湿筛分离^[28]:称取20 g过2 mm筛的风干土样于锥形瓶中,加入200 mL六偏磷酸钠溶液(5 $\text{g} \cdot \text{L}^{-1}$)。先用手摇15 min,再振荡16 h分散后将土壤混合液过0.053 mm筛,多次使用去离子水冲洗至滤液透明,收集土样。土样烘干后测定POC(筛上)和MAOC(筛下)。SOC、POC、MAOC及Fe-OC均采用重铬酸钾氧化-外加热法(HJ 625—2011)测定。

1.3.3 土壤样品中铁含量及铁氧化物含量的测定

使用连二亚硫酸钠-柠檬酸钠-碳酸氢钠(DCB)连续法提取Fe-OC和游离态氧化铁(Fe_d)^[29]。计算公式为:

$$S_{\text{Fe-OC}} = S_{\text{NaCl}} - S_{\text{DCB}} \quad (2)$$

$$f_{\text{Fe-OC}} = S_{\text{Fe-OC}} / S_{\text{SOC}} \times 100\% \quad (3)$$

$$\text{OC/Fe} = (S_{\text{Fe-OC}} / M_{\text{C}}) / (m_{\text{Fe}_d} / M_{\text{Fe}}) \quad (4)$$

式中: $S_{\text{Fe-OC}}$ 为Fe-OC含量, $\text{g} \cdot \text{kg}^{-1}$; S_{NaCl} 为氯化钠-碳酸氢钠提取的SOC含量, $\text{g} \cdot \text{kg}^{-1}$; S_{DCB} 为DCB法提取的SOC含量, $\text{g} \cdot \text{kg}^{-1}$; $f_{\text{Fe-OC}}$ 为Fe-OC占SOC的比值,%; S_{SOC} 为有机碳含量, $\text{g} \cdot \text{kg}^{-1}$; OC/Fe为铁结合态有机碳的碳与铁摩尔比; m_{Fe_d} 为土壤游离氧化铁的含量, $\text{g} \cdot \text{kg}^{-1}$; M_{C} 和 M_{Fe} 分别为碳和铁的摩尔质量, $\text{g} \cdot \text{mol}^{-1}$ 。

土壤中的络合态氧化铁(Fe_p)和 Fe_o 分别用碱性焦磷酸钠和酸性草酸铵提取^[30]。采用邻菲罗啉分光光度法测定土壤样品中全铁(Fe_t)^[9]、 Fe_p 、 Fe_o 和 Fe_d 的含量。

土壤铁的络合指数的计算公式为:

$$\text{铁的络合指数} = \text{Fe}_p / \text{Fe}_d \times 100\% \quad (5)$$

1.4 数据统计分析

采用SPSS 22软件、Graphpad Pism 9.5软件和Avantage软件进行数据分析。基于95%置信区间的显著统计差异通过单因素方差分析和t检验确定。使用双尾检验的Spearman相关分析来确定所有测量参数之间的相关性。

2 结果与讨论

2.1 生物炭与丛枝菌根真菌对土壤理化性质和植物生长的影响

2.1.1 pH值和CEC

盆栽培养90 d后,生物炭添加处理显著提高了土壤的pH值(表1, $P < 0.05$),与CK相比, G_{BC} 和 G_{BA} 的pH值分别增加了0.37个单位和0.32个单位,其原因可能是生物炭含有可溶性碱金属离子(Ca^{2+} 、 Mg^{2+} 、 K^{+} 和 Na^{+})以及碳酸盐、磷酸盐等无机矿物质^[31-34],这些成分通过水解或离子交换释放氢氧根,使土壤pH值升

表1 不同处理的土壤理化性质和植物生长情况

Table 1 Soil physicochemical properties and plant growth in different treatments

处理 Treatment	pH	阳离子交换量 CEC/(mol·kg ⁻¹)	侵染率 AMF colonization rate/%	地上生物量 Aboveground biomass/ (g·m ⁻²)	地下生物量 Underground biomass/ (g·m ⁻²)	根冠比 Root/shoot ratio	SOC含量 SOC content/(g·kg ⁻¹)
CK	7.50±0.07b	1.89±0.02c	<0.01±<0.01c	0.92±0.04c	0.54±0.08c	0.58±0.06b	2.60±0.26c
G _{BC}	7.87±0.01a	2.05±0.01a	<0.01±<0.01c	1.46±0.06b	0.84±0.06b	0.57±0.06b	8.13±0.20b
G _{AMF}	7.55±0.10b	1.91±0.01b	40.87±2.23b	1.52±0.06b	0.89±0.04b	0.58±0.02b	3.00±0.60c
G _{BA}	7.82±0.02a	1.98±0.01b	68.52±0.95a	1.68±0.11a	1.22±0.29a	0.72±0.12a	15.71±0.45a

