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目 次

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菌渣与化肥配施对稻田土壤微生物群落组成及多样性的影响

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摘要: 菌渣是一种独特而丰富的有机物料,与化肥配施不仅能改良土壤质量还可以调控微生物群落. 然而,土壤细菌和真菌对菌渣与化肥配施的响应是否一致仍不清楚. 在水稻田间长期定位试验条件下,设置化肥水平(C)0%、50% 和 100%,菌渣相对用量(F)0%、50% 和 100% 各 3 个水平,共 9 个处理,对土壤肥力与微生物群落的相关指标进行测定. 结果表明,土壤全氮(TN)在 C_0F_{100} 处理中最高,碳氮比(C/N)、全磷(TP)、可溶性碳(DOC)和有效磷(AP)在 $C_{100}F_{100}$ 处理中最高,土壤有机碳(SOC)、碱解氮(AN)、速效钾(AK)和 pH 在 $C_{50}F_{100}$ 处理中最高,较对照分别增加了 55.56%、26.18%、26.46%、17.13%、279.54%、85.57%、41.61%、29.33% 和 4.62%. 菌渣与化肥配施后,不同处理土壤细菌和真菌 α-多样性存在显著变化,与对照 C_0F_0 处理相比较,各处理细菌 β-多样性并未发生显著变化,却使真菌 β-多样性发生了明显差异,且 $C_{50}F_{100}$ 处理显著降低了真菌子囊菌门(Ascomycota)和粪壳菌纲(Sordariomycetes)的相对丰度. 随机森林预测模型表明,AP 和 C/N 分别是细菌和真菌 α-多样性的主要驱动因子, AN、pH、SOC 和 DOC 是细菌 β-多样性的主要驱动因子,而 AP 和 DOC 是真菌 β-多样性的主要驱动因子. 相关性分析表明,真菌子囊菌门(Ascomycota)和粪壳菌纲(Sordariomycetes)与 SOC、TN、TP、AN、AP、AK 和 C/N 显著负相关. 置换多元方差分析表明,菌渣对土壤肥力指标、细菌门及纲水平上优势物种、真菌门及纲水平优势物种、细菌多样性和真菌多样性的变化贡献率分别为 46.35%、18.47%、41.57%、23.84% 和 10.42%,菌渣和化肥的交互效应对细菌和真菌多样性变化的贡献率分别为 9.90% 和 35.00%. 综上所述,施用菌渣较化肥在影响土壤肥力指标含量和微生物群落变化方面更具优势.

关键词: 菌渣; 化肥; 稻田土壤; 细菌多样性; 真菌多样性; 微生物群落
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Effects of Combined Application of Fungal Residue and Chemical Fertilizer on Soil Microbial Community Composition and Diversity in Paddy Soil

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Abstract: Fungal residue is a unique abundant organic material undervalued in agricultural production. The application of chemical fertilizer combined with fungal residue can not only improve soil quality but also regulate the microbial community. However, it is unclear whether the response of soil bacteria and fungi to the combined application of fungal residue and chemical fertilizer is consistent. Therefore, a long-term positioning experiment in a rice field was conducted with a total of nine treatments. Chemical fertilizer (C) and fungal residue (F) were applied at 0, 50%, and 100% to evaluate ① the change in soil fertility properties and microbial community structure and ② the main driving factors of soil microbial diversity and species composition. The results showed that soil total nitrogen (TN) was highest after treatment C_0F_{100} (55.56% higher than in the control), and the carbon to nitrogen ratio (C/N), total phosphorus (TP), dissolved organic carbon (DOC), and available phosphorus (AP) contents were highest after treatment with $C_{100}F_{100}$ (26.18%, 26.46%, 17.13%, and 279.54% higher than in the control, respectively). The amounts of soil organic carbon (SOC), available nitrogen (AN), available potassium (AK), and pH were highest after treatment with $C_{50}F_{100}$ (85.57%, 41.61%, 29.33%, and 4.62% higher than in the control, respectively). Following the application of fungal residue with chemical fertilizer, there were significant changes in the α-diversity of bacteria and fungi in each treatment. Compared with that of the control (C_0F_0), different long-term applications of fungal residue with chemical fertilizer did not significantly change soil bacterial β-diversity but resulted in significant differences in fungal β-diversity, and the relative abundance of soil fungal Ascomycota and Sordariomycetes significantly decreased after the application of $C_{50}F_{100}$. The random forest prediction model indicated that AP and C/N were the main driving factors of bacterial and fungal α-diversity, respectively, and AN, pH, SOC, and DOC were the main driving factors of bacterial β-diversity, whereas AP and DOC were the main driving factors of fungal β-diversity. Correlation analysis suggested that the relative abundance of soil fungal Ascomycota and Sordariomycetes had a significantly negative correlation with SOC, TN, TP, AN, AP, AK, and C/N. PERMANOVA showed that variation in soil fertility properties, dominant species of soil bacteria at the phylum and class level, and dominant species of soil fungi at the phylum and class level were all best explained by fungal residue (46.35%, 18.47%, and 41.57%, respectively), and variation in bacterial diversity was best explained by fungal residue (23.84%) and to a lesser extent by the interaction between fungal residue and chemical fertilizer (9.90%). In contrast, the variation in fungal diversity was best explained by the interaction between fungal residue and chemical fertilizer (35.00%) and to a lesser extent by fungal residue (10.42%). In conclusion, the application of fungal residue has more

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advantages than chemical fertilizer in influencing soil fertility properties and microbial community structure changes.
Key words: fungal residue; chemical fertilizer; paddy soil; bacterial diversity; fungal diversity; microbial community

土壤微生物是土壤的重要组成部分,是维持土壤健康的关键[1,2]. 土壤微生物群落可能对植物生长、凋落物分解、驱动养分循环和碳、氮周转起着关键的作用[3~6]. Wei 等[7]报道了土壤微生物群落的组成和功能决定了植物是否可以抵抗土传病害. 有研究表明,微生物多样性可以显著影响土壤中碳的周转[8~10],Chen 等[11]揭示了微生物 Shannon 多样性指数可以作为植被变化下 SOM 动态的可测量指标,Xu 等[12]的研究发现在底土中,当土壤细菌和真菌多样性较高时,SOC 分解表现出更强的抗变暖能力. 因此,土壤微生物是土壤质量变化的衡量指标.

