

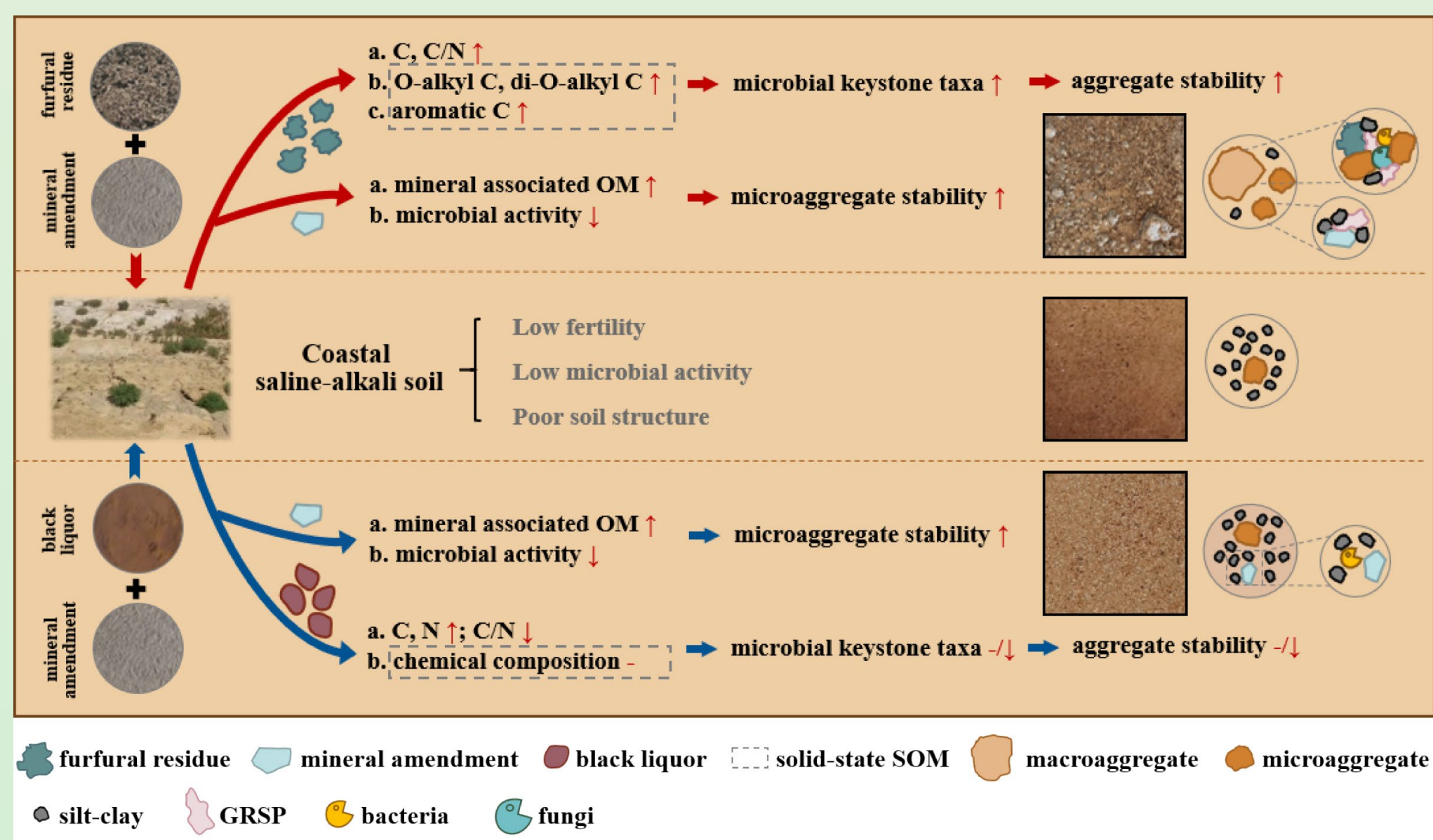
## Effects of different amendments on aggregate stability and microbial communities of coastal saline-alkali soil in the Yellow River Delta

