Vet.Med.J.,Giza. Vol.43,No.4.(1995): 421-427.

EFFECT OF FLUORIDE TOXICITY ON BROILER CHICKEN CHROMOSOMES

By

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SUMMARY

The effect of fluorine on mitotic chromosomes of broiler chicken (60 day old, was investigated. Broielr chickens were exposed to sodium fluoride in ration in two doses (1000 ppm and 1500 ppm) for two months, after which, metaphases from the bone marrow were prepared,stained and exaimed for chromosomal aberrations. Exposure of broiler chicken to fluorine revealed an increase in the frequency of abberant metaphase which reached 11.34%, 5.37% and 0.8% in groups exposed to 1000, 15000 and control respectively at the first month, while the frequency reached 6.10, 5.12 and 0.9 in groups exposed to fluorine in a dose of 1000, 1500 and 0.0 (control) respectively at the second month.

The obtained results illustrated the hazardous effect of fluoride pollution on broiler chicken - genome.

INTRODUCTION

Fluoride is considered as one of the important environmental pollutants in Assuit, Egypt as a result of the presence superphophate factory at Manquabad / and has a severe dangerous effect on bilogical life. Of particular hazard are factors possesing mutogec properties, since they can cause hereditary diseases and changes in the structure of natural or even the exinction of certain species.

In recent years, it has been proposed that cancer cases are high in areas with fluoridated water (Burk and Yiamouyiannis, 1975).

On the other hand many additional epidemiological studies provide no evidence for an association betwecn fluorides ingestion and m·'tality from cancer or other diseases in human (W.H.O., 1970) Mohamed and Chandler (1970) reported a damage in mice chromosomes when given sodium fluoride for 3 weeks otherwise , no significant increase in chromo[illegible]nal aberrations was observed in animals with high NaF intake using different levels up to 1000 ppm for six weeks (Martin et al., 1978).

A series of test systems to determine the genetic effects of environmental pollutants have been described, chromosome aberration test in culture and/or in mammalian marrow are used to investigate the genetic risk (Dubinin and Kalinina, 1980).

The previous contradictive results pushed our ideas to study the effect of NaF (sodium fluoride)

in high doses in hens in hens for a short period of exposure.

MATERIAL AND METHODS

Forty eight broiler chickens (Balady) from Manfaloat (60 days old) were treated with NaF through ration containing 1000 and 1500 sodium fluoride for 2 months.

1- Chromosome preparations:

Chickens were inoculated with colchicine intraperitoneally sacrificed after two hours , colchicine is applied to arrest mitotic metaphases, bone marrow was flushed with KCI/) 0.075 M, incubated at 37°C for 30 minutes, centrifuged and the sediment was fixed with methanol acetic acid (3;1), centrifuged and the supernatant was discarded, washed 3 times, two to three droplets were put on the slide and dried at 37°C overnight, (Tjio and Levan, 1956).

2- Staining:

The slides used for chromosome aberrations were stained at room temperature for 10 min with 10% Giemsa in phosphate buffer (pH 6.8), rinsed in distilled water, air dried and mounted in Entelan (Merk).

3- Scoring or chromosome aberrations:

Metaphases were examined under an oil immersion lens at a magnification of 1000 x . Only methaphases with well spread chromosomes were used for scoring chromosome aberrations. Some of these metaphases were photographed using copex pan film (Agfa Gevaert, Belgium).

4- Statistical analysis:

The significance of the results from treated chicken and control was tested according to two tail t-test (Kalton, 1976).

RESULTS AND DISCUSSION

Table 1 shows the results obtained from examining metaphases cells from bone marrow of both control and treated broiler chickens used in the experiment . From 122 and 330 untreated metaphases 3 and 6 cells showed chromosome aberrations after 1 and 2 months respectively. This frequency of chromosomal aberrations in the present experiment was in normal manner for control groups.

At the first month, out of 326 and 186 scored metaphases from the broilers exposed to 1000, and 1500 ppm of NaF in ration, 37 and 10 aberration metaphases were found which constitute 11.35% and 5.38% of the examined cells respectively. This frequency is nearly 3-4 fold that obtained in the control. The increase in aberration frequency is statisticaly highly significant ($P < 0.01$), Table 1.

In the second month, exposed broiler chickens had 13 and 6 cells out of 210, 117 and 330 scored metaphase that exhibited chromosomal aberrations for groups of broiler chicken exposed to 1000 and 1500 ppm sodium fluoride in ration respectively. These percentages of aberration (6.19, 5.13 and 1.82%) respectiveiy are approximately three times that of the control with highly significant differences ($P<0.01$).

Table 1: Type and frequency of chromatid and chromosome type abnormalities in control and broiler chicken exposed to sodium fluoride in ration (1000 ppm, 1500 ppm) for 2 months.