由于土壤微生物对环境的敏感性[13,14],长期施肥等农艺活动会导致土壤环境因子发生变化,从而影响微生物群落[15,16]. 有研究表明,施氮改变了土壤微生物群落组成,使细菌群落从以寡养菌群为主转变为以共养菌群为主[17,18],Dai 等[19]的研究发现施氮会降低细菌多样性,对于真菌,施氮会降低担子菌(Basidiomycota)的相对丰度[20]. 土壤微生物群落对有机肥施用也极为敏感[21],有机物料和有机肥可以缓解化肥对微生物多样性的负面影响[22,23]. Liu 等[24]报道了与单施化肥相比,有机替代增加了微生物多样性. 马龙等[25]的研究发现有机肥替代部分化肥可以提高土壤细菌和真菌丰度,改变土壤优势菌群的组成. 因此,探究有机物料与化肥配施对土壤肥力指标含量和微生物群落的影响,对正确揭示土壤肥力指标含量与微生物群落之间的关系具有重要意义.

菌渣是一种独特而丰富的有机物料,是农业生产中被低估的生物质资源[26],将其还田能为土壤提供大量的营养物质. 我国是世界上最大的食用菌生产国,但菌渣的利用率仅为 33%[27],不恰当处置可能会造成生态环境污染[28]. 以往对菌渣还田的研究主要集中在土壤养分、土壤酶和作物产量变化方面[29~31]. 菌渣还田后土壤微生物群落可能会发生变化,然而,菌渣还田后对微生物群落的影响目前尚未报道. 为此,在水稻田间长期定位试验条件下,设置不同比例的菌渣与化肥配施处理,利用 Illumina MiSeq 高通量测序技术测定细菌和真菌群落,通过置换多元方差分析和随机森林预测模型,定量评估菌渣对土壤肥力指标和微生物群落的影响,及其微生物多样性和物种组成的主要驱动因子.

1　材料与方法

1.1　试验场地

本试验于 2017 年 6 月至 2017 年 12 月在中国浙江省嘉兴市水稻田(北纬 30°37′~30°40′,东经 120°39′~120°44′,平均海拔为 4 m)进行. 该地区四季分明,属东亚季风区,年平均降水量为1 168.6 mm,主要集中在 5~9 月,年平均气温为 15.9℃,无霜期为 230 d,年平均日照为2 017 h. 长期定位试验始于 2010 年,供试土壤试验前的基本理化性质为:pH 值 6.80,ω[有机碳(SOC)]25.16 g·kg^{-1},ω[碱解氮(AN)]94.29 mg·kg^{-1},ω[有效磷(AP)]37.01 mg·kg^{-1},ω[速效钾(AK)]127.22 mg·kg^{-1}. 土壤耕作方式是休耕轮作. 菌渣选用种植黑木耳后的桑枝屑物料经发酵后的产物. 主要养分为:ω(SOC)451.8 g·kg^{-1},ω[全氮(TN)]11.4 g·kg^{-1},ω[全磷(TP)]1.0 g·kg^{-1},ω[全钾(TK)]6.0 g·kg^{-1},碳氮比(C/N)为 39.6. 成分由纤维素(28.21%)、半纤维素(20.16%)、木质素(15.11%)、灰分(14.83%)和蛋白质(14.3%)组成. 供试水稻品种为甬优 1540. 2017 年 6 月 15 日播种育苗,7 月 13 日插秧移栽秧苗,12 月 13 日收获.

1.2　试验设计

本试验设置施用化肥 C_0、C_{50}和 C_{100},施用菌渣 F_0、F_{50}和 F_{100},采用双因素随机区组排列,共设 9 个处理,每个处理 3 次重复,共 27 个小区,每个小区面积为 20 m^2. 菌渣施用量为 0(0%)、10 t·hm^{-2}(50%)和 20 t·hm^{-2}(100%),化肥施用量为当地常规施肥量的 0%、50%和 100%. 各处理具体如下:不施化肥和菌渣(C_0F_0)为对照,施用菌渣 10 t·hm^{-2}(C_0F_{50}),施用菌渣 20 t·hm^{-2}(C_0F_{100}),50%的化肥用量($C_{50}F_0$),50%的化肥用量配施菌渣 10 t·hm^{-2}($C_{50}F_{50}$),50%的化肥用量配施菌渣 20 t·hm^{-2}($C_{50}F_{100}$),100%的化肥用量($C_{100}F_0$),100%的化肥用量配施菌渣 10 t·hm^{-2}($C_{100}F_{50}$),100%的化肥用量配施菌渣 20 t·hm^{-2}($C_{100}F_{100}$). 结合当地单季稻种植制度,水稻移栽前将土壤进行翻耕,同时施入不同比例混合的菌渣和化肥(基肥). 常规化肥 100%施用量具体如下:基肥施碳酸氢铵(含氮量 17%)300 kg·hm^{-2};分蘖初期追施尿素(含氮量 46%)150 kg·hm^{-2};分蘖盛期追施控释复合肥(N∶P∶K = 22∶11∶13)225 kg·hm^{-2},50%化肥处理则将 100%化肥处理减半进行,0%化肥处理不施肥,其他田间管理

策略遵循传统栽培技术.