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### ABSTRACT

Organic amendments have been widely used in coastal saline-alkali soil remediation; however, the mechanisms involved and the interactions between organic and inorganic amendments are still unclear. In this work, furfural residue and black liquor were tested to examine their effects on chemical properties, water-stable aggregate fractions, chemical compositions of solid-state soil organic matter (SOM), glomalin-related soil protein contents and microbial communities of coastal saline-alkali soil under a 400-day incubation experiment. Furthermore, organic amendments mixed with mineral amendment (4:1) were employed to explore the interactions between organic and inorganic amendments. Furfural residue had stronger and longer effects on soil macroaggregate stability (~240 days, intense) than black liquor (~15 days, weak), and mineral amendment addition had a positive effect on the stability of microaggregates. Our results revealed that qualities (primary form, C/N, and chemical composition) of organic amendment which can change microbial communities by increasing soil C/N and effective chemical compositions of solid-state SOM, are the key factors in promoting the rapid formation and longer stability of coastal saline-alkali soil aggregates. Moreover, inorganic amendment addition can further improve the formation and stability of microaggregates rather than those of macroaggregates. This study provided a much-needed technical basis for remediation of coastal saline-alkali soil.



### INTRODUCTION

The coastal wetland of Yellow River Delta (YRD) is one of the largest, most integrated and youngest wetlands in the warm temperate zone of China, and the YRD has the most reserve land resources in the east coast. While, soil salinization in this region induced and compounded problems like low fertility, low microbial activity, and poor soil structure, seriously affecting ecological balance and agricultural development. In consequence, the remediation and comprehensive utilization of coastal saline-alkali soils in this region are of great significance to solve the shortage of agricultural land and promote the development of ecosystem restoration and agricultural economy. At present, a large number of scholars have studied the effects of different amendments on the remediation of saline-alkali soils in the YRD, but few studies have provided technical guidance on the mechanism for the selection of effective amendments in this region.

Here, we examined the effect of two organic amendments (furfural residue and black liquor) with and without addition of inorganic amendment (mineral amendment) on aggregate stability and microbial communities of coastal saline-alkali soil with a 400-day incubation experiment. We hypothesized that the two organic amendments had different effects on soil microbial communities to affect the formation and stability of soil aggregates because of their different qualities. We further hypothesized that inorganic amendment addition can improve the stability of soil microaggregates instead of macroaggregate. To test our hypotheses, we measured a range of soil chemical properties, water-stable aggregate fractions, and glomalin-related soil protein (GRSP) content during incubation, and selected the days ( $R_{(0.25)} \max$  day) when the water-stable macroaggregate fraction ( $>0.25$  mm,  $R_{0.25}$ ) of each treatment was the highest. We then measured the solid-state SOM chemical compositions ( $^{13}\text{C}$  NMR) and quantified soil microbial communities on the  $R_{(0.25)} \max$  and 400<sup>th</sup> days. Finally, we linked these parameters to explore how organic amendment qualities and inorganic amendment addition affected the formation and stability of aggregates.