注: 同列不同小写字母表示处理间差异显著($P<0.05$)。下同。

Note: Different lowercase letters in a column indicate significant differences among treatments at $P<0.05$. The same below.

高。CEC已被证明是促进SOC积累的关键因素^[35]。G_{BC}、G_{AMF}和G_{BA}的CEC较CK分别提高了8.47%、1.06%和4.76%，这会进一步影响土壤质量和缓冲能力^[36]。生物炭表面的官能团提供了大量的络合位点，导致较高的CEC^[33]，而AMF通过分泌的GRSPs形成土壤团聚体，改善了土壤质量，提高了CEC^[37]。

2.1.2 AMF侵染率和水稻生物量

培养90 d后，G_{AMF}的根系侵染率为40.87%，生物炭添加显著提高了AMF在水稻根系中的侵染率，G_{BA}较G_{AMF}的根系侵染率提高了67.65% (表1, $P<0.05$)。生物炭提高了水稻根系中AMF的泡囊发育，促进菌丝分支与生长(图1)，这与Wen等^[38]的研究结果相似，即生物炭提高了低水分土壤中水稻根系AMF侵染率(约40%)。一方面，生物炭带来的氮、磷、钾等营养物质及其多孔结构能为AMF生长提供养分和栖息地^[39-40]。另一方面，生物炭还可能改变植物和AMF之间的信号传导或化感物质的吸收，影响其共生关系^[17]。土壤的通气条件影响植物和AMF间的共生关系，如不良的土壤通气环境抑制水稻根系和AMF的磷转运蛋白，影响根系吸收磷素，降低AMF的侵染

率^[41]。Xu等^[42]认为干湿交替增加AMF对共生信号(几丁质化合物的混合物)的分泌，并激活下游共生信号通路，促进AMF定殖。干湿交替是一个循环过程，当生物炭和AMF联用后，生物炭可以通过改善土壤孔隙结构提高通气条件^[43]，这不仅能在淹水阶段减缓氧气流失，还能在落干阶段加速土壤微环境的氧气恢复，从而有利于AMF与水稻根系的共生。

由表1可知，与CK相比，不同外源添加物均提高了水稻的地上、地下生物量($P<0.05$)。与CK相比，G_{BC}和G_{AMF}使水稻地上部分生物量分别增加了58.70%和65.22%，地下部分生物量分别增加55.56%和64.81%。生物炭中的氮、磷、碳等营养物质提供了植物生长所需的养分，多孔性结构改善了土壤孔隙度、容重、持水率等，这些均有利于植物生长^[38]。而AMF与水稻共生，不仅可扩大根系的养分吸收面积^[42]，还可提高植物光合作用能力^[44]，促进植物生长。G_{BA}的水稻具有最高的生物量和根冠比。G_{BA}的地上部分生物量为1.68 g·m⁻²，较CK、G_{BC}和G_{AMF}分别增加82.61%、15.07%和10.53%。G_{BA}地下部分生物量为1.22 g·m⁻²，较CK、G_{BC}和G_{AMF}分别增加125.93%、45.24%和37.08%。根冠比是反映植物光合碳分配的重要特征^[45]，G_{BA}的根冠比相较于CK、G_{BC}和G_{AMF}分别增加24.14%、26.32%和24.14%。生物炭和AMF联用对水稻促生的原因：首先是植物根系较粗难以进入生物炭微孔中，AMF菌丝纤细可以进入生物炭内部，吸收生物炭内部暂未释放的营养物质，同时生物炭可以调节植物-真菌信号，及时将营养反馈给植物，提高养分利用率^[46]。其次是生物炭和AMF联用能促进固氮和铁载体的产生，提高植物所需营养物质的可用性^[47]。