1.3 土壤采样与分析

土壤样品于水稻收获后,从每个小区 0 ~ 20 cm 土层多点采集土壤,清除石块和植物残留物,彻底混合取样土壤,通过 2 mm 筛网进行筛分,并分为 3 部分. 第一部分用于土壤可溶性碳(DOC)分析,第二部分存于 -84℃用于土壤 DNA 提取,第三部分通风阴干用于土壤理化性质分析.

1.4 测定方法

SOC、TP、TN、pH、AN、AP 和 AK 采用常规土壤农化分析方法测定, DOC 按水土比为 2∶1 比例浸提.

土壤 DNA 提取及测序:使用 DNA 试剂盒(MP Biomedicals, Santa Ana, CA, USA)从 1.0 g 新鲜土壤中提取 DNA 总量. 为了生成细菌和真菌高通量测序的扩增子库,细菌 16S rRNA 基因用引物 F515(5′-GTGCCAGCMGCCGCGGTAA-3′)和 R806(5′-GGACTACHVGGGTWTCTAAT-3′)在 V4 区进行 PCR 扩增,真菌 ITS 基因用引物 ITS1F(5′- CTTGGTCATTTAGAGGAAGTAA-3′)和 ITS2(5′-GCTGCGTTCTTCATCGATGC-3′)在 ITS1 区进行 PCR 扩增. PCR 扩增完成后,使用 QIAquick PCR 纯化试剂盒(OMEGA, USA)对产物进行纯化,用 Qubit® 2.0 荧光仪(Invitrogen, USA)进行定量. 然后将纯化后的扩增子以等量浓度汇集,用 NEB Next® UltraTM DNA 文库预备试剂盒(Annoline Biotechnology Ltd., London, UK)构建文库. 用 Agilent 2100 生物分析仪(Agilent Technologies, Inc., Santa Clara, CA, USA)检测文库的最终质量和浓度,用 KAPA 文库定量试剂盒(Kapa Bio, Wilmington, MA, USA)进行测定. 文库测序所有准备工作均在 Illumina MiSeq 平台上进行(Shanghai Majorbio Bio-Pharm Technology Co., Ltd., Shanghai, China).

1.5 统计分析

使用 SPSS 26.0(IBM, Chicago, IL, USA)对不同处理的 SOC、DOC、TN、TP、AN、AP、AK、pH 和微生物 α-多样性指数进行单因素方差分析和 Duncan 显著性检测(统计显著水平为 $P<0.05$),以比较不同处理间的差异. 利用 R4.1.2 的“vegan”包计算微生物 α-多样性指数和绘制非度量多维尺度图以辨别微生物群落的差异. 利用 R4.1.2 的“rfPermute”包预测 SOC、DOC、TN、TP、AN、AP、AK 和 pH 对细菌和真菌 α-多样性及 β-多样性的重要性,以确定细菌和真菌 α-多样性及 β-多样性的主要驱动因子. 使用 R4.1.2 的“vegan”包中的“adonis”函数执行置换多元方差(PERMANOVA)分析,定量评估菌渣和化肥对土壤肥力指标含量和微生物群落的影响. 使用 R4.1.2 的“pheatmap”包对细菌和真菌的门及纲水平上优势物种与土壤肥力指标进行相关性分析. 使用 R4.1.2 的“ggplot2”包绘制细菌和真菌的门及纲水平上物种组成堆叠图. 数据可视化在 RStudio 上操作,用 Adobde Illustrator CC 2018(Adobe Inc., San Jose, CA, USA)进行图形排版.

2 结果与分析

2.1 菌渣与化肥配施对土壤肥力指标的影响

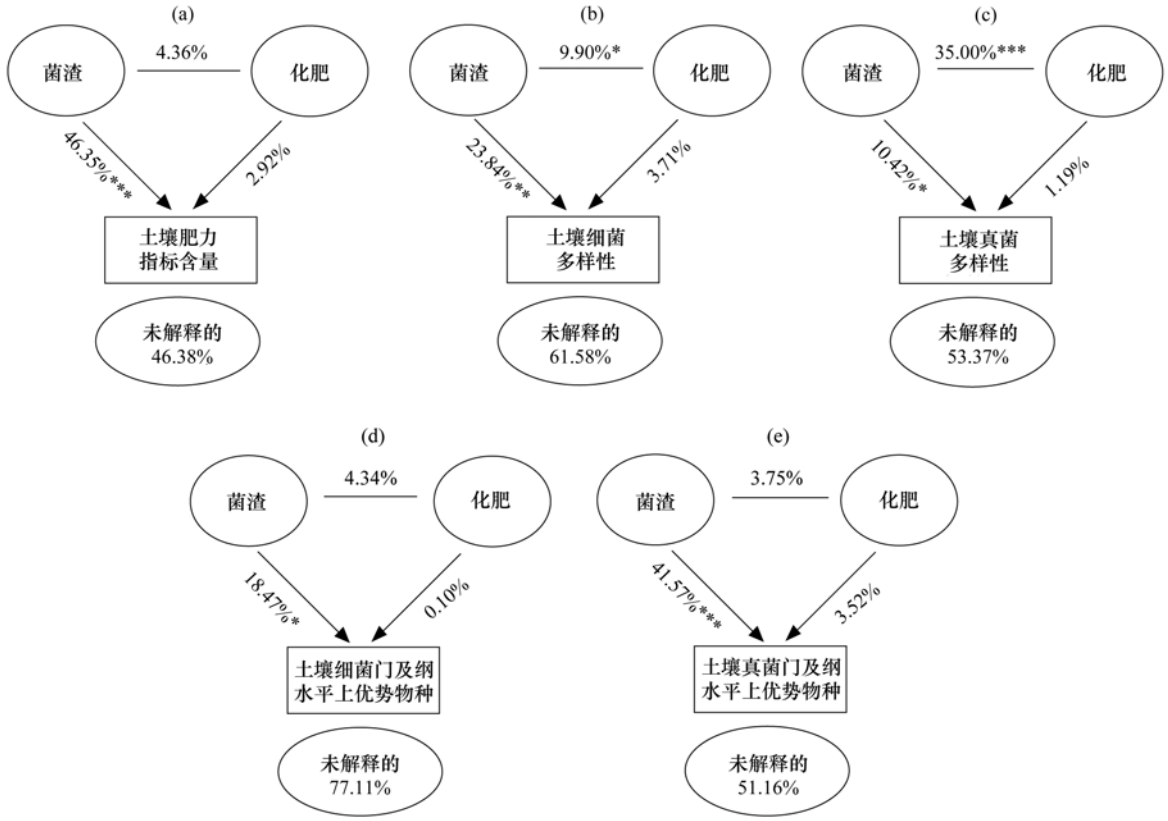
菌渣与化肥配施后,各处理土壤肥力指标均有所增加(表 1). TN 在 C_0F_{100} 处理中最高,C/N、TP、DOC 和 AP 在 $C_{100}F_{100}$ 处理中最高,pH、SOC、AN 和 AK 均在 $C_{50}F_{100}$ 处理中最高,较对照分别增加了 55.56%、26.18%、26.46%、17.13%、279.54%、4.62%、85.57%、41.61% 和 29.33%. 在化肥施用 C_0、C_{50} 和 C_{100} 水平下,各处理土壤肥力指标的含量均随菌渣施用量的增加而增加. 此外,菌渣对土壤肥力指标含量变化的贡献率为 46.35%[图 1(a)].

