Hematology of Mercury Toxicity in Young Chickens[1]

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Dietary mercury (Hg) when administered in the form of mercuric chloride is known to cause several physio-pathological changes in avian species. Among these changes are growth reduction accompanied by decreased feed and water efficiencies (PARKHURST and THAXTON, 1973), gross changes in the sizes of several vital organs (THAXTON and PARKHURST, 1973b), decreased immunological responsiveness (THAXTON and PARKHURST, 1973c), eggshell thinning (TEJNING, 1967; STOEWSAND et al., 1971), abnormal mating behavior coupled with decreased reproductive efficiency in both sexes (THAXTON and PARKHURST, 1973a), and neurological dysfunction (FEMREITE and KARSTAD, 1971). However, the effects of dietary Hg on the cellular hematology of avian, as well as mammalian, species remains unknown. The purpose of this study was to determine the cellular hematological changes of chickens which were administered mercuric chloride during their juvenile development.

MATERIALS AND METHODS

Day-old broiler cockerels which were obtained from a commercial hatchery were randomly assigned to 24 groups of five chicks each. Three replicate trials were conducted. Hg in the form of mercuric chloride was added to the drinking water at the concentration of 0, 150 or 300 ppm of Hg. In each trial eight groups of five chicks received each concentration of Hg ad libitum from hatching until termination of the trial at six weeks. A commercial chick-starter ration which was formulated to provide adequate amounts of the essential nutrients was fed throughout the trials.

The chicks were maintained in electrically heated metal batteries for the first three weeks. The birds were then transferred to non-heated batteries for the remainder of the trials.

At intervals of 2, 4 and 6 weeks the chicks were weighed and then bled by veni-puncture. The blood samples were collected directly from the venal flow. Packed cell volumes (PCV) were determined by the capillary tube technique according to the procedure described by SCHALM (1965). Total erythrocyte and leucocyte numbers were estimated microscopically using a hemocytometer. The fresh blood samples were diluted with Natt-Herrick stain prior to quantitation (NATT and HERRICK, 1952). Additionally, blood smears were made, and following staining with Wright's stain, 300 leucocytes per sample were differentiated into the cells of the mature leucocytic series (LUCAS and JAMROZ, 1961). The differential numbers of each leucocyte were determined by calculating the absolute numbers, i.e., the numbers of a particular leucocyte-cell type were expressed as cells per mm^3 of blood.

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The data of each trial were evaluated statistically by the analysis of variance and significant differences which were attributable to trials were not found. Therefore, the data were pooled over trials and the differences among the treatment means were partitioned by the method of least significant differences. Statements of significance are based on $P \leq 0.05$.

RESULTS

The effects of the Hg-treatments on the growth of the chicks through six-weeks are expressed graphically in Figure 1. Significantly lower body weights were found at 2, 4 and 6 weeks in the chicks which received Hg at the level of 300 ppm. The body weights of the birds which received 150 ppm of Hg were lower at eacn of the bi-weekly periods; however, these differences were not significant.

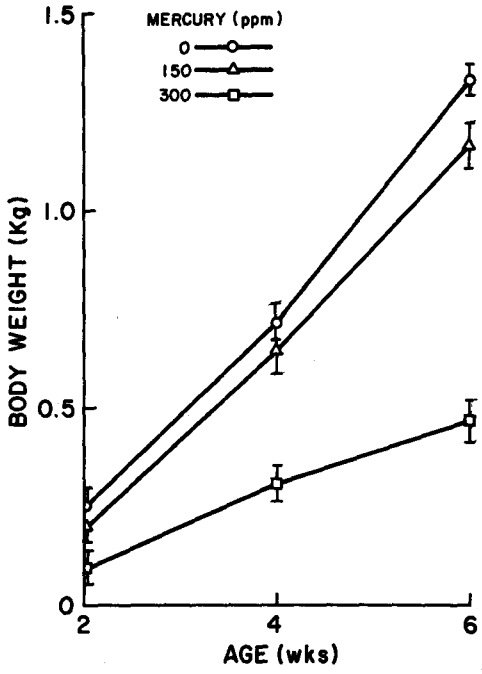

Figure 1. Effect of dietary mercury on the growth of broiler cockerels. The bars which accompany the means in all the Figs. represent standard error of the means.

In addition to inhibited growth, 300 ppm of Hg caused an increase in PCV at all times of measurement. These data are shown in Figure 2. The lower dose of 150 ppm of Hg did not significantly affect the PCV at 2 or 4 weeks, but this level of Hg did cause increased PCV at 6 weeks of age. As shown in Figure 3, the total circulating numbers of erythrocytes of the birds which received the growth inhibitory dose of 300 ppm of Hg corresponded to the PCV, i.e., significantly greater numbers of erythrocytes were found throughout the experimental period. The erythrocyte numbers of the 150 ppm birds did not differ from the controls at 2 or 4 weeks; however, these birds did have significantly less erythrocytes at 6 weeks of age.