Time post exposure	Dose	No. of examined cells	No. of Metaphases	Mitotic index	Metaphases with aberration		Aberration									
							Single chromatid gap		Dots		Single chromatid break		Plodihloy		Other types	
					No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
One month	1000 ppm	8000	326	4.08	37	11.35**	2	0.60	14	4.30	4	1.27	15	4.60	2	0.60
	1500 ppm	5000	186	3.72	10	5.38**	1	0.53	1	0.54	0	0.00	2	1.07	6	3.22
	Control	3000	122	4.06	3	2.45	0	0.00	3	2.45	0	0.00	0	0.00	0	0.00
Two month	1000 ppm	3000	210	7.00	13	6.19**	0	0.00	1	0.47	11	5.23	0	0.00	1	0.47
	1500 ppm	3000	117	3.90	6	5.13**	0	0.00	2	1.71	2	1.71	0	0.00	2	1.71
	Control	13000	330	2.54	6	1.82	1	0.30	2	0.61	1	0.30	0	0.00	2	0.61

** Highly significant at P<0.01

Fig. (1): Satillite chromosomes

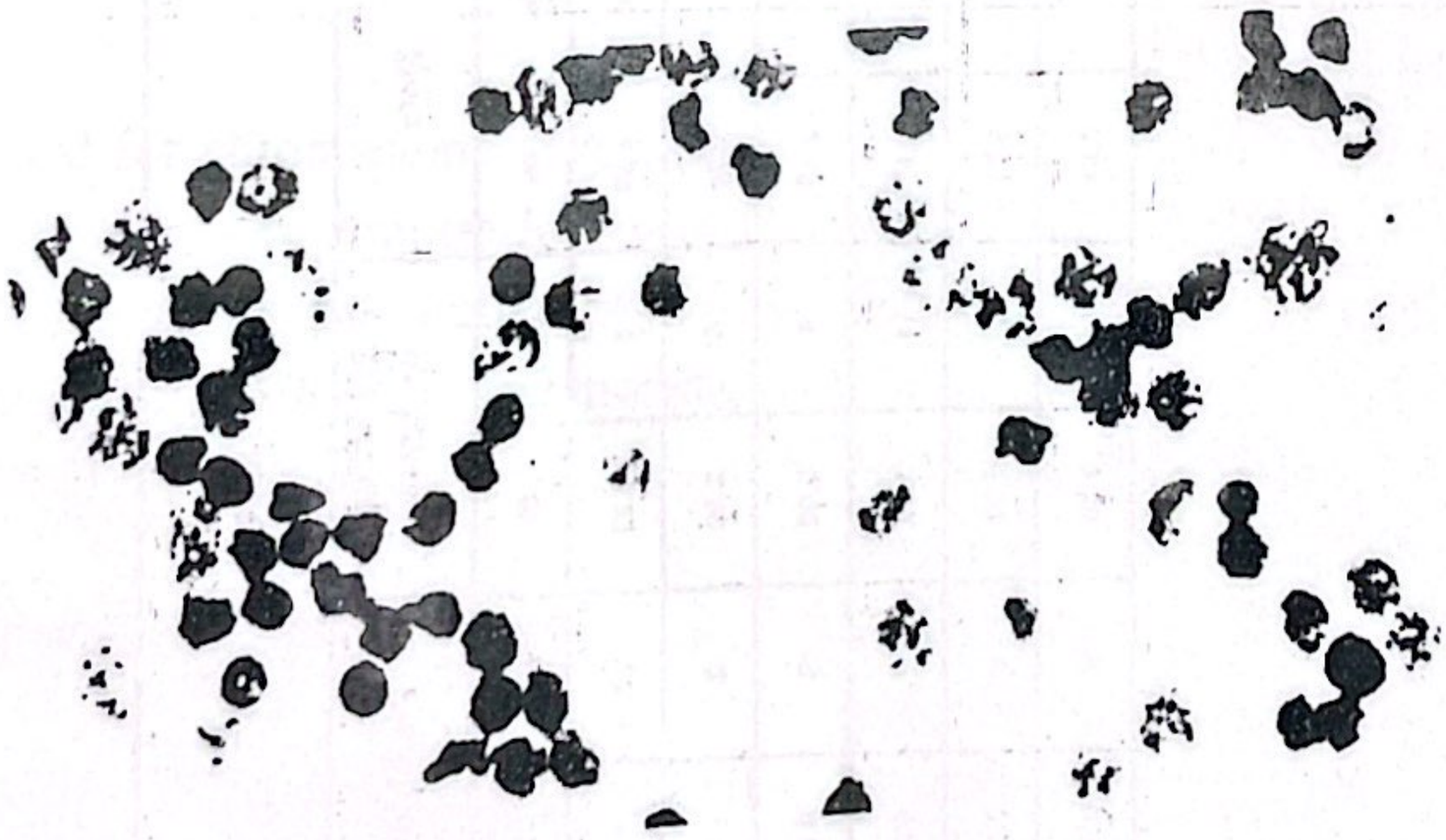
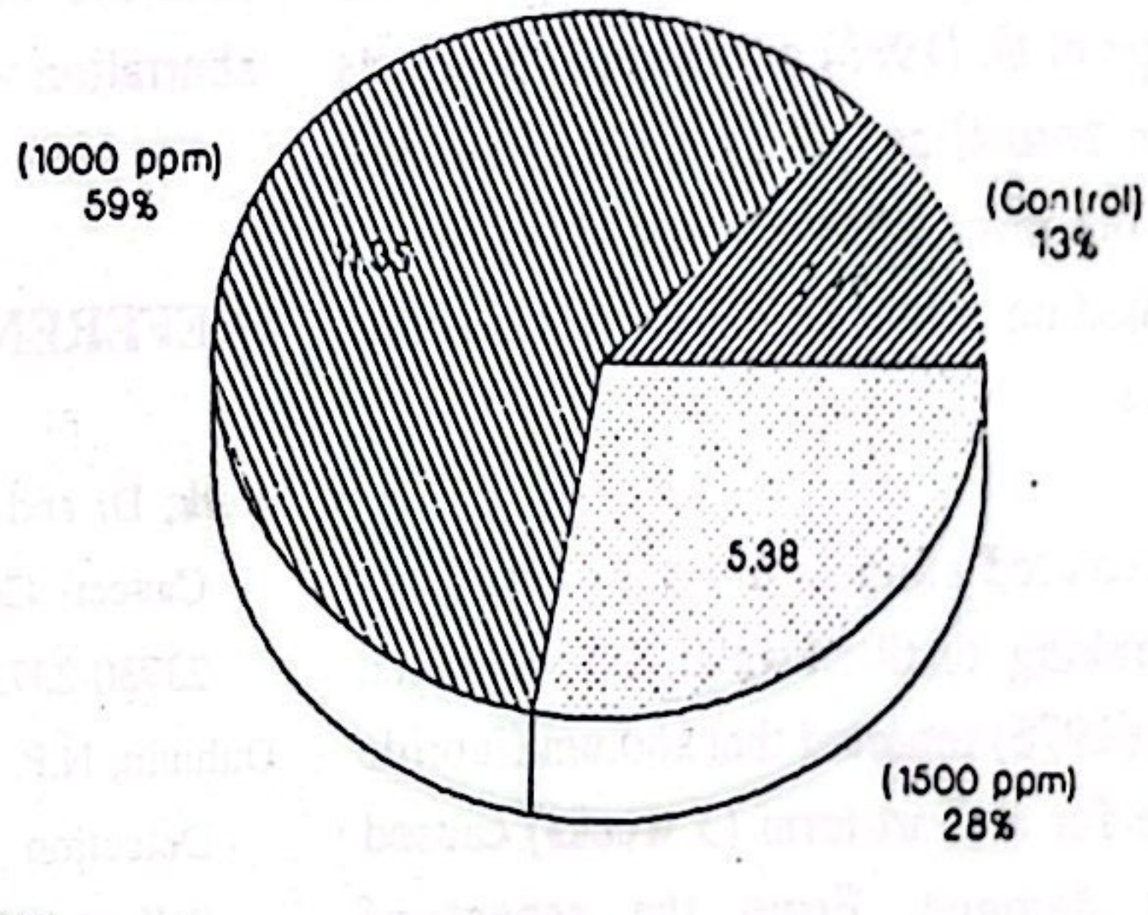


