EFFICACY OF MILBOND TX® AGAINST THE MYCOTOXIN'S EFFECT ON THE CELLULAR IMMUNITY RESPONSE VACCINE OF BROILER[A]

Santos, I.L.[1]; Muniz, E.C.[1]; Holtz,L.[1]; Fávaro Junior, C.[2]; Beirão, B.C.B.[2]; Ingberman, M.[2]; Fernandes Filho, T.[2]; Caron, L.F.[2]; Barbosa, T.[3]; Osuna, O.[4]

[1] LABORATORIO ZOETIS, SÃO PAULO,SP, BRAZIL [2] IMUNOVA, CURITIBA, PR, BRAZIL.[3] ZOETIS GLOBAL POULTRY, DURHAM, NC, SA [4] MILWHITE, BROWVILLE, TEXAS, USA

INTRODUCTION

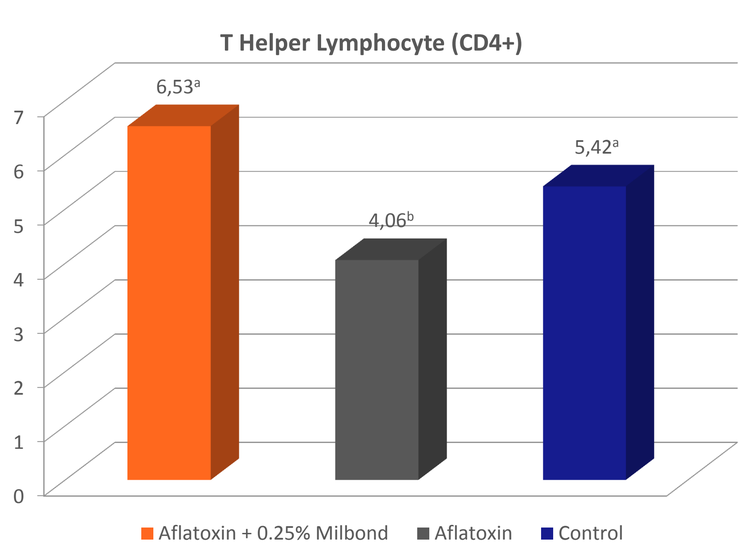
Aflatoxin is metabolite of *Aspergillus flavus* and it is potent liver toxins and carcinogens in animals and humans. Mycotoxins can interfere with the production of cytotoxic and helper T lymphocytes in the thymus directly through the altered metabolism of lipids. T Helper Lymphocyte (CD4+) are the cells responsible for steering the responses of the adaptive immune system. These cells are present in peripheral circulation and then migrate to the secondary lymphoid tissues in the presence of a challenge. Mucosal T Helper lymphocytes are cells present in these locations that act aiding in the production of antibodies or in defending against challenges. Aflatoxin intoxication has been reported as a cause of immunosuppression and its effects can affect the vaccine immune response. The measures used by the industry to protect the animals from the toxic effects of AF include assessment of grain, use of inhibitors of fungal growth, fermentation, microbial inactivation, physical separation, thermal inactivation, irradiation, use of ammonia, ozone degradation and the use of adsorbents. Currently, one of the most auspicious and practical approaches is the use of adsorbents. Selected adsorbents when added to diets contaminated with AF can hijack the aflatoxin during the digestive process, allowing the mycotoxin to pass through the gastrointestinal tract of the animal without harm Milbond-TX® is a commercial available mycotoxin binder used to diminish aflatoxin effects. The objective on this study was to evaluate the Milbond TX® influence in the vaccine immune response in broiler.