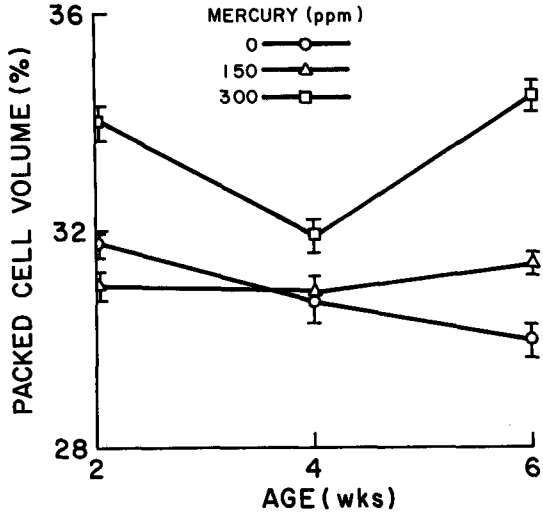

Figure 2. The effect of dietary mercury on the packed cell volumes of broiler cockerels.

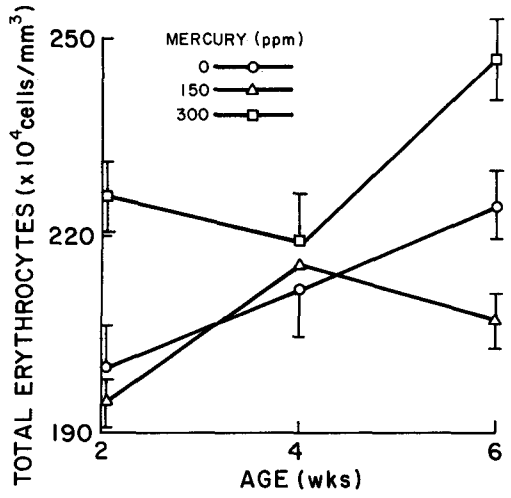

Figure 3. The effect of dietary mercury on the total numbers of erythrocytes in broiler cockerels.

The effects of the Hg-treatments on the total leucocyte numbers are presented in Figure 4. The trend was that the toxic level of 300 ppm of Hg resulted in decreased numbers of leucocytes. This effect was apparent at 4 and 6 weeks of age. Additionally, 150 ppm of Hg resulted in decreased leucocyte numbers at 2 and 4 weeks of age, but not at 6 weeks of age.

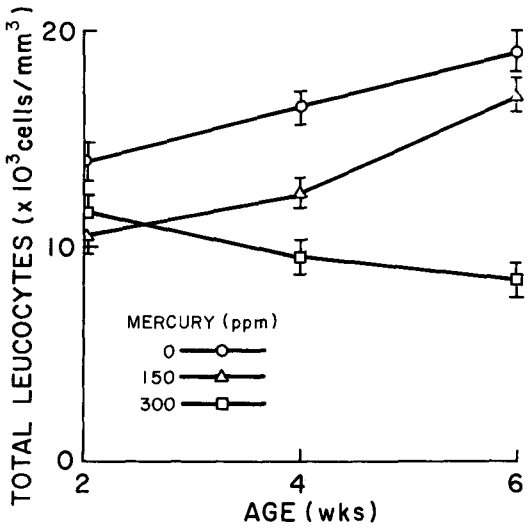

Figure 4. The effect of dietary mercury on the total numbers of leucocytes in broiler cockerels.

Figures 5 and 6 demonstrate the effects of Hg on the absolute numbers of lymphocytes and heterophils, respectively. Both 150 and 300 ppm of Hg resulted in significantly fewer numbers of circulating lymphocytes at 4 and 6 weeks. The effect of the Hg-treatments on the numbers of heterophils was not apparent until 6 weeks. At this time both doses of Hg caused significant increases in the numbers of heterophils. It should be noted that the greatest increase was attributable to the 300 ppm dose of Hg.

Although the data are not presented, neither dose of Hg caused significant alterations in the absolute circulating numbers of monocytes, eosinophils or basophils at any of the times at which the measurements were made.