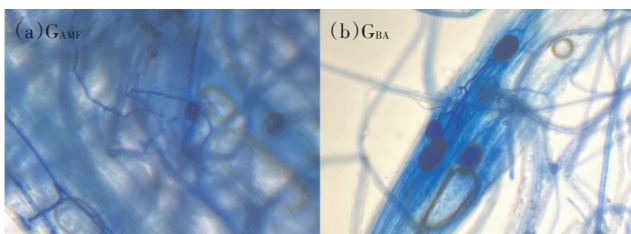


图1 培养90 d后单独接种丛枝菌根真菌(G_{AMF})和生物炭-丛枝菌根联用(G_{BA})的水稻根系侵染情况(显微镜放大40倍)

Figure 1 Root infection situation of single inoculation arbuscular mycorrhizal fungi(G_{AMF}) and biochar-arbuscular mycorrhizal fungi combination(G_{BA}) after 90 days of cultivation(Microscope magnified 40 times)

2.2 生物炭与丛枝菌根真菌对SOC及其组分的影响

2.2.1 SOC

培养90 d后不同处理SOC含量如表1所示。CK

的SOC为 $2.60 \text{ g} \cdot \text{kg}^{-1}$,单独接种AMF未显著提高SOC,但单独添加生物炭或生物炭与AMF联用均能显著提高SOC。与CK相比, G_{BC} 和 G_{BA} 的SOC分别增加了212.69%和504.23% ($P < 0.05$)。 G_{BA} 对SOC的提升最显著,较 G_{BC} 和 G_{AMF} 分别增加了93.23%和423.67%。生物炭可能通过自身较高的含碳量^[48],以及抑制有机物分解等提高SOC^[49]。AMF可能通过分泌GRSPs,促进土壤团聚体形成,通过物理包裹或化学键合对SOC形成保护^[15]。二者联用后,生物炭可以提高水稻根系中AMF侵染率,这不仅增强了植物的光合碳固定^[44],更促进了GRSPs分泌,进而促进土壤团聚^[50],所以二者联用更能提高SOC的含量和稳定性。

2.2.2 POC和MAOC

土壤有机碳库的稳定性不仅取决于SOC总量,更与其组成密切相关。通常将SOC分为POC和MAOC,其中,POC主要由动植物残体形成,是MAOC形成的前体物,MAOC可以由土壤矿物质与POC分解产物(小分子有机物)通过配体交换或范德华力形成^[51-52],是较稳定的有机碳组分,对SOC的长期固存至关重要。本实验使用分馏法将SOC分为POC和MAOC,进一步研究生物炭和AMF单独施用或联用对SOC组成及稳定性的影响。经过90 d培养,各处理的POC和MAOC变化显著(表2)。和CK相比, G_{BC} 的POC增加19.1%。其原因一方面可能是生物炭颗粒中大于0.053 mm的组分在较短时间内不易分解,直接贡献POC^[53]。另一方面可能是生物炭添加后其养分缓慢释放,促进植物生长,增加根系分泌物和植物残体输入,进一步增加了土壤POC含量^[54]。而 G_{AMF} 和 G_{BA} 的POC较CK分别降低了63.06%和22.29%,因为AMF自身不直接提供碳源,但其定殖于水稻根系后(实验测定时根系已移除),可能通过增强微生物活性加速动植物残体的分解,从而减少POC累积。 G_{BA} 的降幅低于 G_{AMF} ,可能是生物炭的物理保护作用部分抵消了AMF的分解效应。与CK($1.03 \text{ g} \cdot \text{kg}^{-1}$)相比, G_{BC} 的MAOC增加了

507.77%,这可能是生物炭中的复杂碳结构(如芳香族碳)更易于与矿物质结合形成MAOC,从而增强了SOC的稳定性^[55]。 G_{BA} 的MAOC较CK增加了13.07倍,可能原因是生物炭除了自身能提供MAOC外,还刺激AMF分泌更多的GRSPs,二者共同促进矿物和有机质的胶结作用,提高MAOC含量^[56]。 G_{BC} 、 G_{AMF} 和 G_{BA} 的MAOC/SOC较CK分别提高了95.85%、102.14%和134.53%,POC/SOC较CK分别降低了62.06%、66.13%和87.10%,这说明生物炭和AMF单独施加或联用均能提高SOC中稳定态碳组分,促进POC(不稳定态碳)向MAOC(稳定态碳)转化,提高SOC的稳定性,这与Sun等^[57]的研究结果一致。以上结果表明,联用对SOC总量和碳稳定性的增加效果显著高于单一生物炭和AMF处理。

