Vet.Med.J.,Giza. Vol.41,No.1.(1993):33-37.

PRELIMINARY STUDY ON FORMULATION AND APPLICATION OF AFLATOXINS- ANTIDOTE

BY

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(Received: 16.1.1993).

SUMMARY

Thorough looking to aflatoxin metabolic pathways, mode of action, its physical and chemical properties leads to suggest a formula of aflatoxins-antidote. The 9 fine and well mixed components of such formula were packaged into gelatin capsules, each contain 1 gm of this proposed aflatoxins-antidote. Fortunatelly, also a highly aflatoxin B_1 contaminated finisher ration was available. Chemical assay showed that the concentration of aflatoxin B_1 was proved to be 250 ug/each kg finisher feed. To testify the validity of this appropriate aflatoxins-antidote, 200 lohman broilers aged 4 weeks and weighed approximately 1065 gms, each were obtained and randomly distributed into equal four groups. Two groups only were received single oral capsule daily for 30 successive days, meanwhile they were fed on aflatoxin B1 contaminated ration contained 250 and 125 ug/kg feed for the high and low level treated groups, respectively. The other two groups were identified as positive and negative control groups. The negative control group fed on aflatoxins free ration, while the positive control group fed on a ration of 250 ug aflatoxin B_1/kg, both control received no aflatoxinsantidote. Data showed that the performance of both treated groups showed an abvious resistance to aflatoxin B_1-contamination. The average total body gain of both high and low level treated groups were 93.8 and 104.8% relative to the negative control group, while the corresponding figur obtained from the positive control group reduced up to 25.9% after 30 successive days of feeding on aflatoxin B_1 contaminated ration. Similarly, the percentages of average feed conversion of both high and low level treated foups showed 104.3 and 94% relative to negative control, while the corresponding figur obtained from positive control was 353%. In other words, consuming 2244 and 2262 gms aflatoxin B_1 contaminated ration resulted in body weight gain of 800 and 894 gms in the two treated groups which received daily capsules of aflatoxins-antidote, while the average body weight gained from positive control consuming 2106 gms was 221 gms after the same period. Its worthy to mention that no deaths had occurred during this trial neither in treated nor control groups. Such data needs further research work to approve and/or to reject the validity of the appropriate antidote considering the variables of specie response, sex, age, physiological and nutritional status... etc.

INTRODUCTION

Both human and animal health has been dramatically affected in outbreaks of acute mycotoxicosis, but these targic events may by only a part of the cost to society in terms of impaired health and productivity from the ingestion of sub-clinical levels of mycotoxins (WHO, 1981). Recent stud-

ies have indictated that the differences in response to aflatoxins in different livestock species has been attributed to their differential metabolism (Park, 1983). The rate of metablism, the repeativeness of exposure, dosage and type of aflatoxins are important factors in determining the type of toxic action of aflatoxins (Hsieh, 1983). So far, the net work of research on mycotoxins and mycotoxicosis was focused on methods of detection, surveys, inhibition and promotion of mould activity...etc. But, regading our best of knowledge, there are few if any attempts to synthesize mycotoxins-antidote (Saad, 1991). So, this work aims to formulate an aflatoxins-antiote based on digestion, absorption, detoxification and elimination of such contaminants inside the body, then applying the appropriate formula in controlled experimental trial.

MATERIAL AND METHODS

Aflatoxins-antidote (formulation, dosage and application):

The components and the quantities included in this appropriate formula were suggested considering aflatoxins metabolic pathways as reported by Hsieh (13), Raina et al. (1991) and Saad (1991).

An empty gelatin capsules were purchased from El-Gomhoria Pharma. Inc., each was filled with 1 gm of the above mentioned well-mixed components. These capsules were administered orally once per day to both treated groups which fed on aflatoxin B_1 contaminated rations.

Aflatoxin B_1 contaminated ration:

A finisher ration of broilers showed high concentrations of aflatoxin B_1 was available. Analytical methods of analysis using HPLC confirmed that the level of contamination was 250 ug aflatoxin B_1 per each kg of feed (AOAC, 1980). The nutritive value of each kg of this ration was 17.5% digested protein, 1975 kcal metabolizable energy, while its content of Ca, P, lysine and methionine were 0.8, 0.45, 0.85 and 0.36%, respectively. Also, vitamins and minerals were within the range recommended by the legalised issue 554 (1984).

Experimental animals (grouping and management):

Two hundred of Lohman white broilers aged 4 weeks and weighted about 1065 gms, were obtained after receiving their starting and growing rations. the birds were randomly distributed into 4 groups each of 50 heads. The 1st group was negative control which was fed on aflatoxins-free ration, while the 2nd group was positive control received aflatoxin B_1contaminated ration the was containing 250 ug/kg. Both the treated groups received single oral capsule (1gm) daily of aflatoxins-antidot. One treated group was fed on the available contaminated ration the was containing 250 ug aflatoxin B1/kg, while the 2nd group were fed on a ration consisted of 50% available contaminated ration + 50% of aflatoxins-free ration which was equivalently contain 125 ug aflatoxin b1/kg mixed feed. Chean drinking water and light were available for all groups all over the day.

Table (1). The composition of aflatoxins-antidote (qualitative and quantitative).

No.	Components	Concentrations/ kg.
1.	Vitamin A	3.000.000 IU.
2.	Cholecalciferol	1.000.000 IU
3.	Choline chloride	150 gm
4.	Methionine	50 gm
5.	Glutathione	50 gm
6.	Ascorbic acid	25 gm
7.	Zinc sulphate	20 gm
8.	Folic acid	75 mg
9.	Cyanocobalamine	4 mg
10.	Calcium carbonate	up to 1 kg.