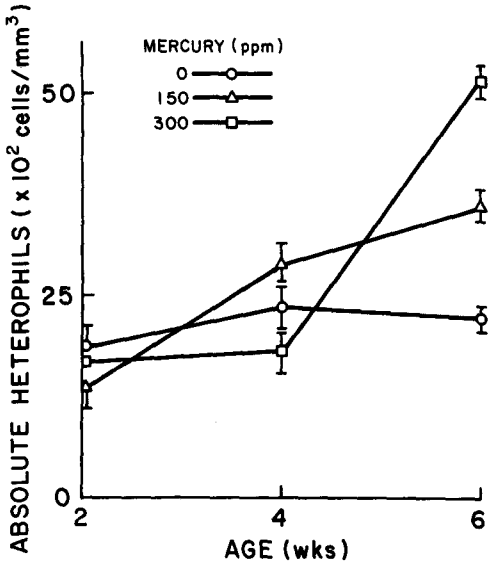

Figure 5. The effect of dietary mercury on the absolute numbers of lymphocytes in broiler cockerels.

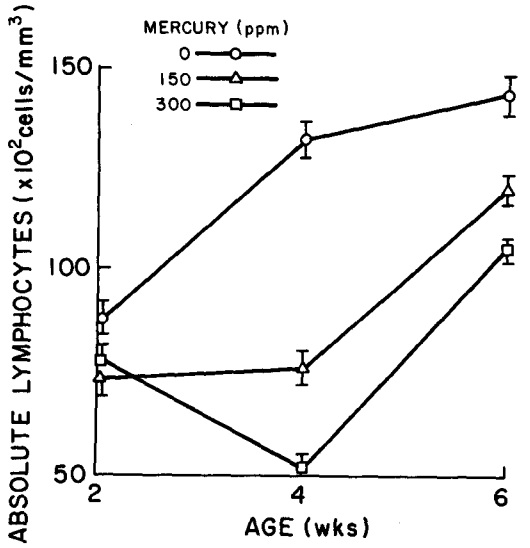

Figure 6. The effect of dietary mercury on the absolute numbers of heterophils in broiler cockerels.

DISCUSSION

We have reported previously that 250 ppm of Hg in the drinking water of young chickens caused a growth inhibition (PARKHURST and THAXTON, 1973). In the present study, 300 ppm was growth inhibitory while 150 ppm was not. HILL and MATRONE (1970) showed that 400 ppm of Hg in the form of mercuric chloride when added to the feed of young chickens caused a growth reduction. Since chickens normally consume greater quantities of water than feed, it appears that the results of these reports are in agreement.

The relationship of increased PCV concomitant with increased circulating numbers of erythrocytes in the birds which received the growth inhibitory dose of 300 ppm suggests that Hg either directly or indirectly enhanced erythropoiesis. However, the conflicting results in the birds which received 150 ppm is difficult to explain. Excessive numbers of abnormal erythrocytes and erythroblastic cells were not observed. An explanation of the Hg-induced erythrocytosis on the basis of a dehydratory hemoconcentration appears unwarranted, since in a previous study we demonstrated that juvenile broilers which experienced Hg-toxicity maintained a normal blood pH (THAXTON et al., 1973). Additionally, the circulating levels of uric acid, glucose, protein, total lipids and cholesterol of the birds were not affected significantly. The apparent Hg-stimulation of erythropoiesis is similar to that observed with toxic levels of cobalt (DAVIS et al., 1945).

The reduction in the total numbers of leucocytes suggests that rather than a decreased production of leucocytes, the circulating numbers of cells were decreased. This possibility is feasible because the reticuloendothelial system (RES), of which the leucocytes are component parts (ASCHOFF, 1942) is involved in the phagocytic removal of heavy-metal complexes (EVDOKIMOFF and WAGNER, 1972). Thus, circulating Hg-complexes, formed by interactions of Hg with sulfhydryl groups of proteins (CLARKSON, 1972), could have been actively phagocytised and account for the leucopenic condition in the toxic birds. The decreased numbers of lymphocytes simultaneous with the increase in heterophils suggests enhanced granulo-leucocytic involvement.

An equally attractive explanation for these results is that Hg evoked a physiological stress response. SIEGEL (1972) has reviewed the subject of stress in avian species and documented the associated leucocytic changes. Lymphopenia with heterophilia is the characteristic leucocytic response in birds which are exhibiting stress and, unlike in mammals, birds do not experience eosinophilia (SELYE, 1953). Our results are in agreement with the leucocytic changes which are characteristic of stress. Additionally, we reported that Hg-toxicity caused increased adrenal size, a reduction in the sizes of the bursa of Fabricius and an impaired ability to mount normal immune responses (THAXTON and PARKHURST, 1973b and c). These physiological changes are also characteristic stress-indicators in birds (SIEGEL, 1972). The results of this study could be interpreted to indicate that a generalized stress condition existed in the birds which received the toxic level of Hg.

These results expand the considerations of Hg-toxicity in birds to include the hematological system. However, it remains unknown whether Hg caused the hematological alterations by acting directly on the hematologically associated tissues or by covertly affecting interrelated systems.

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