2.3 生物炭与丛枝菌根真菌对土壤含碳官能团的影响

利用FTIR探究添加生物炭、接种AMF和二者联用对含碳官能团变化(图2),揭示碳结构对有机碳稳定性的影响。FTIR图谱分析表明,不同处理的SOC均含有脂肪族碳甲基C—H的弯曲振动(1362 cm^{-1})、芳香族碳C=C的伸缩振动(1603 cm^{-1})和脂肪族碳甲基/亚甲基C—H的伸缩振动(2832 cm^{-1})^[58-61]。但生物炭或AMF单独或联合添加对土壤含碳官能团的相对含量产生显著影响。 G_{BC} 、 G_{AMF} 和 G_{BA} 的芳香族碳的相对含量较CK(58.10%)分别增加13.80%、12.74%和30.57%; G_{BC} 、 G_{AMF} 和 G_{BA} 的甲基/亚甲基C—H伸缩振动的脂肪族碳的相对含量较CK(10.03%)分别减少70.79%、115.36%和104.06%,甲基C—H的弯曲振动的脂肪族碳的相对含量较CK(31.87%)分别减少2.89%、12.61%和34.01%。3个处理均提高了土壤中芳香族碳的相对含量,降低了脂肪族碳的相对含量,且 G_{BA} 的芳香族碳的相对含量最高(75.86%)。原因可能是:首先,生物炭制备的热解过程提高了材料难降解芳香族碳含量^[62];其次,AMF分泌的GRSPs中含高达49%的芳香烃^[63],可进一步贡献芳香族碳;最后,

表2 不同处理对土壤颗粒态有机碳和矿物结合态有机碳含量的影响及颗粒态有机碳和矿物结合态有机碳占有有机碳的比值

Table 2 Effects of different treatments on soil POC and MAOC content, POC/SOC and MAOC/SOC

处理 Treatment	颗粒态有机碳 POC/($\text{g} \cdot \text{kg}^{-1}$)	矿物结合态有机碳 MAOC/($\text{g} \cdot \text{kg}^{-1}$)	颗粒态有机碳占有有机碳的比值 (POC/SOC)/%	矿物结合态有机碳占有有机碳的比值 (MAOC/SOC)/%
CK	1.57±0.07ab	1.03±0.20d	60.70±3.88a	39.30±3.88c
G_{BC}	1.87±0.24a	6.26±0.18b	23.03±2.59b	76.97±2.59b
G_{AMF}	0.57±0.16c	2.42±0.76c	20.56±9.87b	79.44±9.87b
G_{BA}	1.22±0.31b	14.49±0.73a	7.83±2.18c	92.17±0.02a

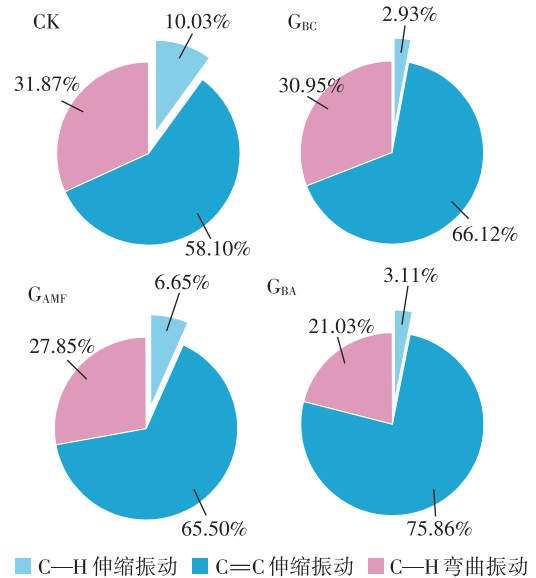
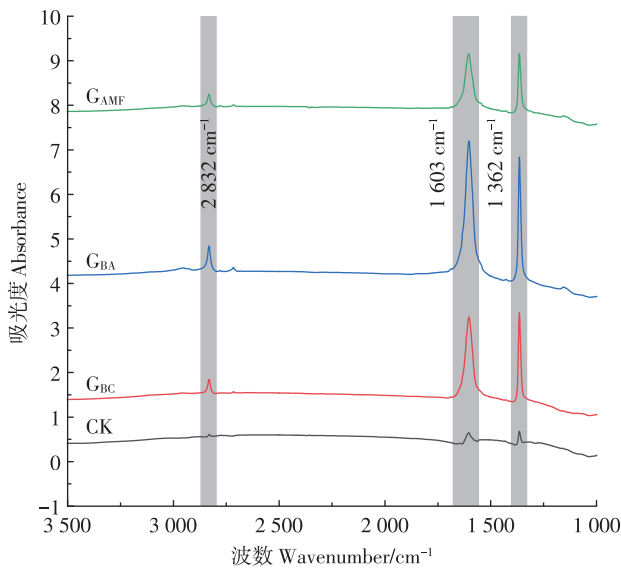


