Berberine inhibits lipopolysaccharide-induced expression of inflammatory cytokines by suppressing TLR4-mediated NF-κB and MAPK signaling pathways in rumen epithelial cells

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Short title: **Berberine inhibits inflammation in REC**

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Summary

Subacute ruminal acidosis (SARA) can increase the level of inflammation and induce rumenitis in dairy cows. Berberine (BBR) is the major active component of *Rhizoma Coptidis*, which is a type of Chinese anti-inflammatory drug for gastrointestinal diseases. The purpose of this study was to investigate the anti-inflammatory effects of BBR on lipopolysaccharide (LPS)-stimulated rumen epithelial cells (REC) and the underlying molecular mechanisms. REC were cultured and stimulated with LPS in the presence or absence of different concentrations of BBR. The results showed that cell viability was not affected by BBR. Moreover, BBR markedly decreased the concentrations and mRNA expression of pro-inflammation cytokines, including tumor necrosis factor-α, interleukin-1β, and interleukin-6 in the LPS-treated REC in a dose-dependent manner. Importantly, Western blotting analysis showed that BBR significantly suppressed the protein expression of toll-like receptor 4 (TLR4) and myeloid differentiation primary response protein and the phosphorylation of nuclear factor-κB (NF-κB), inhibitory kappa B (IκBα), p38 mitogen-activated protein kinase (MAPK), and c-Jun N-terminal kinase (JNK) in LPS-treated REC. Furthermore, the results of immunocytofluorescence showed that BBR significantly inhibited the nuclear translocation of NF-κB p65 induced by LPS treatment. In conclusion, the protective effects of BBR on LPS-induced inflammatory responses in REC may be due to its ability to suppress the TLR4-mediated NF-κB and MAPK signaling pathways. These findings suggest that BBR can be used as an anti-inflammatory drug to treat inflammation induced by SARA.