表 1 土壤肥力指标[1)]

Table 1 Soil fertility properties

处理	pH	ω(SOC) /g·kg^{-1}	ω(TN) /g·kg^{-1}	C/N	ω(TP) /g·kg^{-1}	ω(DOC) /mg·kg^{-1}	ω(AN) /mg·kg^{-1}	ω(AP) /mg·kg^{-1}	ω(AK) /mg·kg^{-1}
C_0F_0	6.71abc	12.68f	0.63e	20.17d	0.63b	435.65c	212.07c	13.59f	100.00e
C_0F_{50}	6.62bc	19.12c	0.84cd	22.73bc	0.71ab	445.51c	222.70c	16.89f	103.67de
C_0F_{100}	6.90ab	23.06a	0.98a	23.62ab	0.77a	480.16b	295.10a	47.85a	113.33bcd
$C_{50}F_0$	6.46c	13.60e	0.65e	20.98cd	0.70ab	487.74ab	223.53c	30.79c	100.33e
$C_{50}F_{50}$	6.64bc	14.05e	0.89bc	15.76e	0.71ab	487.80ab	266.00b	28.45cd	106.33cde
$C_{50}F_{100}$	7.02a	23.53a	0.93ab	25.41a	0.79a	493.89ab	300.30a	39.32b	129.33a
$C_{100}F_0$	6.72abc	12.44f	0.78d	16.05e	0.71ab	422.79c	216.73c	21.57e	105.33de
$C_{100}F_{50}$	6.68bc	17.92d	0.88bc	20.32cd	0.78a	393.98d	263.70b	25.70d	116.67bc
$C_{100}F_{100}$	6.89ab	21.69b	0.85bcd	25.45a	0.80a	510.28a	222.40c	51.57a	120.62ab

1)数据为平均值,同一列中不同小写字母表示不同处理在 $P<0.05$ 水平差异显著



* 为 $P<0.05$，** 为 $P<0.01$，*** 为 $P<0.001$

图 1　基于 PERMANOVA 值的菌渣和化肥对土壤肥力指标和微生物群落变化的贡献率

Fig. 1　PERMANOVA values showing the contribution rate that fungal residue and chemical fertilizer contributed to the variation in soil fertility properties and microbial community

2.2　菌渣与化肥配施对土壤微生物多样性的影响

菌渣与化肥配施后，土壤细菌和真菌多样性指数存在显著差异. 细菌 Simpson 和 Chao 指数分别在 C_0F_{100} 和 C_0F_{50} 处理中最大［图 2(a)和 2(b)］，真菌 Simpson 和 Chao 指数分别在 $C_{50}F_0$ 和 $C_{100}F_0$ 处理中最大［图 2(c)和 2(d)］，较对照（C_0F_0）分别增加了 64.01%、2.82%、85.46% 和 50.89%. 真菌 Simpson 和 Chao 指数在 $C_{50}F_{100}$ 处理中最小［图 2(c)和 2(d)］，与对照相比分别降低 90.60% 和 29.37%. 另外，各处理土壤真菌群落与对照明显分离［图 3(b)］. 菌渣对土壤细菌和真菌多样性变化的贡献率分别为 23.84% 和 10.42%［图 1(b)和 1(c)］，且菌渣和化肥的交互效应对真菌多样性变化的贡献率为 35.00%［图 1(c)］.