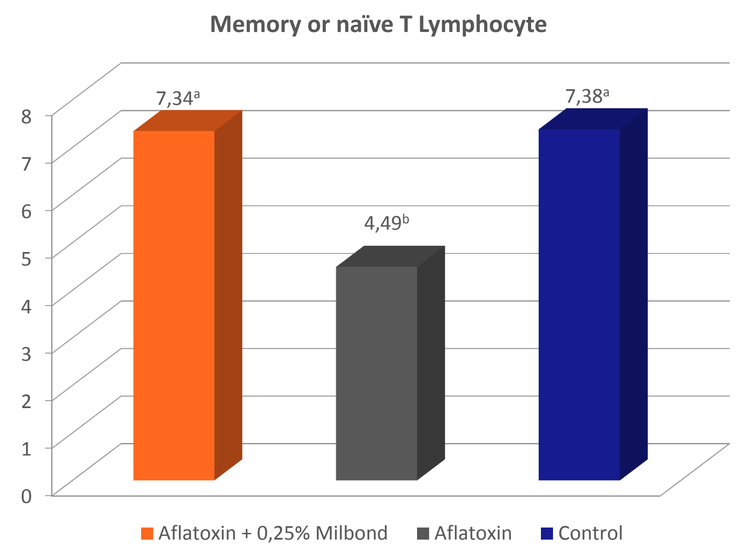
MATERIAL AND METHODS

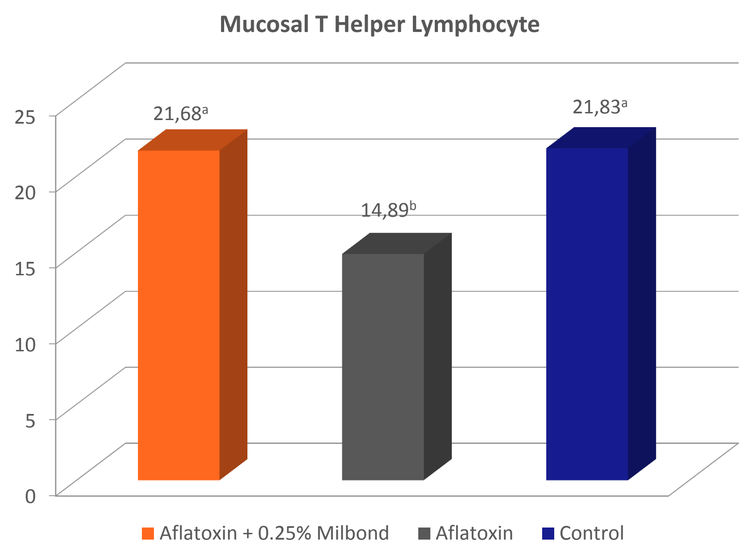
90 male broilers with same origin were vaccinated with Newcastle Disease virus (Poulvac® NDW) at 1 day of age and, divided into 3 treatments of 30 birds each. Treatment differences were the type of feed given to the different groups. The treatments consist of the Negative control (T1), regular broiler feed only, Positive control (T2), and where at the regular feed was included 2.8 ppm of Aflatoxin and the Milbond group (T3), which included the aflatoxin (2.8 ppm) and Milbond at 0.25 %. Animals were kept in cages with 10 birds each (3 replicates per treatment). Blood samples were collected at 3, 7 and 21 days post vaccination. Samples were immediately transported and process at the Imunova Biological Analysis Laboratory for Flow cytometry. The was technique used to quantify the presence for T Helper Lymphocyte (CD4+), Mucosal T Helper Lymphocyte and Memory or naïve T Lymphocyte in individual samples. This procedure is currently an established method to study the animal immune system. Flow cytometry was performed on a FACSCalibur flow cytometer (Becton Dickinson). Green fluorescence (from FITC) was detected on the FL1 channel (530/30 nm), and orange fluorescence was detected on the FL2 channel (585/42 nm). Cells were analyzed at up to 10,000 events in the lymphocyte gate (based on forward and side scatter, including contaminating thrombocytes. Data were analyzed with FlowJo software (TreeStar, Inc). Statistical analyses was for each type of cell and day by generalized linear mixed model (Poisson distribution and log link) with treatment fixed, and sample random.

RESULTS AND CONCLUSIONS

The flow cytometry technology allows the characterization of the immunological status, enabling the evaluation of the mechanism of action of certain vaccines. Small interferences on the immune system can thus be detected with great sensibility, predicting how the immune system has been able to mount a response facing the vaccination. The Immune response evaluated by flow cytometry, mapping 3 different cell subpopulations (T Helper Lymphocyte (CD4+), Mucosal T Helper Lymphocyte and Memory or naïve T Lymphocyte), showed that the Milbond TX® fed group and the negative control group had significantly greater cellular immunity response than Aflatoxin group. All evaluated cell subpopulations were reduced in aflatoxin group compared to controls, whereas Milbond TX® helped to numerically reduce such deleterious effects (results not shown). The addition of Milbond TX® in feed led to increase the number of lymphocytes, at similar levels observed in the unchallenged control group, while the groups which received only the challenge had the lymphocyte count decreased (Figure below). The presence of mycotoxin alters the normal development of the immune system of birds tested in this experiment. The use of additive anti mycotoxin Milbond TX® was able to reverse the effects of mycotoxins in relation to vaccine response. We conclude that Milbond TX® helped to protect the immune response when birds were fed aflatoxin positive feed, which could help to a better adaptive immune response after Newcastle Disease vaccination.







Different letters in the same graphic indicate significant differences p≤0.05.

REFERENCES

Beirão, B. C. B., Fávaro Jr, C., Nakao, L. S., Caron, L. F., Zanata, S. M., & Mercadante, A. F. (2011). Flow cytometric immune profiling of specific-pathogen-free chickens before and after infectious challenges. *Veterinary Immunology and Immunopathology, In Press.*

Chowdhury, S. R., Smith, T. K., Boermans, H. J., Woodward, B., & Chowdhury SR. (2005). Effects of feed-borne Fusarium mycotoxins on hematology and immunology of laying hens. *Poultry Science, 84(12), 1841–1850.*

Girish, C. K., & Smith, T. K. (2008). Impact of feed-borne mycotoxins on avian cell-mediated and humoral immune responses. *World Mycotoxin Journal, 1(2), 105–121.*

Jung, B.-G., Toan, N. T., Cho, S.-J., Ko, J., Jung, Y.-K., & Lee, B.-J. (2010). Dietary aluminosilicate supplement enhances immune activity in mice and reinforces clearance of porcine circovirus type 2 in experimentally infected pigs. *Veterinary microbiology, 143(2-4), 117–25.*

Tessari, E. N. C., Oliveira, C. A. F., Cardoso, A. L. S. P., Ledoux, D. R., & Rottinghaus, G. E. (2006). Effects of aflatoxin B1 and fumonisin B1 on body weight, antibody titres and histology of broiler chicks. *British Poultry Science, 47(3), 357–364.*

[A] Data on file, Study Report No. 03-13-70AQO, Zoetis Inc.