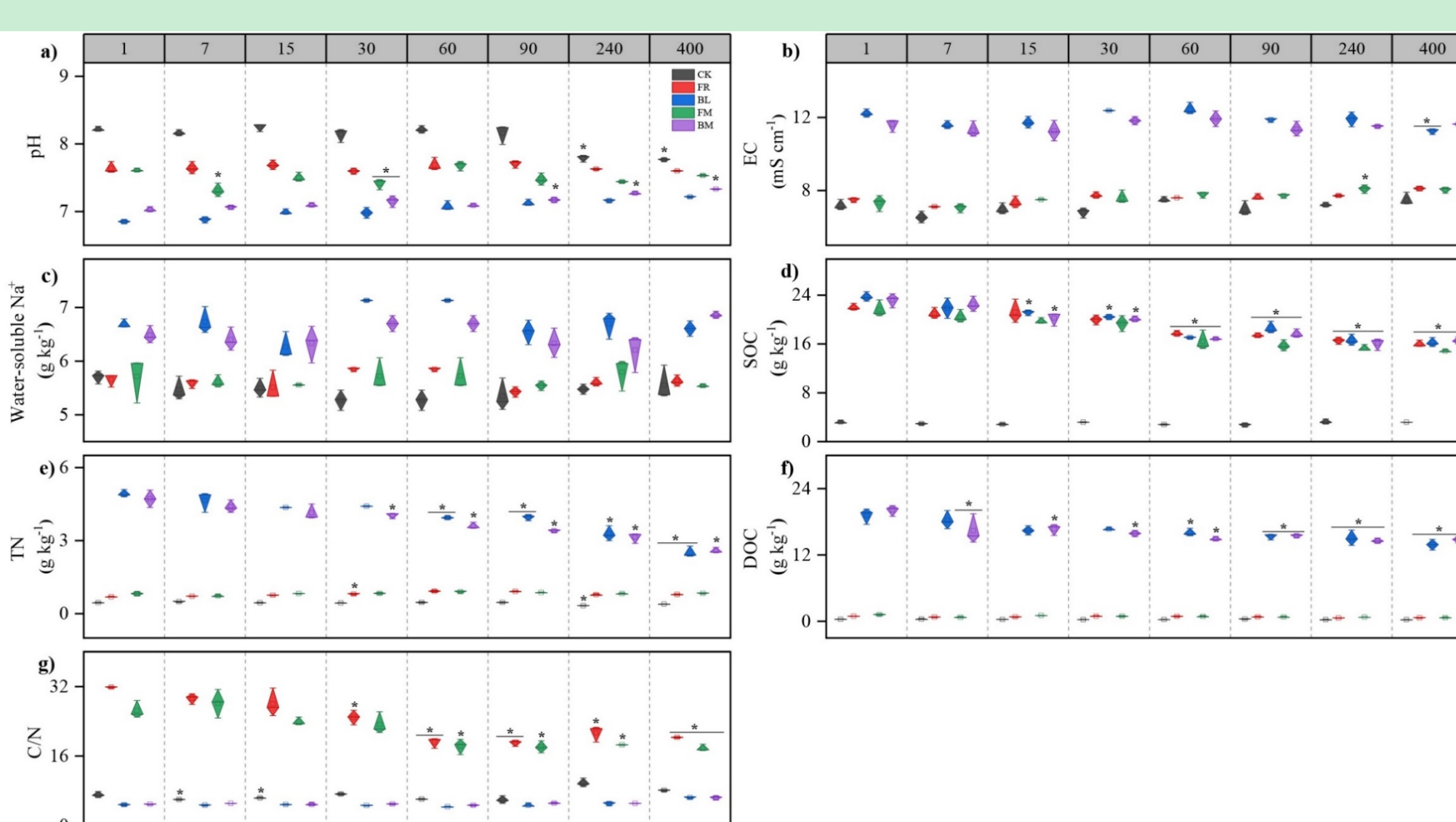
### MATERIALS AND METHODS

**Preparation of soil and amendments.** Soil samples for the experiment were taken from fallow land (0–20-cm depth) in Kenli County, Shandong Province, China (118° 36' 5" E, 37° 35' 37" N). The soil is classified as a Salic Fluvisol (the texture is sandy loam). The organic amendments include furfural residue and black liquor, and the inorganic amendment is potassium-silicon-calcium mineral fertilizer (mineral amendment), which is made by a hydrothermal reaction involving potassium feldspar ( $\text{K}_2\text{O} \cdot \text{Al}_2\text{O}_3 \cdot 6\text{SiO}_2$ ) and quicklime (CaO). The amendments were ground to  $<0.25$  mm by a mill before the incubation experiment.