2.3　菌渣与化肥配施对土壤微生物物种组成的影响

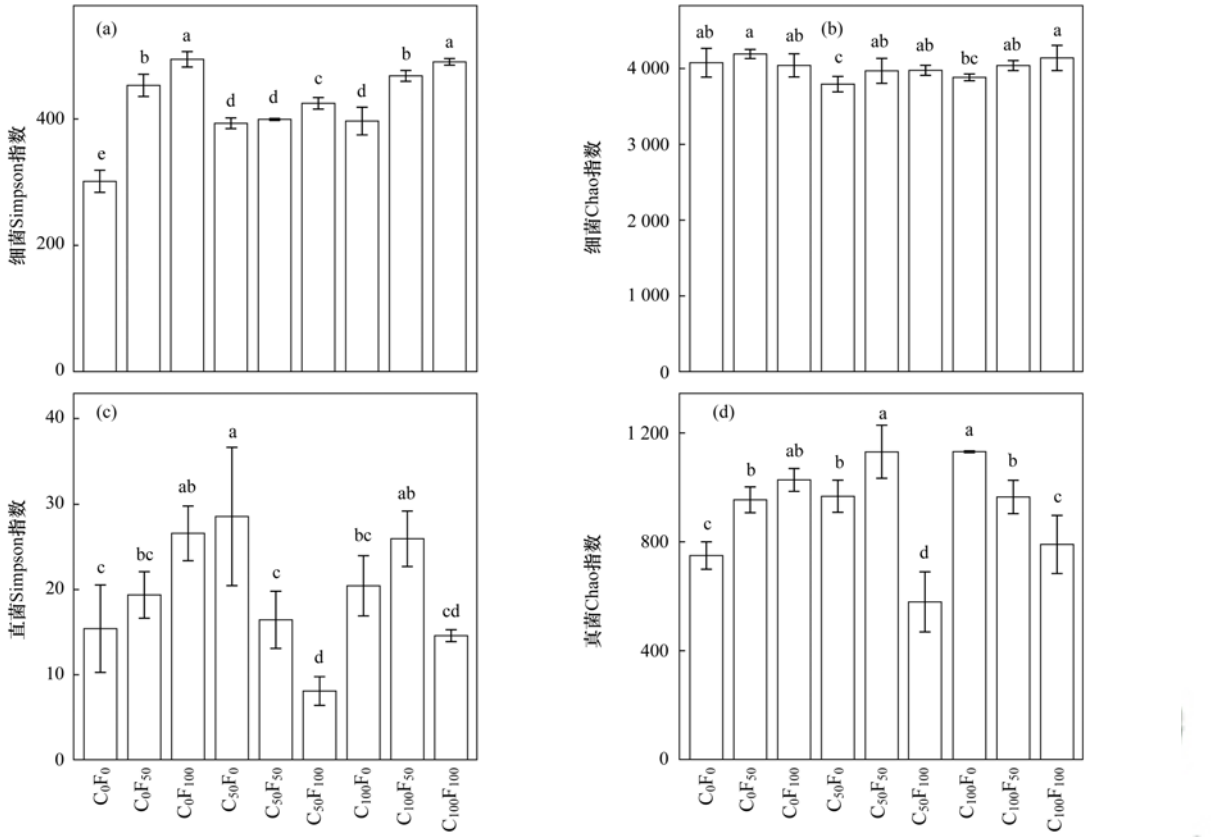
菌渣与化肥配施后，各处理土壤细菌门及纲水平上物种组成无明显变化［图 4(a)和 4(b)］. 然而，真菌门及纲水平上物种存在明显差异［图 4(c)和 4(d)］，真菌子囊菌门（Ascomycota，40.40% ~ 70.57%）和粪壳菌纲（Sordariomycetes，30.17% ~ 55.03%）分别为最优势菌门和菌纲，其相对丰度在对照 C_0F_0 处理中最高，在 $C_{50}F_{100}$ 处理中最低，且在施用化肥 C_0 和 C_{100} 水平下随着菌渣用量的增加而减小，C_{50} 水平下呈先增大后减小［图 4(c)和 4(d)］. 施用菌渣对真菌门及纲水平上优势物种变化的贡献率为 41.57%［图 1(e)］.

2.4　土壤微生物多样性与土壤肥力指标的相关性分析

SOC、AP 和 TN 对土壤细菌 α-Simpson 多样性有极显著贡献［$P<0.01$，图 5(a)］，AP 对 α-Chao 多样性有显著贡献［$P<0.05$，图 5(c)］. AP 和 SOC 对真菌 α-Chao 多样性呈显著贡献［$P<0.05$，图 5(d)］，C/N 对真菌 α-Simpson 和 Chao 多样性分别有显著［$P<0.05$，图 5(b)］和极显著贡献［$P<0.01$，图 5(d)］. AN、pH、SOC 和 DOC 对细菌 β-多样性有显著贡献［$P<0.05$，图 5(e)］，AP 和 DOC 对真菌 β-多样性有显著贡献［$P<0.05$，图 5(f)］.

2.5　土壤细菌和真菌的优势物种与土壤肥力指标的相关性分析

土壤细菌变形菌门（Proteobacteria）和 δ-变形菌纲（δ-Proteobacteria）与 DOC、TN、TP 和 AP 显著负相关［图 6(a)］，绿弯菌门（Chloroflexi）和厌氧绳菌纲（Anaerolineae）与 SOC、DOC、TN、TP、AN、AP 和 AK 显著正相关［图 6(a)］. 真菌子囊菌门



不同小写字母表示不同处理在 $P<0.05$ 水平显著差异

图 2 菌渣与化肥配施对土壤细菌和真菌 α-多样性的影响

Fig. 2 Effects of combination of fungal residue with chemical fertilizer on soil bacterial and fungal α-diversity