Fig. (2): Condensation chromosomes

Fig. 4: Percentages of chromatid aberrations in treated broiler chickens



At the early of mitosis each chromosome can be seen to have a centromere that binds two identical chromatides. The centromere attaches to the spindle fibre and within a short , all chromosomes migrate to the centre of the spindle apparatus, with condensation of the chromosomes in the peripheral site (Fig.2). These results agree with those of Shupe et al. (1983) who tested the effects of fluoride in fruit flies and on sister chromatid exchange in the mice bone marrow. They found that fluoride had no genotoxic effect on chrosome aberration rate

The fluoride treated animals did not differ from those mice drinking distilled water, but Mohamed and Cnandler (1976) reported that sodium fluoride given to mice for a short term (3 weeks) caused chromosome damage. From the aspect of extrapolating mutageniciy data are, that biotransformation pathways vary between species and individuals (Norpoth and Garner, 1980).
Chromosomes in chickens are small and of varying shape and size, hence they are difficult to count. Evidently they are 5 or 6 pairs of macro crmosomes (large) and 33 pairs of micro chromosomes (small) (North, 1984). In our study the most cases counted 33 pairs of chromosomes (Fig.1). Satillite chromosomes had been detected in case of chickens treated for 2 months with 1500 ppm. Also metacertic chromosome and submetacentric chromosome was detected in one case taken from group (1000 ppm) for 2 months (Fig.2).

Jagillo and Lin (1974) reported that in increase in sodium fluoride intake was followed by an increase in chromosomal aberrations in mice given 500 ug NaF intravenously or 250 ug NaF subcutaneously for 16 days.

In recent years however it has been proposed tha cancer rates are high in areas with fluoridate water (Burk and Yiamouiannis, 1975 an Yamouiannis and Burk, 1977). Chromosoma aberrations were present at similarity low leve (less than 1 present of the cells with aberration Also that no significant increase in chromosomal aberration was observed by Martin et al., 1979.

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