Body weights and feed consumption all groups were checked every 3 days up to 30 successive days, then the average of both daily intake and feed conversion were calculated either absolutely or relativly to average negative control.

RESULTS AND DISCUSSION

Table 2. Average body weight gain (gms.) of the different groups.

Age	Control		Treatments	
	Negative	Positive	250 ug B_1/Kg	125 ug B_1/Kg
0 time	1070	1060	1065	1070
3 rd day	1140	1100	1110	1115
6 th day	1190	1142	1176	1180
9 th day	1250	1120	1240	1220
12 th day	1310	1160	1290	1280
15 th day	1400	1140	1380	1390
18 th day	1490	1190	1450	1480
21 th day	1580	1220	1550	1610
24 th day	1700	1280	1680	1730
27 th day	1810	1272	1791	1905
30 th day	1923	1281	1865	1964
Total gain	853	221	800	894

It's worthy to mention that no deaths had been occurred during this trial neither in control group nor in treated one. considering that the average body weight gain of negative control group (110%), data showed that the corresponding figure obtained from negative control group represented only 25.9%. This obvious losses of average body weight gain was mainly due to the repeatitiveness of feeding on a finisher ration contminated with 250 ug aflatoxin B_1/kg for 30 successive days. These findings were in agreemer with those reported by **Wogan (1967) and Rainer et al. (1991)**, who reported that ingestion of sub-clinical aflatoxins resulted in slow growth and weight loss, considering the dosage of contamination and the repeatitiveness of exposure to such diets. On the other hand, the high and low level treated

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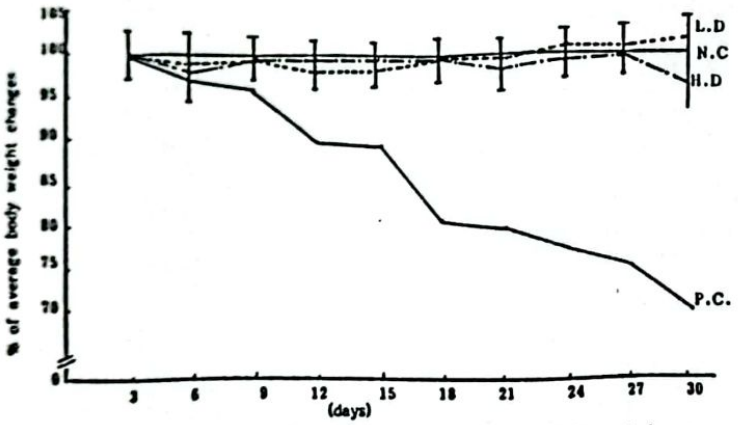

Fig.1: The percentages of average body weight changes of both control groups and treated groups.

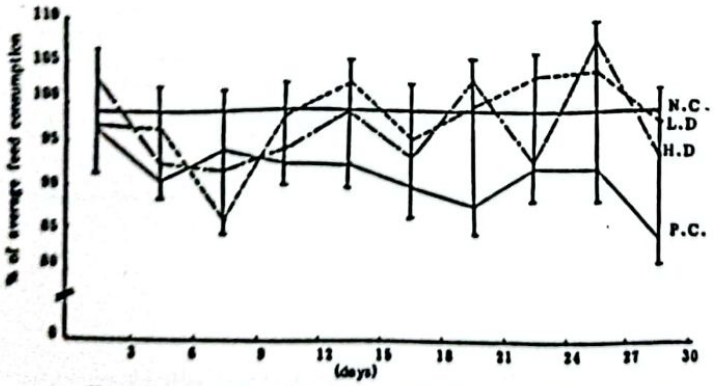

Fig.2: The percentages of average feed consumed by the different groups during the experiment.

groups which received orally single capsule of aflatoxins-antidote once a day, meanwhile they were fed on contaminated ration containing 250 and 125 ug aflatoxin B_1/kg respectively, both showed more or less the same body weight gain of negative control group (-6.2 & + 4.8%). Comparing the average body weight gain obtained from positive control group (221 gm) and high level treated group (800 gm), which all fed on contaminated ration of 250 ug aflaoxin B_1/kg for 30 successive days, it's accepted to mention that such obvious effect was mainly due to aflatoxins-antidote. Many reports exhibited that vitamin A, glutathione, Zn and ascorbic acid had some effects on such cellular systems such as the structure of cell membranes and cell particles as well as on certain microsomal enzymes responsible for cell metabolism and interactions of alephatic compounds. Undoubtedly, vitamin A and cyanocobalamine had wide effect on the symptoms caused by impairing food utilization , e.g. show growth and weight loss (Bains, 1979 and Raina et al., 1991). Moreover, feeding on a ration containing 125 ug aflatoxin B_1/kg and receiving daily capsule of aflatoxins antidote exhibited 104.8% body weight gain after 30 days relative to negative control group. Such improvement might be due to some components of aflatoxins-antidote which had certain effect on growth (Table 1). However, such promising data leads to recommend this unique aflatoxins-antibote because of its double action, 1) minimizing aflatoxins errects and 2) maximizing feed utilization.

ACKNOWLEDGEMENT

My deepest appreciation to Dr. Akram Shoukfa (Ph.D) the general Manager of El-Mansoria Co. for Agric. Projects, for his sincere help facilitating this work.

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