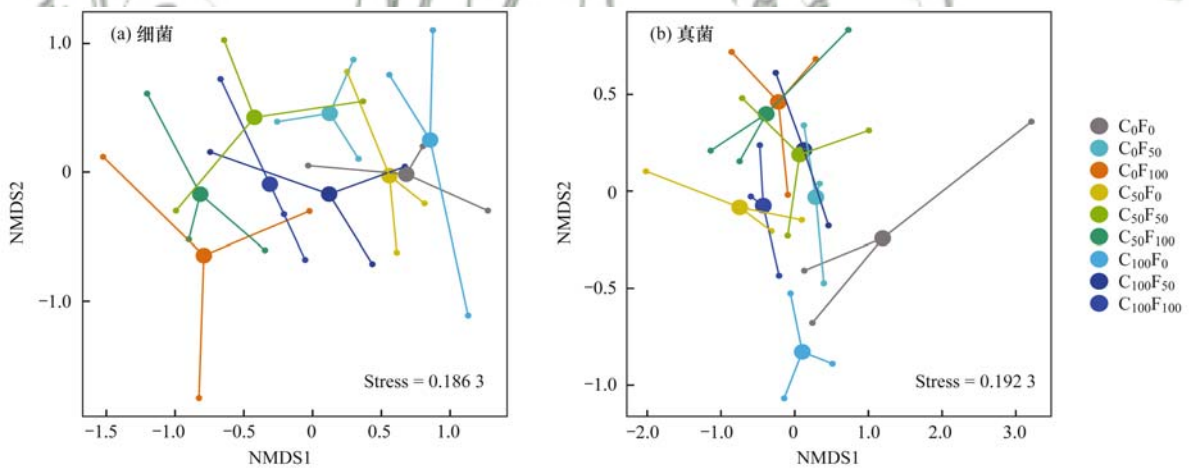


图 3 菌渣与化肥配施对细菌和真菌 β-多样性的影响

Fig. 3 Effects of combination of fungal residue with chemical fertilizer on soil bacterial and fungal β-diversity

(Ascomycota)和粪壳菌纲(Sordariomycetes)与 SOC、TN、AN、AP 和 C/N 显著负相关[图 6(b)],接合菌门(Zygomycota)、单子菌门(Basidiomycota)和接合菌纲(norank_Zygomycota)都与 SOC、DOC、AN、AP、AK 和 C/N 显著正相关[图 6(b)].

3 讨论

3.1 菌渣与化肥配施对土壤肥力指标的影响

Tarkalson 等[32]的研究发现,施用化肥是导致土壤酸化的主要因素,有研究表明土壤中添加有机物料不仅能调节 pH,还能提高土壤肥力[33]. 化肥减量配施 20 $t\cdot hm^{-2}$ 菌渣能够提高土壤 pH,说明菌渣与化肥配施可以缓解土壤酸化[34]. SOC 是维持土壤肥力和微生物群落稳定的重要角色[35],相比单施化肥,菌渣与化肥配施后 SOC 含量显著增加,这与温广婵等[36]的研究结果一致,马欣等[37]的研究也发现有机物料配施化肥对农田土壤固碳效果更好. DOC 是表征土壤活性碳库的重要指标,虽然其

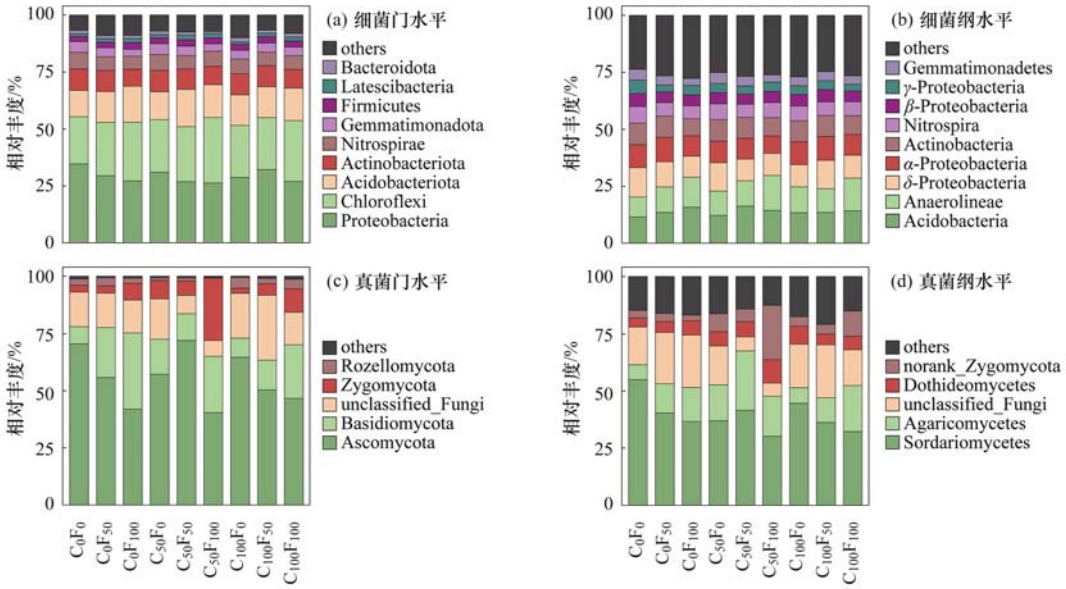
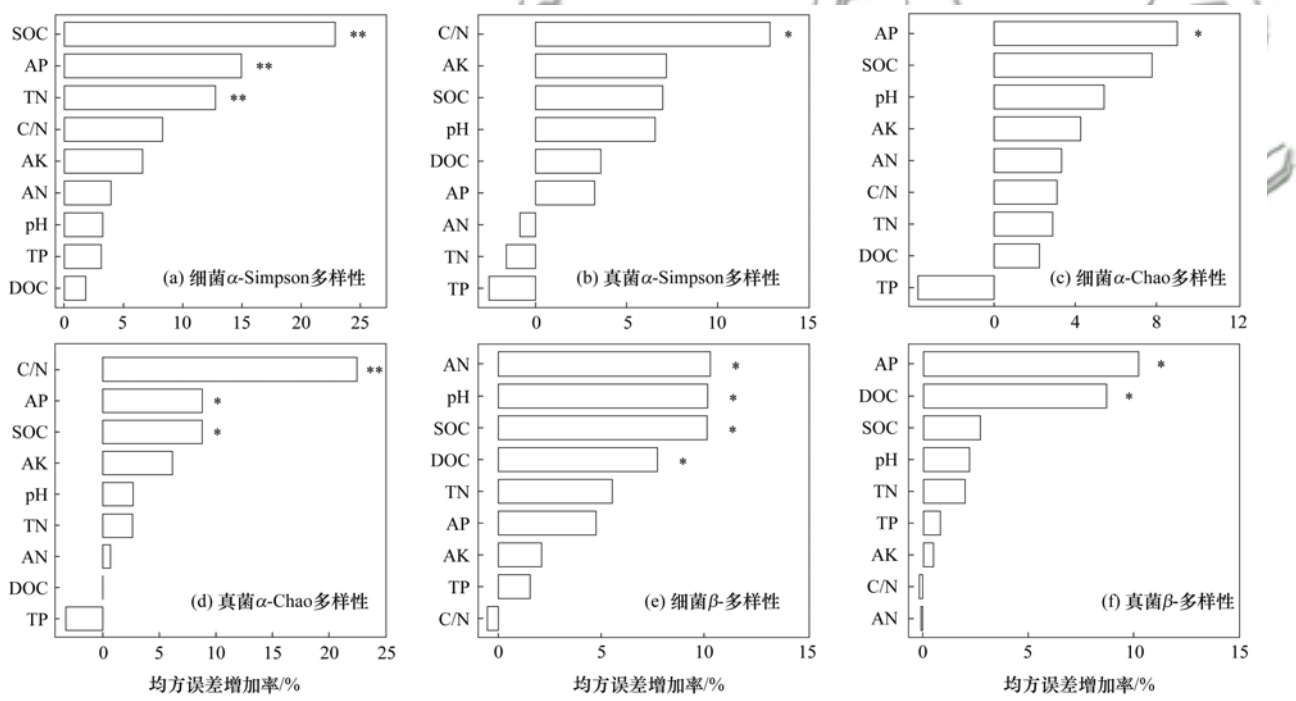


