Lactiplantibacillus plantarum attenuates Coxsackievirus B3-induced pancreatitis through the BAX/BCL2/CASP3 signaling pathway

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Supplementary Materials

Attachment and acid bypass Assay

For virus attachment, HT-29 cells were infected with the CVB3 at 4°C for 1 h after pretreated with *L. plantarum* FLPL05 2 h. And then, the cells were washed three times with cold PBS to remove the unbound virus particles.

For entry assays, CVB3 (MOI=1) was prebound to HT-29 cells for 1 h at 4°C. Cells were washed with cold PBS three times to remove unbound virus. And then, *L. plantarum* FLPL05 was added, incubating at 37°C for 1 h, and washed three times with cold PBS. Finally, cells were further incubated at 37°C for 1 h. These absorbed and penetrated viruses were collected separately to plaque assay.

Plaque Assay

The viruses which absorbed on the surface of HT-29 cells or penetrated into the HT-29 cells were collected respectively and used for plaque assay. Viral titers [plaque-

forming units (PFU)/ml] were measured by plaque assays on HeLa cells. HeLa cells were re-plated in 6 well plates and were infected after confluency reached about 95%. Removed the virus after infected for 1 h and overlaid with a 1:1 mixture which contained DMEM and 0.9% agarose (YEASEN, Shanghai, China). After 48 h of incubation period, the cells were fixed with 10% formaldehyde. The mixture was removed before the cells were stained with 1% crystal violet, and plaques were counted finally.

Fig. S1 Basic blood biochemical and body weight changed of *L. plantarum* FLPL05 overall protection mice against CVB3 infection. (A) total bilirubin (TBIL), (B) direct bilirubin (DBIL), (C) indirect bilirubin (IBIL), (D) Alanine aminotransferase (ALT), (E) aspartate aminotransferase (AST), (F) cholesterol (CHOL), (G) triglyceride (TG), (H) high density lipoprotein cholesterol (HDL-C), (I) low-density lipoprotein cholesterol (LDL-C), (J) glucose (GLU), (K) UREA, (L) Creatinine (CREA), (M) Uric acid, and (N) body weight. Data are presented as mean \pm SD, with *p*-values based on unpaired two-tailed Student's *t*-tests. Key: *, p < 0.05; **, p < 0.01; ***, p < 0.001.

Figure S2. *L. plantarum* **FLPL05** inhibited the process of adsorption and penetration of **CVB3** into cells. (A) CVB3 which adsorbed on the HT-29 cells forms plaques in HeLa cells. (B) Western blot detected the adsorption of CVB3 in the different groups. (C) CVB3 which penetrated into the HT-29 cells forms plaques in HeLa cells. (D) Western blot detected the penetration of CVB3 in the different groups.



Figure S1. YU et al.,



Figure S2. YU et al.,