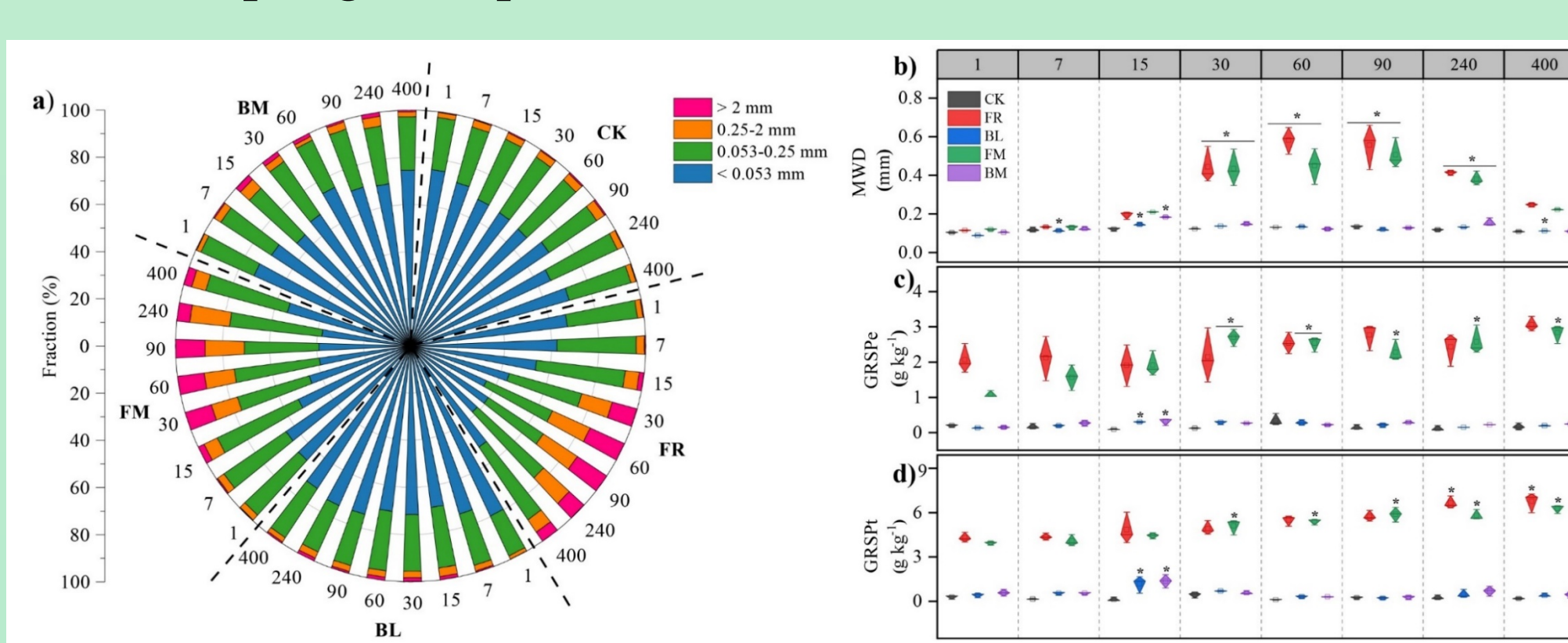
**Incubation experiment.** A 400-day incubation study was carried out in a growth incubator without light at 25°C. The sieved (2mm) soil samples (100-g dry weight) were stored in 180-mL plastic cups, and the amendments were added into the soils at a dose of 20 g-OC·kg<sup>-1</sup> dry soil and mixed thoroughly. The incubation experiment consisted of a control (no amendment, CK) and four amendment treatments, with three replicates for each: (1) FR: with 4.33-g furfural residue; (2) BL: with 7.17-g black liquor; (3) FM: with 4.33-g furfural residue and 1.08-g mineral amendment (mass ratio is 4:1); and (4) BM: with 7.17-g black liquor and 1.79-g mineral amendment (mass ratio is 4:1). Deionized water was added into soils to fill 60% of the pore space per week, and a layer of aluminum foil (with six 0.5-cm diameter holes) was wrapped over each plastic cup. Each treatment had 24 samples, which were used for sampling on days 1, 7, 15, 30, 60, 90, 240, and 400 during incubation.

