

LIVE COCCIDIOSIS VACCINE FOR BREEDERS AND LAYERS (EVALON®) IMMUNE MODULATION AND ENHANCEMENT OF IMMUNITY BY THE USE OF AN ADJUVANTED SOLVENT (HIPRAMUNE®T)

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INTRODUCTION

EVALON® is a live attenuated vaccine against avian coccidiosis. It is composed of five attenuated strains: *Eimeria acervulina* 003, *E. brunetti* 034, *E. maxima* 013, *E. necatrix* 033 and *E. tenella* 004. These strains were selected to maximise immunogenicity and minimise the side effects of *Eimeria* parasites. Avian *Eimeria* have a complex life cycle with a combination of exogenous and endogenous stages that trigger a response by the host's immune system. However, *Eimeria* parasites have also been described as being highly elusive to the immune system, as well as producing chemokines that can slow or inhibit the host's immune response (Jang 2011, Schmid 2014, Miska 2013). Although it is well known that live vaccines can induce adequate immunity without being combined with an adjuvant, we firmly believe that immune modulation is crucial in providing strong, fast and long-lasting immunity (Dalloul 2005).

MATERIALS AND METHODS

In this study, four treatment subgroups received EVALON®, EVALON® in combination with HIPRAMUNE®T or phosphate buffered solution (control group). Five birds from each subgroup were used to obtain intestinal lymphocytes from the mucosa and Peyer's patches at different time points post-vaccination (7, 23 and 43 days p.v.). The lymphocytes were then incubated in appropriate medium and stimulated overnight with *Eimeria* whole antigen. Later, the lymphocytes were fixed and stained using monoclonal antibodies marked with fluorescein and studied with flow cytometry to detect lymphocytes producing IL-2, IFN- γ , IL-4 and IL-10.

RESULTS AND DISCUSSION

The results obtained in the first experiment indicated that HIPRAMUNE®T is able to increase the level of Th1 cytokines, as indicated by the results obtained for IL-2 (Figure 1; significant differences were detected at days 23 and 43 for the mucosa and at day 43 for the Peyer's patches) and IFN- γ (Figure 2; significant differences were detected at days 7 and 23 for the mucosa and at days 23 and 43 for the Peyer's patches).

Figure 1. Mean percentage of positive results for IL-2. Results obtained with lymphocytes isolated from the mucosa and intestinal Peyer's patches. Superscripts (a,b) indicate statistical differences.

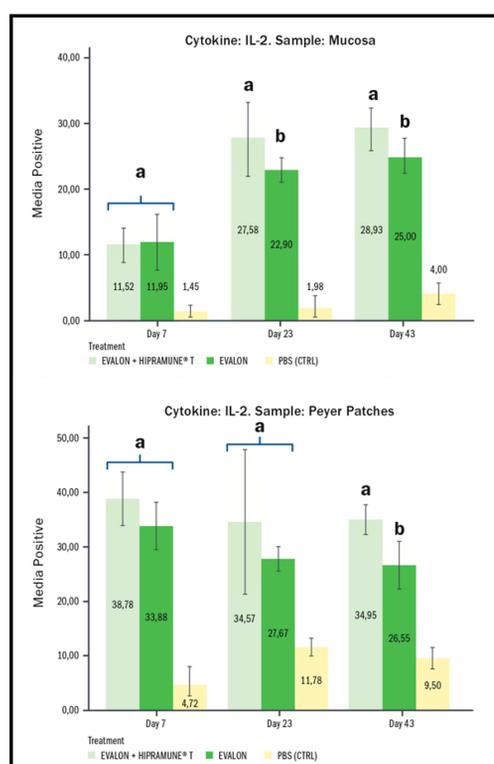
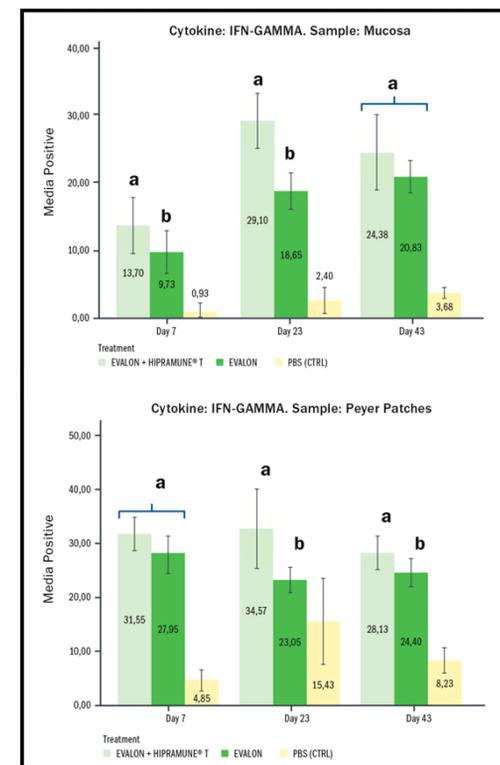


Figure 2. Mean percentage of positive results for IFN- γ . Results obtained with lymphocytes isolated from the mucosa and intestinal Peyer's Patches. Superscripts (a,b) indicate statistical differences.



By contrast, the level of IL-4 and IL-10 at days 23 and 43 was equal or lower when EVALON® and EVALON® + HIPRAMUNE®T were compared. These results, combined with the results recorded for IL-2 and IFN- γ , confirm the ability of HIPRAMUNE®T to stimulate a cellular immune response. Therefore, it is hypothesised that EVALON® administered together with HIPRAMUNE®T is able to polarise the immune response towards a Th1 response with more intensity, as the study indicates, than the live vaccine without the adjuvant. The Th1 response is crucial for protection against *Eimeria* (del Cacho 2011 and 2012). In vaccines designed for layers and breeders, which are long-lived birds, it is of paramount importance to provide extended protection throughout the life cycle. EVALON®'s efficacy is boosted by co-administration with HIPRAMUNE®T.

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