图 4　菌渣与化肥配施对土壤微生物物种组成的影响

Fig. 4　Effects of combination of fungal residue with chemical fertilizer on soil microbial species composition

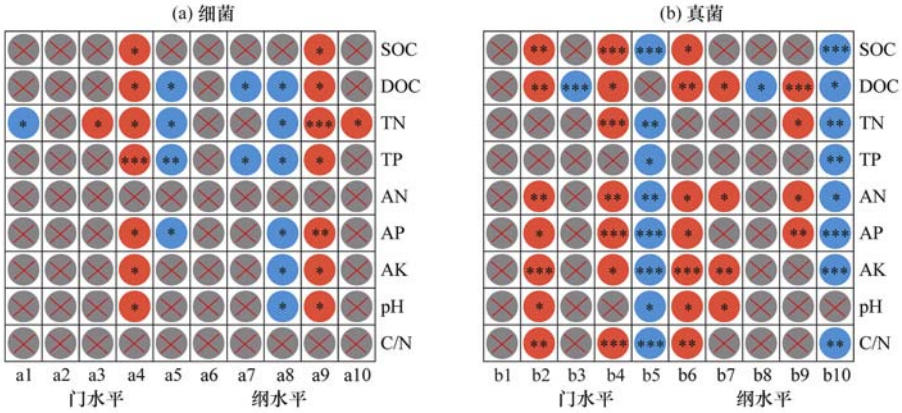


变量的均方误差增加率用于评估这些预测值的重要性，均方误差增加率越大，则意味着预测值越重要；* 为 $P<0.05$，** 为 $P<0.01$

图 5　土壤肥力指标作为土壤微生物 α-和β-多样性驱动因子的随机森林预测重要性均值

Fig. 5　Random forest mean predicted importance of the soil fertility properties as drivers for soil microbial α- and β-diversity

含量占土壤活性碳库的比例较小，但却是土壤碳库中最为重要和活跃的部分，且在土壤物质转化和养分循环中起着关键的作用[38~40]. 顾春朝等[41]的研究发现，有机肥配施化肥更有利于土壤 DOC 的释放，这与本研究的结果类似. N、P 和 K 被认为是大多数生态系统中主要的营养元素以及养分限制因素[42,43]，土壤 N、P 和 K 含量高低不仅影响着农作物的产量和品质，而且还是影响微生物群落结构的重要因子[4]，有研究表明 AP 显著增加会抑制微生物生长[44]. 菌渣与化肥配施后，较对照土壤 TN、AN、TP、AP 和 AK 都显著增加（$P<0.05$，表 1），这些结果与诸多研究一致[30,31,34,35]，土壤 C/N 是土壤质量变化的敏感指标之一，在本研究中土壤 C/N 随着菌渣添加量的增加而增加，土壤 C/N 高低对作物生长发育和土壤养分平衡具有重要的影响，且决定着有机氮的矿化以及 NH_4^+ 的硝化作用[45,46]，另外，还会介导微生物对有机质分解过程中养分的释放[45].



a1. Nitrospirae, a2. Actinobacteriota, a3. Acidobacteriota, a4. Chloroflexi, a5. Proteobacteria, a6. Actinobacteria, a7. α-Proteobacteria, a8. δ-Proteobacteria, a9. Anaerolineae, a10. Acidobacteria, b1. Rozellomycota, b2. Zygomycota, b3. unclassified _ Fungi, b4. Basidiomycota, b5. Ascomycota, b6. norank_Zygomycota, b7. Dothideomycetes, b8. unclassified_Fungi, b9. Agaricomycetes, b10. Sordariomycetesc; 红色表示显著正相关,蓝色表示显著负相关,灰色表示无显著相关; * 为 $P<0.05$, ** 为 $P<0.01$, *** 为 $P<0.001$

图 6 土壤细菌和真菌优势物种与土壤肥力指标的相关性

Fig. 6 Correlation between soil bacterial and fungal dominant species and soil fertility properties