### RESULTS

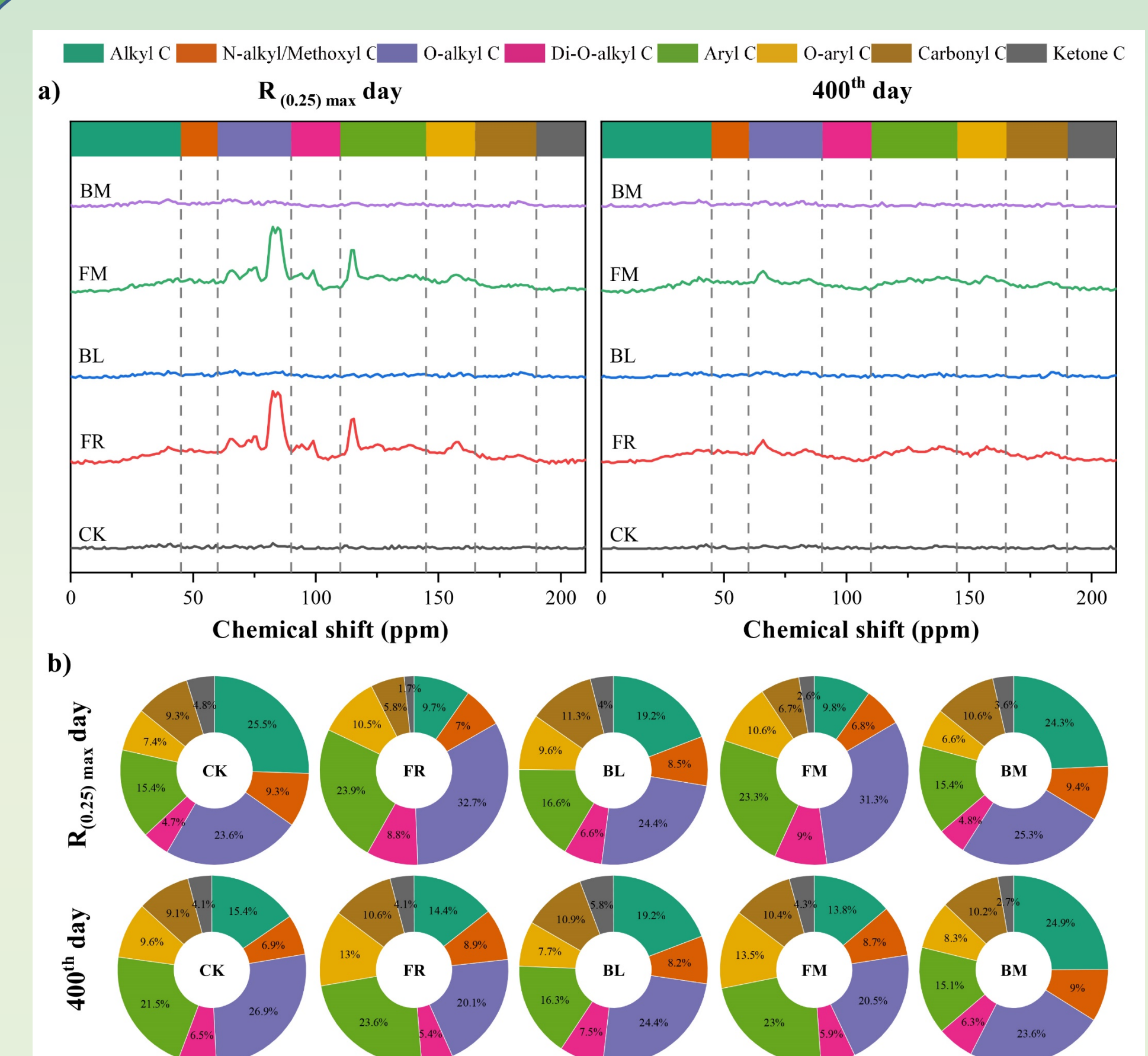
- 1) Furfural residue reduced the soil pH and increased SOC, TN, C/N, and DOC; black liquor reduced the soil pH and C/N, but increased EC, water-soluble Na<sup>+</sup>, SOC, TN, and DOC; and the addition of mineral amendment limited these effects on soil properties.
- 2) Furfural residue improved the water-stability of aggregates while black liquor had a slight negative effect, and mineral amendment further increased the fraction of water-stable microaggregate; Furfural residue and black liquor significantly affected the increase of GRSP, while mineral amendment slightly reduced the effect of furfural residue.
- 3) The chemical composition of solid-state SOM changed greatly with the input and decomposition of furfural residue, compared with black liquor.
- 4) Furfural residue and black liquor significantly changed the compositions of soil bacterial and fungal communities, while mineral amendment obviously influenced the effects of black liquor more than those of furfural residue; Furfural residue reduced the alpha diversities of fungi while mineral amendment limited these effects, and black liquor reduced the bacterial alpha diversities.
- 5) The Pearson correlation analysis showed that water-stability of aggregates were positively correlated with soil C/N and GRSP, and negatively correlated with EC, Na<sup>+</sup>, TN, and DOC. Furfural residue had obvious impacts on soil C/N, water-stability of aggregates, and GRSP, but black liquor obviously affected soil EC, Na<sup>+</sup>, TN, and DOC.