图2 不同处理后土壤的傅里叶红外光谱图

Figure 2 Fourier transform infrared spectra of soil after different treatments

生物炭还能促进革兰氏阳性菌优先利用脂肪族碳^[64], 加速易分解碳的消耗, 间接提高芳香族碳的相对含量。芳香族碳是土壤中的难降解成分, 其含量越高有机碳越稳定, 而脂肪族碳较芳香族碳稳定性较差^[65], 因而生物炭和AMF联用提高了芳香族碳的相对含量, 从而进一步稳定了土壤碳库。已有研究表明芳香族碳更优先与铁铝氧化物吸附结合形成MAOC^[66], 芳香族碳的富集通过矿物结合途径实现长期碳封存。综上, 本实验中芳香族碳的相对含量增加可能是造成MAOC积累的一个原因。

2.4 生物炭与丛枝菌根真菌对土壤碳铁结合特征及铁结合有机碳的影响

铁氧化物是形成MAOC的主要胶结矿物^[67], 本实验通过分析不同处理下氧化铁的形态变化, 揭示生物炭和AMF对土壤铁碳结合固存机制的调控作用(表3)。培养90 d后, 与CK相比, G_{BC}、G_{AMF}和G_{BA}的Fe_d分别降低了0.91%、3.20%和4.00%, Fe_o分别提高了8.11%、5.41%和2.70%, Fe_p分别增加了4.88%、4.88%

和14.63%。生物炭添加和AMF接种均降低了Fe_d, 但提高了土壤Fe_o和Fe_p。He等^[68]认为生物炭施加可提高土壤pH值, 诱导Fe_d转化为非结晶氧化铁(Fe_o和Fe_p), 同时生物炭的高比表面积和孔结构可增强对铁氧化物的吸附^[69], 抑制Fe_d结晶并促进Fe_o生成。AMF产生的有机酸通过配体交换结合针铁矿^[70], 这可能抑制氧化铁的结晶^[71], 导致Fe_d降低。干湿交替的土壤环境通过氧化还原环境循环促进土壤铁氧化物的溶解-重结晶过程, 进一步增加Fe_o, Fe_o较高的比表面积和吸附亲和力比晶型氧化铁更能吸附固定SOC^[72]。

Fe-OC是形成MAOC的关键组分, 其含量可进一步证明本研究铁氧化物与SOC的结合^[73], 从而促进有机碳在土壤中的保留^[74]。如图3a所示, 与CK相比, G_{BC}、G_{AMF}和G_{BA}的Fe-OC含量分别增加了18.53%、167.12%和1397.51%, 铁络合态指数(图3c)分别增加了5.48%、8.68%和18.20%。各处理土壤Fe-OC占SOC的比例见图3b, 其中G_{BA}的Fe-OC占SOC的比例最高, 约为31.22%, 较CK和G_{BC}分别提高了204.98%

表3 不同处理下的土壤铁含量及铁氧化物形态变化

Table 3 Changes in soil iron content and iron oxide forms under different treatments

处理 Treatment	总铁 Fe/(g·kg ⁻¹)	游离态氧化铁 Fe _d /(g·kg ⁻¹)	无定形氧化铁 Fe _o /(g·kg ⁻¹)	络合态氧化铁 Fe _p /(g·kg ⁻¹)	结合态有机碳的碳铁 摩尔比OC/Fe
CK	16.42±0.07ab	8.75±0.03a	2.96±0.01a	0.41±0.01b	0.14±0.08d
G _{BC}	16.43±0.02ab	8.67±0.02b	3.20±0.01a	0.43±0.02b	0.21±0.03c
G _{AMF}	16.33±0.08b	8.47±0.02c	3.12±0.02b	0.43±0.01b	0.39±0.08b
G _{BA}	16.47±0.04a	8.40±0.05d	3.04±0.05c	0.47±0.01a	2.72±0.32a