3.2 菌渣与化肥配施对土壤细菌和真菌多样性的影响

土壤微生物多样性在可持续农业中发挥着重要作用[5,6]. 本研究发现,菌渣与化肥配施后,微生物多样性存在显著变化,细菌 α-多样性在施用化肥水平下随着菌渣用量的增加而增加[图 1(b)],这与 Liu 等[24]秸秆替代部分化肥增加了细菌多样性类似. 相反,化肥减量配施 20 t·hm^{-2}菌渣显著降低了真菌 α-多样性[图 1(c)和 1(d)]. 细菌和真菌对施用菌渣的反应不同,这可能是细菌和真菌群落获取营养素资源的偏好和策略不同. Six 等[47]的研究表明,与真菌相比较,细菌更喜欢容易分解的资源. 因此,菌渣对真菌类群的选择性影响更大,从而降低了真菌 α-多样性. 对于细菌 α-多样性增加可能是菌渣添加后为细菌提供了大量的营养物质,有利于细菌的生长. 秦红灵等[48]的研究表明土壤 AP 增加能够显著增加细菌群落多样性,在本研究中随机森林预测模型表明土壤 AP 对细菌 α-多样性有显著贡献($P<0.05$),这与姜霓雯等[49]的研究结果类似,说明土壤 AP 是细菌 α-多样性的主要驱动因子. 本研究发现 AN、SOC、pH 和 DOC 是细菌 β-多样性的主要驱动因子,与 Zheng 等[50]的报道类似,陈桂鲜等[51]的研究也表明,DOC 是细菌 β-多样性的主要驱动因子,可能解释为 DOC 易于被微生物分解利用,为微生物的活动提供了能量. 然而,在一项关于全球变化因子对土壤微生物多样性和功能的影响报道中,土壤 pH 主导着 α-多样性的反应[52],但在本研究中并非如此. 化肥减量配施 20 t·hm^{-2}菌渣降低真菌 α-多样性可能归因于菌渣本身含有丰富的碳源,高量菌渣施用增加了土壤 C/N,土壤 C/N 是决定微生物多样性的重要因素[53],N 源不足从而导致真菌 α-多样性降低;随机森林模型表明,C/N 对真菌 α-多样性有显著贡献[$P<0.05$,图 5(c)和 5(d)],与 Ning 等[54]的研究结果一致,在本研究中 AP 和 DOC 是真菌 β-多样性的主要驱动因子. 综上所述,菌渣对土壤细菌多样性的变化发挥了重要作用,可能是通过增加 AP 的含量影响细菌 α-多样性,AN、SOC、pH 和 DOC 驱动了细菌 β-多样性变化. 然而,菌渣和化肥的交互效应介导了真菌多样性变化,且化肥减量配施 20 t·hm^{-2}菌渣降低了真菌 α-多样性,C/N 在其中发挥了重要的作用,AP 和 DOC 主导了真菌 β-多样性变化.

3.3 菌渣与化肥配施对细菌和真菌优势物种组成的影响

微生物群落对维持土壤生态系统功能至关重要,且受到土地利用、耕作和施肥等的影响[55~57]. 在本研究中与细菌相比,真菌在门及纲水平上物种变化更明显,这可能是真菌对环境的敏感性,菌渣与化肥配施后土壤环境因子发生明显变化,其丰度受到影响[14]. 子囊菌门(Ascomycota)是所有处理中最优势的菌门,这与 Chen 等[58]和 Bei 等[59]的研究结果一致. 化肥减量配施 20 t·hm^{-2}菌渣降低了子囊菌门(Ascomycota)和粪壳菌纲(Sordariomycetes)的相对丰度[图 4(c)和 4(d)],但 Wu 等[60]的研究表明,化肥减量与有机物料相结合显著增加了子囊菌门(Ascomycota)的相对丰度. 子囊菌门(Ascomycota)的相对丰度降低可能归因于两方面:首先,可能是过高的土壤养分抑制了子囊菌门(Ascomycota)生长[61],子囊菌门(Ascomycota)与 SOC、TN、TP、AN、AP、AK、pH 和 C/N 呈显著负

相关能解释这一观点[图 6(b)]. 其次,可解释为与菌渣本身有较高的 C/N 密切相关,子囊菌门(Ascomycota)是土壤中有机物质的主要分解者,对腐烂有机基质具有降解能力[62],子囊菌门(Ascomycota)分解需要大量的 N,可能由于 N 缺乏抑制了子囊菌门(Ascomycota)的活性,从而导致子囊菌门(Ascomycota)的相对丰度降低. 另外,本研究发现子囊菌门(Ascomycota)与担子菌门(Basidiomycota)之间可能存在竞争关系,这与 Ye 等[63]的研究结果类似. 担子菌门(Basidiomycota)与子囊菌门(Ascomycota)一样,是典型的腐生菌门[64],具有环境友好和分解有机物质的特点[63],在降解土壤中高木质素含量的凋落物方面发挥了重要的作用[65]. 综上所述,化肥减量配施 20 t·hm^{-2}菌渣通过增加土壤养分和 C/N 抑制了子囊菌门(Ascomycota)和粪壳菌纲(Sordariomycetes)的相对丰度.

4　结论

(1)与对照相比较,菌渣与化肥配施显著增加了土壤肥力指标的含量,菌渣主导了土壤肥力指标含量的变化.

(2)菌渣对土壤细菌多样性的变化发挥了重要作用,可能是通过增加 AP 的含量影响细菌 α-多样性,AN、SOC、pH 和 DOC 驱动了细菌 β-多样性的变化. 然而,菌渣和化肥的交互效应介导了真菌多样性变化,且化肥减量配施 20 t·hm^{-2}菌渣降低了真菌 α-多样性,C/N 在其中发挥了重要的作用,AP 和 DOC 主导了真菌 β-多样性的变化.

(3)化肥减量配施 20 t·hm^{-2}菌渣降低了真菌子囊菌门(Ascomycota)和粪壳菌纲(Sordariomycetes)的相对丰度,且施用菌渣较化肥在影响土壤细菌和真菌的门及纲水平上优势物种变化方面更具优势.

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HUANJING KEXUE

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Vol. 44 No. 4

Apr. 15, 2023

CONTENTS