**Abstract**

**Problem:** This study aims to investigate the effects of alpha-linolenic acid (ALA) on the gut microbiota (GM) and the abdominal environment in mice with endometriosis (EMS).

**Methods:** The effects of faecal microbiota transplantation (FMT) from EMS mice on mice treated with antibiotic cocktail were conducted. The 16S rRNA sequencing and PICRUSt software were used to detect the structure and function of GM respectively. The protein levels of Claudin 4 and ZO-2 in the intestinal wall were detected using the western blotting. The level of LPS in the abdominal cavity was detected using enzyme-linked immunosorbent assay (ELISA). The content of macrophages in the abdominal cavity was detected using flow cytometry.

**Results:** The exogenous supplementation of ALA could restore the abundance of Firmicutes and Bacteroidota in EMS mice. After the ALA treatment, the abundance of 125 functional pathways and 50 abnormal enzymes related to GM in EMS mice was significantly improved (p < .05). The expression of the ZO-2 protein in the intestinal wall was decreased, and the level of LPS in the abdominal cavity was significantly increased after FMT from EMS mice (p < .05). ALA could increase the expression of the ZO-2 protein in the intestinal wall of EMS mice, reduce the level of LPS in the abdominal cavity (p < .05) and reduce the aggregation of peritoneal macrophages (p < .05).

**Conclusion:** Alpha-linolenic acid can improve the GM, intestinal wall barrier and abdominal inflammatory environment and reduce the level of LPS in mice with EMS.