COMPARISON BETWEEN ATTENUATED AND NON-ATTENUATED COCCIDIOSIS VACCINES

Martina Dardi*, Massimiliano Baratelli1, Oriol Boix1, Marc Pagès2

1HIPRA, Amer (Girona), Spain. 
2HIPRA Scientific S.L.U., Amer (Girona), Spain.
*Author for correspondence: Avda. La Selva 135, 17170, Amer (Girona), Spain. Email: martina.dardi@hipra.com. Tel. +34 671 519564.

## 1 Introduction

Most of the coccidiosis vaccines available for chickens are living parasites that have to go through two and sometimes even three life cycles in the intestine of the host to activate the immune system and thus achieve fully protective immunity. Live attenuated and non-attenuated vaccines are available on the market. Live non-attenuated vaccines consist of parasites that continue to maintain their natural virulence. Control of the occurrence of adverse reactions (coccidiosis disease) is achieved by the use of a reduced number of oocysts in the vaccine preparations and, in some cases, even by the use of anticoccidial drugs to control the excessive spread of the vaccine strains. Live attenuated vaccines are specifically designed to generate an immune response but limit the risk of possible adverse events. The most used attenuation system is the selection of the strains by precocious development (1), although attenuation through embryonated egg passages in E. tenella has also been used for some live attenuated vaccines for commercial use. Therefore, there may be different degrees of attenuation due to the use of different processes to achieve the attenuation. There has been some concern among users regarding the safety of live coccidiosis vaccines. The aim of this study was to compare the safety parameters of the attenuated and non-attenuated Eimeria vaccines which are currently marketed together with EVALON®, a live attenuated vaccine against coccidiosis for use in breeders and layers.

## 2 Materials and methods

One-day-old COBB chicks were purchased from a commercial supplier in Spain. The weight of the birds at one day of age was between 40 g and 50 g. The animals were introduced into experimental facilities coccidia-free and were raised in pens with the ground covered with wood shavings as bedding material; the litter was not changed during the study. The temperature and humidity were checked daily and maintained within a range of between 25 °C and 26 °C and over 50%, respectively. Feed and water were provided ad libitum. The commercial feed did not contain coccidiostats. The animals were divided into 5 groups (1-5) of 105 individual animals each, using weight as a stratification factor. Table 1 shows the study design and the parameters evaluated.

## 4 Discussion

The results of this study show the presence of the following differences by product in comparison with EVALON®:

- Clinical signs and alteration of droppings: more persistent in groups 3 and 5, less in group 4.
- Intestinal lesions: highest in groups 3, 4 and 5.
- OPG in the litter: replication peaks of the vaccines very high at 7 dpv in groups 3 and 5, while in group 4 there was a delayed peak at 21 dpv.

In conclusion, the results of this study indicate that both the attenuation itself and its degree may be important factors that must be taken into account when choosing live vaccines against coccidiosis; the lack of attenuation and the low degree of attenuation may partly justify the deterioration of the health of vaccinated animals. On the other hand, this study confirms the non-interference of EVALON® in productive parameters, as well as the absence of impact in the birds’ intestines. In addition to the durable protection offered by EVALON®, these factors put the birds in a better position to achieve the objectives of uniformity and weight during the rearing period.

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### 3 Results

There were no clinical signs in groups 1, EVALON® and 4 throughout the study. Mild clinical signs, such as the presence of ruffled feathers and less activity, were observed in groups 3 and 5 (treated with non-attenuated vaccines). In addition, mild clinical signs were also observed in group 5, such as paleness in the legs and the crest, the body slightly arched and huddling. The duration of the mild clinical signs was variable. These manifestations were detected in group 3 for 2 days only, while in group 5 they lasted from 15 to 43 dpv.

Sporadic changes in the appearance of the droppings were observed in the EVALON® group and group 4, whereas in groups 3 and 5 the changes occurred almost continuously from the second or third week after vaccination.

There were no statistically significant changes in body weight at any time during the study (Mann-Whitney U Test; p <0.05); however at 37 dpv, the weight in control group 1 and the EVALON® group reached its maximum value. These results are consistent with what was expected with EVALON®: since it is an attenuated vaccine, its replication is fast with a peak at 21 dpv, leaving the digestive tract clear so that the animals can continue to grow without interferences linked to the vaccine strains (Figure 1).

Throughout the study, no statistically significant differences were observed in the EVALON® group (Mann-Whitney U Test; p <0.05) in the average of the intestinal lesions compared to the control group (group 1). In contrast, there were statistically significant differences (Mann-Whitney U Test; p <0.05) in the lesion score average in different intestinal regions in groups 3 to 5 compared to the control group. In particular, in group 3 lesions were observed in the rectum at 28 dpv. In group 4 lesions were observed in the duodenum and the jejunum at 28 dpv; in the rectum, 37 dpv. In group 5 lesions were observed in the rectum at 28 dpv and in the caeca at 37 dpv. The Total Mean Lesion Score is the sum of the average lesion values for the different intestinal tracts in a number of birds (20 in this study).

Eimeria oocysts were detected in the litter of all vaccinated groups; however, in group 4, they appeared later compared to the other groups. The number of oocysts was higher in groups 3 and 5 compared to the other groups; the differences reached 14.9 × 10⁴ oocysts/g at day 7 (figure 3). However, after day 7, both groups 3 and 5 showed an evident decrease of oocysts per gram (OPG) in the litter, which may be due to the fact that, under experimental conditions, the densities of the individual animals are lower compared to field conditions and this may affect the correct distribution of vaccine oocysts. The EVALON® group behaved as expected with regard to OPG: since it is an attenuated vaccine, replication never reaches levels as high as that of the non-attenuated vaccines (groups 3 and 5) and, in addition, the profile of the OPG curve shows the peak of the first replication at 7 dpv, and the second peak at 21 dpv, which corresponds to the second replication, after which the animals develop immunity against coccidiosis.
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Eimeria oocysts were detected in the litter of all vaccinated groups; however, in group 4, they appeared later compared to the other groups. The number of oocysts was higher in groups 3 and 5 compared to the other groups; the differences reached $14.9 \times 10^4$ oocysts/g at day 7 (Figure 3). However, after day 7, both groups 3 and 5 showed an evident decrease of oocysts per gram (OPG) in the litter, which may be due to the fact that, under experimental conditions, the densities of the individual animals are lower compared to field conditions and this may affect the correct distribution of vaccine oocysts. The EVALON® group behaved as expected with regard to OPG: since it is an attenuated vaccine, replication never reaches levels as high as that of the non-attenuated vaccines (groups 3 and 5) and, in addition, the profile of the OPG curve shows the peak of the first replication at 7 dpv, and the second peak at 21 dpv, which corresponds to the second replication, after which the animals develop immunity against coccidiosis.
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References