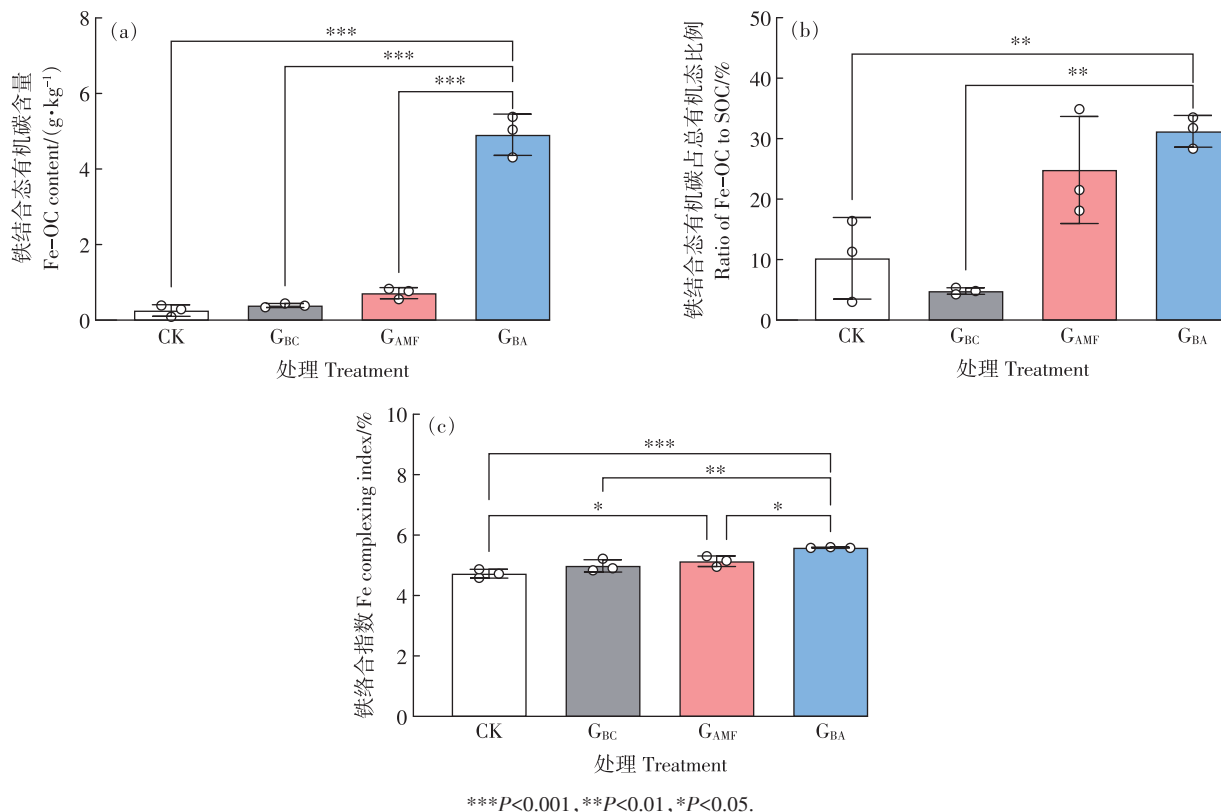


图3 不同处理对土壤铁结合态有机碳含量、铁结合态有机碳占总有机碳的比值及铁的络合指数的影响

Figure 3 Effects of different treatments on soil Fe-OC content, the ratio of Fe-OC to SOC and Fe complexing index

