



## Induction of chicken cytokine responses *in vivo* and *in vitro* by lipooligosaccharide of *Campylobacter jejuni* HS:10



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### ABSTRACT

*Campylobacter jejuni* is a pathogen of the gastrointestinal tract of humans, but colonizes chickens for prolonged periods without causing disease. It is unclear what host and bacterial mechanisms maintain a non-inflammatory state in chickens. The present work was undertaken to characterize cytokine responses of chickens to purified lipooligosaccharide (LOS) of *C. jejuni* HS:10. Chickens were injected with purified LOS, and expression of interleukin (IL)-1 $\beta$  (pro-inflammatory cytokine), IL-8 (pro-inflammatory chemokine), interferon (IFN) $\gamma$  (Th1-like cytokine), IL-10 (immune regulatory/anti-inflammatory cytokine) and IL-13 (Th2-like cytokine) was evaluated in spleen using quantitative RT-PCR, up to 24 h post-injection. In an *in vitro* study, splenocytes were incubated with LOS, and cytokine expression followed up to 18 h. Chickens injected with LOS had increased expression of IL-1 $\beta$  up to 24 h later. Expression of IL-8 was significantly increased at 2 h but then declined below baseline. Expression of IFN $\gamma$  and IL-10 was increased significantly at 2 h, but declined thereafter. Splenocytes incubated with LOS had increased expression of IL-1 $\beta$  and IL-8 up to 18 h of incubation. Expression of IFN $\gamma$  was increased at 6 and 18 h, IL-10 was increased at 2 h, but expression of IL-13 did not differ significantly up to 18 h. It is concluded that LOS of *C. jejuni* can induce expression of pro-inflammatory IL-1 $\beta$  and IL-8, as well as IFN $\gamma$  and IL-10 in chickens. More extensive studies with more prolonged exposure to LOS are needed to further clarify the interaction between *C. jejuni* and the chicken host.

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### 1. Introduction

*Campylobacter jejuni* is a major bacterial cause of food-borne illness in humans (Scallan et al., 2011) but colonizes chickens for prolonged periods without causing inflammation or clinical disease (Dhillon et al., 2006). It is unclear what host and bacterial mechanisms maintain a non-

inflammatory state in chickens. Lipopolysaccharides (LPS) and lipooligosaccharides (LOS) are integral structural components of the outer membranes of Gram-negative bacteria, that interact with pattern recognition receptors (PRR) of innate immune defenses and induce inflammatory responses. The diverse forms of LOS expressed by different strains of *C. jejuni* play an important role in colonization of chickens (Javed et al., 2012), and may assist in immune evasion. Chickens express PRR homologous to mammalian TLR1/6/10, TLR2, TLR3, TLR4, TLR5, TLR7 (Boyd et al., 2007). A homolog to mammalian TLR9 has not been identified, but unmethylated CpG can activate NF- $\kappa$ B via chicken TLR21 (Brownlie and Allan, 2011). In mammals TLR4, in

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