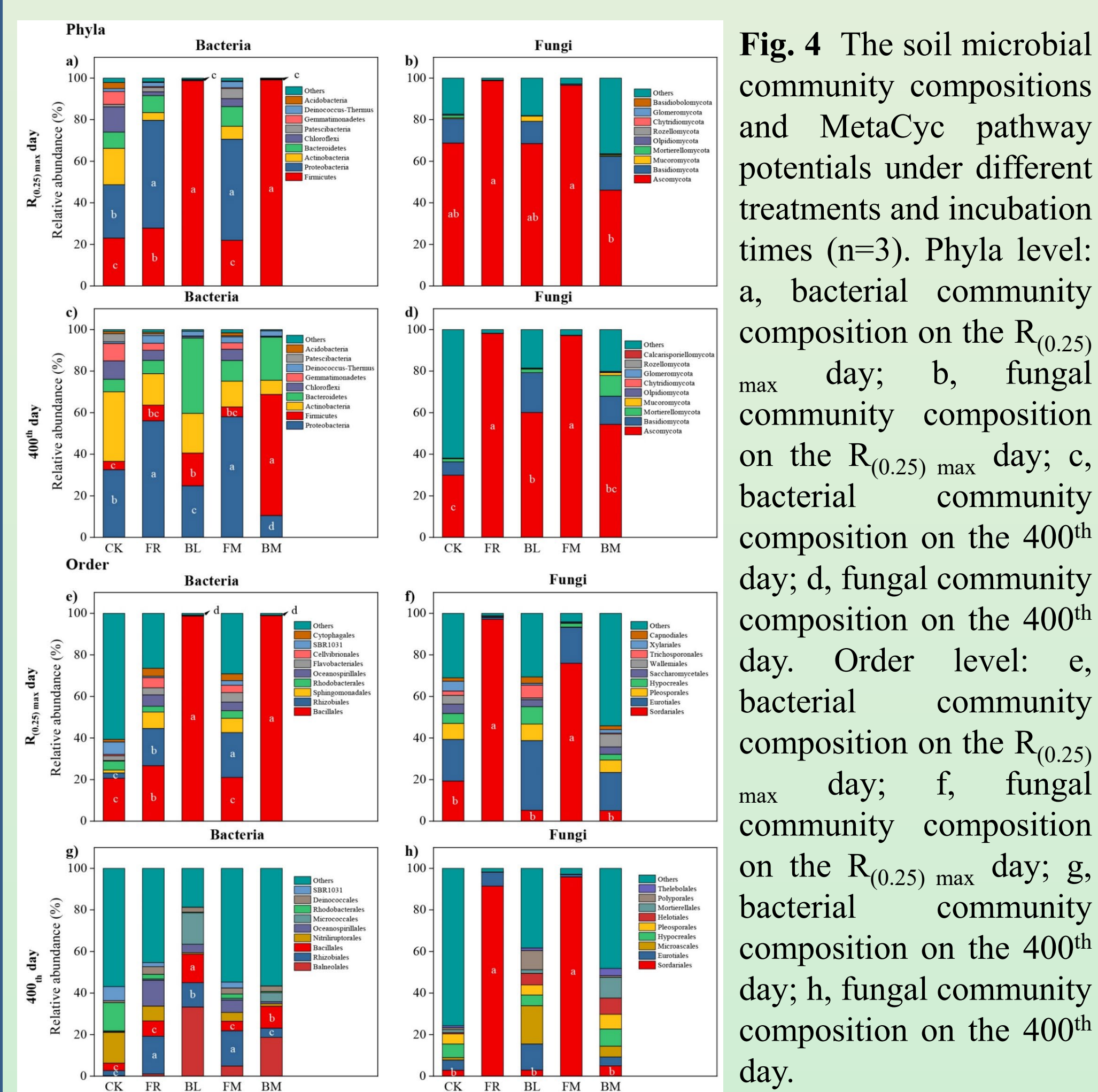
**Fig. 1** Dynamics in soil chemical properties during incubation (n=3). Significant differences ( $P < 0.05$ ) between the 1st day and other sampling time points of the same treatment indicated with \*.