和550.03%,说明铁氧化物和有机碳结合是生物炭和AMF联用下SOC固存的主要结合形式^[75]。相关研究表明,添加生物炭、接种AMF和二者联用均可增强铁氧化物与有机配体(如有机酸、氨基酸等)间的相互作用,促进形成稳定的矿物-有机质复合物^[76]。AMF通过菌根共生分泌的草酸、柠檬酸等有机酸,与铁氧化物表面羧基/羟基发生配位反应,推动矿物-有机质复合物形成^[77]。而生物炭诱导Fe_o与芳香族碳结合通过配位键形成芳烃-Fe络合物,其疏水性和化学惰性可增强团聚体内的保留,增加Fe-OC的含量^[78-79]。OC/Fe可作为氧化铁与SOC形成Fe-OC的途径的判断指标(OC/Fe<1为吸附,OC/Fe>6为共沉淀,OC/Fe介于1~6则为双重作用)^[80]。培养90 d后,G_{BC}、G_{AMF}和G_{BA}的OC/Fe分别为0.21、0.39和2.72,表明在干湿交替条件下种植水稻土壤中单独添加生物炭或AMF主要通过铁氧化物吸附有机碳形成Fe-OC,而二者联用时吸附和共沉淀共同促进Fe-OC的形成。

通过XPS的Fe 2p精细谱图分析进一步揭示各处理的铁氧化还原形态的分布特征(图4及表4),分析SOC稳定化的作用机理。各处理的Fe²⁺和Fe³⁺在Fe 2p_{3/2}的结合能范围分别为710.94~711.27 eV和

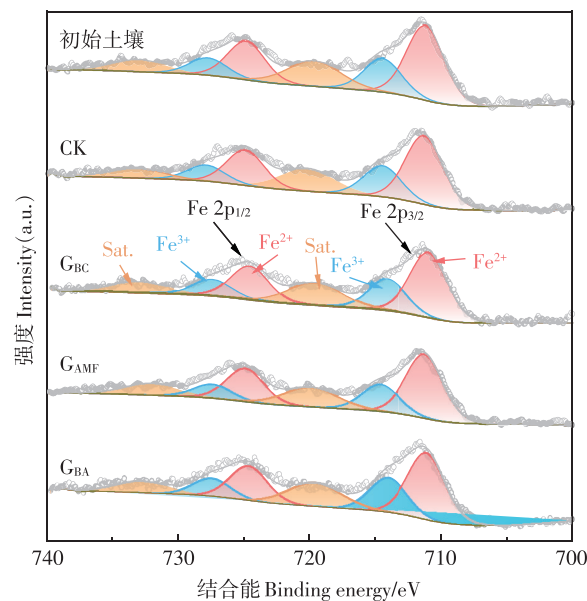


图4 不同处理下土壤Fe 2p XPS光谱图

Figure 4 Soil Fe 2p XPS spectra under different treatments

714.01~714.46 eV,在Fe 2p_{1/2}的结合能范围分别为724.57~724.91 eV和727.38~727.861 eV^[81-82]。初始土壤中Fe²⁺的相对含量为60.60%,Fe³⁺的相对含量为25.76%。90 d培养后,相较于初始土壤,CK中Fe²⁺的

表4 不同处理下土壤 Fe 2p XPS主要吸收峰的
相对强度及其比例

Table 4 Relative intensity and ratio of the main absorption peaks
of soil Fe 2p XPS under different treatments

处理 Treatment	Fe ²⁺ /%	Fe ³⁺ /%	Fe ²⁺ /Fe ³⁺
初始土壤	60.60	25.76	2.35
CK	59.76	27.11	2.20
G _{BC}	57.98	28.38	2.04
G _{AMF}	59.56	26.85	2.22
G _{BA}	56.55	29.61	1.91

相对含量降低 1.39%, Fe³⁺的相对含量增加 5.24%, 说明水稻通过根系泌氧一定程度上促进了 Fe²⁺氧化^[83], Fe³⁺较 Fe²⁺具有更高的电荷密度, 更易与有机配体结合, 这种结合也是 MAOC 形成的一个重要原因^[84]。所有处理组中 Fe²⁺含量均显著高于 Fe³⁺ (Fe²⁺/Fe³⁺>1), 表明此时土壤整体处于还原环境^[24]。对于 Fe²⁺而言, 相较于 CK, G_{BC}、G_{AMF} 和 G_{BA} 中 Fe²⁺的相对含量分别降低 2.98%、0.33% 和 5.37%, G_{BC} 和 G_{BA} 中 Fe³⁺的相对含量分别增加了 4.68% 和 9.22%。生物炭的多孔结构改善了土壤通气性, 且表面官能团可作为电子穿梭体, 促进 Fe²⁺的氧化。不仅如此, G_{BA} 的 Fe³⁺含量最高 (29.61%), Fe²⁺/Fe³⁺最低 (1.91), 根际土壤中约 66% 的 Fe³⁺为无定形态^[85], 这可能证实了 Fe_o的增长。单独添加生物炭以及生物炭与 AMF 联用可能促进了更多的 Fe²⁺被氧化为 Fe³⁺, 而单独接种 AMF 可能对土壤氧化还原影响不大。

