¹ Effect of domestication and microbial

² community on medium chain fatty acids

production from Chinese liquor distillers' grain

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HIGHLIGHTS:

- Pit mud is a suitable inoculum for MCFA prodction from residue
- Domestication of pit mud improves caproate production
- Caproiciproducens and Lactobacillus coexist in pit mud help CE
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BACKGROUND: Chinese strong-aroma type liquor used food crops as 16 substrate, and in the later stage of fermentation, relied on the pit mud for 17 the esterification reaction of ethanol with caproate, butyrate and acetate 18 to produce the aforementioned flavour substances. Chinese liquor 19 distillers' grain (CLDG) is an incomplete fermentation byproduct, which 20 contained polysaccharides, and has a huge output. For the chain 21 elongation (CE) process, pit mud is an appropriate inoculum rich in 22 caproate-producing microbiome. 23

In this study, CLDG was used as fermentation substrates, pit mud and domesticated one were used as inoculum to produce MCFAs with complex substrates was investigated; the fermentation effects and microbial community changes were studied.

28 **RESULTS & DISCUSSION**:

The caproate concentration inoculated by shallow pit mud and the 29 domesticated one were 394 and 449 mg COD/g VS (substrate) 30 respectively. And the caproate carbon selectivity increased from 32.5% to 31 37.1% in total fatty acid yield. The abundance of caproate producing 32 bacteria was improved. The domestication shallow pit mud makes more 33 butyrate participated in the CE process and converts more caproate. The 34 concentration of lactate and ethanol during fermentation showed shallow 35 pit mud can effectively utilize lactate to produce caproate. 36

The characteristics of microbial composition in shallow pit mud and domesticated one showed the rich *Caproiciproducens* and *Lactobacillus* coexisted, and their abundance has increased from 12% to 25% and 3.7% to 18%, respectively. The strain of *Caproiciproducens* spp in the pit mud produce caproate by metabolizing lactate (Figure 1).

42 **CONCLUSION**: Using the pit mud of CSAL as inoculum and CLDG as 43 substrate, the shallow pit mud fermentation system produced caproate. 44 By domestication of shallow pit mud, the caproate concentration and the 45 caproate carbon selectivity increased. The analysis of microbiome showed 46 that lactate in CLDG was used as the main ED, and the main caproate 47 producing species was *Caproiciproducens* spp.

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