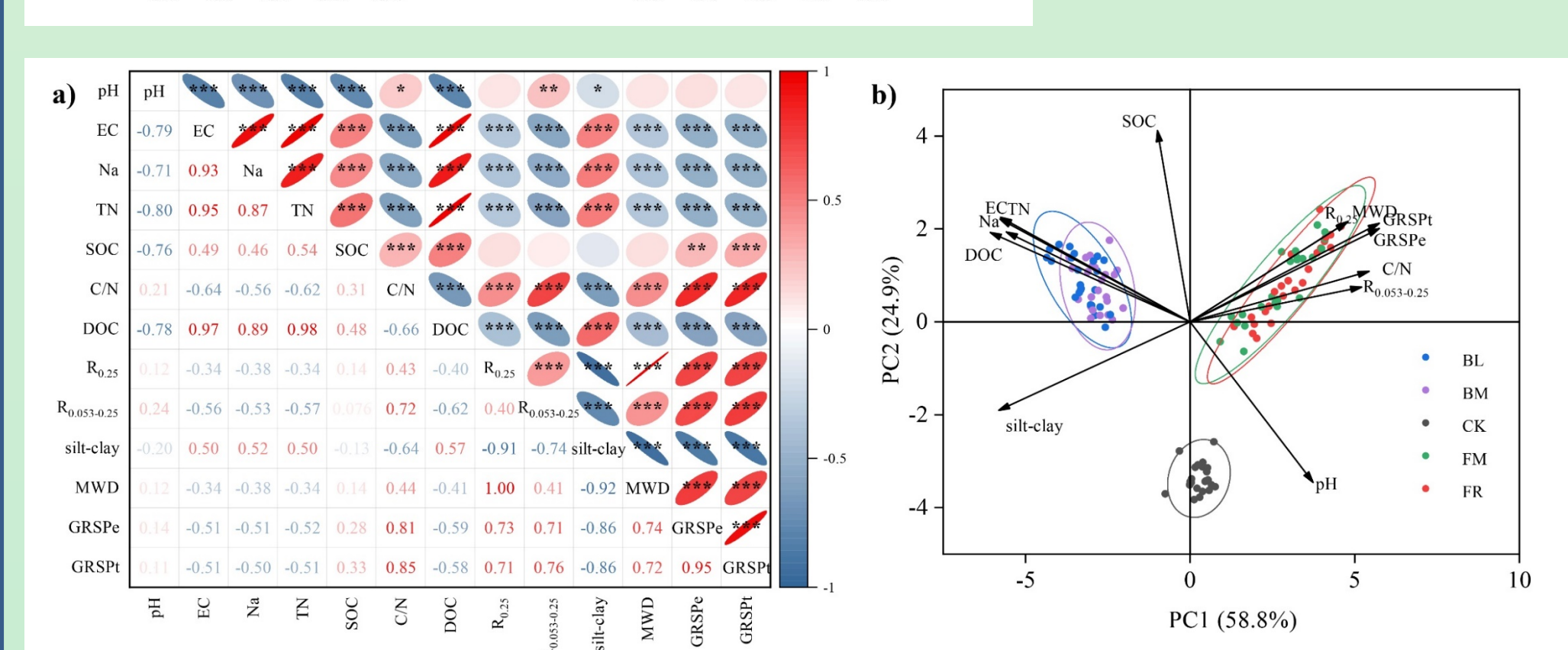
**Fig. 2** Dynamics in soil water-stable aggregate fractions, mean weight diameter (MWD), and glomalin-related soil protein content (GRSP) during incubation (n=3).