2.5 土壤铁氧化物、SOC 及其组分的关系分析

生物炭的多孔隙结构及表面官能团为 AMF 菌丝扩展提供了空间和养分, 促进了 AMF 在水稻根部的高效定殖^[86]; 高侵染率驱动 AMF 分泌更多的 GRSPs, 其胶结作用可促进土壤团聚, 提高对 SOC 的物理保护。生物炭表面和 GRSPs 中所含有的芳香碳与 Fe_o 发生配位络合, 显著提升 Fe-OC 含量^[6], 进而提高 SOC 总量及稳定性。MAOC 是 SOC 的总量和稳定性的重要组成部分。Zhang 等^[87]发现外源碳输入显著提升土壤 MAOC, 且 MAOC 占总 SOC 的 60%~90%, 这与本实验的结果相似。如图 5 所示, SOC 与 MAOC 显著正相关 (P<0.001), MAOC/SOC 高达 92.17% (表 2), 说明 SOC 的赋存形态主要为稳定矿物结合态有机碳, 联用处理提升了 SOC 的稳定性。已有研究表明, Fe-OC 对 SOC 有着 8.33%~41.9% 的贡献率^[88]。与其他处理相比, G_{BA} 处理的 Fe-OC 占 SOC 的比例最高, 为 31.22% (图 3b), 说明铁氧化物与有机碳的结合是联用处理

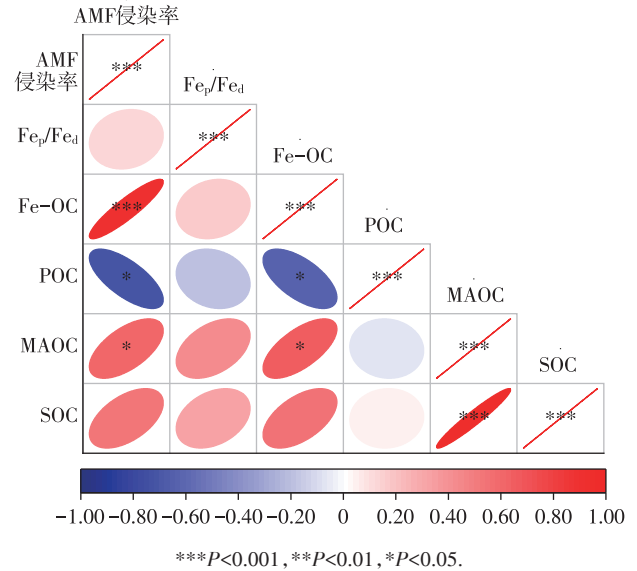


图5 有机碳及其组分和 AMF 侵染率的 spearman 相关性分析
Figure 5 Spearman correlation analysis of organic carbon, its
components and AMF colonization rate

下 SOC 稳定的主导途径。AMF 侵染率与 MAOC (P<0.05) 和 Fe-OC (P<0.001) 均表现为显著正相关, 说明 AMF 的繁殖、代谢可促进 MAOC 和 Fe-OC 的积累^[89], 是 SOC 固存的驱动因子之一。

3 结论

(1) 单独施用生物炭、接种丛枝菌根真菌或二者联用均显著增加了稻田土壤有机碳、矿物结合态有机碳和铁结合态有机碳的含量, 且二者联用处理的土壤有机碳稳定性最高, 增加矿物结合态有机碳是二者联用对土壤固碳的主导途径。

(2) 在干湿交替的稻田土壤中, 单独施用生物炭或丛枝菌根真菌的主要固碳途径为铁氧化物对有机碳的吸附作用, 而二者联用时主要固碳途径为吸附与共沉淀协同促进铁结合有机碳的生成, 表明铁元素是稻田多维土壤固碳的关键调控因子。

(3) 未来研究将结合团聚体分级与矿物表征进一步解析生物炭和丛枝菌根真菌协同固碳的作用过程和固碳机理。

致谢: 感谢江苏大学环境生态研究所戴志聪教授和祁珊珊教授提供的实验用 AMF 菌剂。

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