

## ABSTRACT

**Title** : **Development and evaluation of *Eimeria tenella* sporozoite vaccine in broiler chickens**

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In the present work on development and evaluation of *E. tenella* sporozoites vaccine from field isolates of caecal coccidiosis in broiler chickens, the protective immune response and efficacy of the vaccine were assessed by ELISA and IFN- $\gamma$  assay, and challenge studies, respectively. The purity of the sporozoite protein from the field isolates revealed the molecular weight in the range of 30, 97 and 140 KDa by SDS-PAGE which was used as a potential vaccine candidate.

The experimental study was carried out with 130 day old Cobb 400 broiler chicks which were divided for two experimental trials, each comprising five treatment groups. In the experimental trial-I, the groups, T1 and T2 were administered 10 and 20  $\mu$ g of live sporozoite antigen, respectively on 2<sup>nd</sup> day of age, and T3 and T4 were administered 10 and 20  $\mu$ g of

live sporozoite antigen, respectively on 6<sup>th</sup> day of age. In experimental trial-II, T1 was administered 25 µg of live sporozoites as primary and booster doses on 2<sup>nd</sup> and 18<sup>th</sup> day of age, respectively and T2 with 25 µg of live sporozoites with FCA, as primary vaccination on 2<sup>nd</sup> day and booster vaccination without adjuvant on 18<sup>th</sup> day of age. T3 was administered live anticoccidial vaccine (Livacox-Q®) at the recommended dosage on the 2<sup>nd</sup> day of age and the group T4 was administered the anticoccidial drug (diclazuril) via feed at the recommended dosage from 2<sup>nd</sup> day of age, till the end of experiment. Group T5 was kept as control in both the trials I and II.

Humoral (IgG) and cell mediated immune response (CMI) were assessed by sporozoite specific indirect ELISA and gamma interferon assay, respectively from day 7 of life at weekly intervals. Significantly high (IgG) antibody levels ( $P<0.01$ ) were observed in group T4 with a ELISA mean OD value of  $0.399\pm 0.016$  at 35 days followed by a decline of  $0.316\pm 0.016$  at 42 days of age in experimental trial-I. However, in experimental trial-II, the antibody levels (IgG) were significantly high ( $P<0.01$ ) in group T2 (mean OD of  $0.616\pm 0.022$ ) at 42 days and significantly high ( $P<0.01$ ) in group T3 at 49 days of life. The mean IFN- $\gamma$  concentration (in pcg) was significantly high ( $P<0.01$ ) in group T4 ( $226.61\pm 6.41$ ) with a decline after 14 days of age in experimental trial – I and significantly high ( $P<0.01$ ) in T2 ( $681.39\pm 39.22$ ) with a steady and better concentration of IFN- $\gamma$  in T1 and T2 than other groups in experimental trial-II.

The mean weekly weight gain in chickens in experimental trial-I was higher in group T4 with a relative ratio of 47.81 per cent and a low feed conversion ratio of 2.18 after challenge. Whereas, in experimental trial II, the mean weekly weight gain was higher in group T2 than in T1 and T4 and lower than in T3, with a relative ratio of 83.0 per cent. However, the mean total weight gain was significantly higher ( $P<0.01$ ) in group T2 than in group (T3) which received the commercial vaccine and the feed conversion ratio after challenge was low in T3 (2.04).

The lesion decrease ratio was high in group T4 (24.92%) with no mortality in trial-I and significantly low ( $P<0.01$ ) in group T2 (58.04%), but inferior to group that received the commercial vaccine (T3) in trial-II. The protection percentage in terms of oocyst output was significantly low ( $P<0.05$ ) in group T4 (51.21%) and in T2 (65.39%) in trial-I and II, respectively.

In terms of safety, potency and efficacy, the developed vaccine administered with FCA adjuvant showed no significant difference ( $P>0.05$ ) in weight gain, no untoward reactions like transmission of oocysts via faeces, clinical disease or mortality before

challenge and had significantly better ( $P<0.01$ ) protective response and anticoccidial index than controls. The efficacy of candidate vaccine was superior to that of commercial vaccine in terms of the total weight gain, however, inferior in terms of mean weight gain after challenge, FCR, oocyst decrease ratio, lesion decrease ratio and ACI. Whereas in comparison with the commercial anticoccidial, the FCA adjuvanted sporozoite vaccine was superior in terms of weight gain, FCR, lesion decrease ratio, oocyst decrease ratio and ACI.