**Fig. 3**  $^{13}\text{C}$  NMR spectra of solid-state SOM on the  $R_{(0.25)} \max$  and 400<sup>th</sup> days. a, solid-state  $^{13}\text{C}$  NMR spectra; b, relative abundance of chemical composition.



**Fig. 4** The soil microbial community compositions and MetaCyc pathway potentials under different treatments and incubation times (n=3). Phyla level: a, bacterial community composition on the  $R_{(0.25)} \max$  day; b, fungal community composition on the  $R_{(0.25)} \max$  day; c, bacterial community composition on the 400<sup>th</sup> day; d, fungal community composition on the 400<sup>th</sup> day. Order level: e, bacterial community composition on the  $R_{(0.25)} \max$  day; f, fungal community composition on the  $R_{(0.25)} \max$  day; g, bacterial community composition on the 400<sup>th</sup> day; h, fungal community composition on the 400<sup>th</sup> day.



**Fig. 5** The interrelation between soil chemical properties, water-stable aggregate fractions and GRSP contents by amendments during incubation (n=120). a, Pearson correlation analysis; b, principal component analysis (PCA). \*:  $P < 0.05$ , \*\*:  $P < 0.01$ , \*\*\*:  $P < 0.001$ .

### CONCLUSION

Our findings demonstrate that organic amendment qualities and inorganic amendment addition could significantly influence the formation and stabilization of aggregates in coastal saline-alkali soil. High soil C/N and the relative abundance of effective chemical compositions (O-alkyl C, di-O-alkyl C, and aromatic C) of solid-state SOM have stronger and longer effects on the water stability of aggregates by increasing the relative abundance of keystone taxa (such as orders *Rhizobiales* and *Sordariales*) and GRSP contents in soils. The qualities (primary form, C/N, and chemical composition) of organic amendment are the key factors in affecting the form, C/N, and chemical composition of SOM. In addition, O-alkyl C and di-O-alkyl C fractions of solid-state SOM promote the rapid and intense formation of macroaggregates, while aromatic C make the aggregate stability more durable. We also found that abundant N inputs have no positive or even negative influence on the formation and stability of aggregates. Inorganic amendment addition further increased the fractions of microaggregates, while having negative effects on microbial activities.