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The study of chromosomal aberrations in mice infested with radiated and non-radiated protoscolices of Echinococcus sanulosus .

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Abstract
The mitotic index (M1) and the chromosomal aberration in cells have been studied in white bulb mice subjected to radiated and non-radiated protosolices of E. granulosis . In this experimental infestation , gamma ray of 81.58416 x 10.3 wave length have been used. The protoscolices were subjected to a high degree of 25 Gy and low degree of 5 Gy. The results revealed no significant variation in both M1 and chromosomal aberration were found in bone marrow cells of infested mice in comparison to control.

الخلاصة:

تم خلال هذا البحث ايجاد معامل الانقسام الخلوي MIودراسة التغيرات الكروموسومية في خلايا نقي العظم لفئران بيضاء حقنت برؤيسات واكياس لطفيلي المشوكات الحبيبية مشعة بدرجة ٥ كري و٢٥ كري بأشعة كاما ذات طول موجي ١٠ x ٨١,٥٥٤١٦ واظهرت النتائج وجود فرق بسيط في معامل الانقسام الخلوي لحيوانات التجربة المحقونة بالرؤسيات والاكياس المشعة مقارنة بنفس النوع من الخلايا في حيوانات السيطرة الموجبة والسالبة ، كما ظهرت تغيرات كروموسومية بسيطة، لذلك ليس هناك تأثير للرؤسيات المشعة على كروموسومات المعاملات .

Introduction

The exposure of living cells to mutagenic and carcinogenic factors may lead to chromosomal and chromatic changes (Evans & Orriodan , 1975). Chromosomal aberration in the cells of living organisms could be raised after exposure of such cells to different chemical and physical factors. These factors may cause damage in the cellular chromosomes . Abnormal repair for these nicks may confirm the ability of these factors to cause changes in the DNA molecules (Wo1ff,1981).

There are many tests can be used to investigate the ability of chemical and physical factors to induce changes in the genetic materials. Microorganisms were used widely to detect and identify the carcinogenic chemicals . Most of these tests depend on the detection of changes induced by such chemicals in the DNA molecules of bacteria (Ames, et. al. 1975) fungi (Zimmerman , et. at. 1975) and insects (Tease,1982).

The use of very sensitive bacterial strains to many carcinogenic and mutagenic chemicals to study the changes in the DNA molecules failed to explain the same changes in highly developed primates. Accordingly , many other biological systems closely to humans were used such as mice and rats (huong, et. at. 1990 ; Tice, et. at. 1989).

Some other investigators found that tissue culture can be used to effect of carcinogenic chemicals such as human lymphoid cells (Lamberti, et. at. 1983) and hamster lung cells (Boys, et al. 1990). In the present study we used mice as a model to investigate the chromosomal aberration due to Gamma radiation.

Material and Methods

120 white bulb mice (Al-Razi Research Centre) of 28-35 days old were used. The mice were randomly divided into six groups. Each group contains 20 mice kept in cages with optimal feeding and environment. Fertile of Echinococcus granulosus were isolated from hydatid cyst of liver and lung of sheep origin. The viability of protoscolices were checked and found to be 90%.

The hydatid cyst and protoscolices were subjected gamma rays Co 60 with varying dose of 5 Gy and 25 Gy for 1.2 and 6 minutes respectively. According to the method described by Smyth & Davies,l974 two groups of mice were infested intra- peritoneally with irradiated hydatid cysts of 5 Gy and 25 Gy respectively and another two groups were infested intra-peritoneally with irradiated protoscolices of 5 Gy and 25 Gy respectively. The other two groups were used as positive and negative controls. Mice of positive control group were inoculated with non-irradiated protoscolices of the same dose.

The negative control mice group was inoculated with sterile normal physiological saline.

One month later all the groups were challenged with the dose of 2000 protoscolex/animal intraperitoneally. Five mice from each group were killed after 2 , 4 , 8 and 12 weeks gust challenge and tissue samples were collected and examined grossly and microscopically to investigate the macro and micro changes from cytological and genetic points of view in comparison to positive and negative controls.

Bone marrow samples were collected from femur of mice and processed according to Cook & Pallister, 2000. Processed cells were allowed to grow for 24 hours, collected by precipitated by centrifugation. The precipitated cells were fixed on microscopical slides and stained with Giemza for 5-7 minutes. The mitotic index (M1) was calculated according to the following equation (Ghosh, et. al. 1991) :

$$MI = \frac{\text{Abnormal chromosome}}{\text{Total number}} \times 100$$

The chromosomal aberration was estimated according to Bauchiger et. al. 1983 by calculation of the ratio of chromosomal changes in each 100 cells selected randomly in first metaphase stage of division.

Results and Discussion

The results revealed the M1 of cells was decreased in mice infested with

5 Gy and 25 Gy irradiated protoscolices in comparison to positive control (Table 1) while it was increased in bone marrow cells of mice infested with 5Gy and 25 Gy irradiated hydatid cyst. Such increase or decrease was found to be low and not significant.

grouping	MI
Control (-)	10.8
Control (+)	11.3
5 gray cyst	11.6
5 gray fluid	10.3
5 gray cyst	11
25 gray fluid	10.5

The estimated chromosomal aberration of studied cells (Table 2) was found to be very little and not significant (figures 2,4, and 5) in comparison to control (Figures 1,3,and 6)

grouping	Total count	Gap chromosome	Break chromosome	Gap chromatide	Break chromatide	Ring chromosome	Diecentric chromosome	Acentric chromosome
Control(-)	0.9	0.3	0.1	0.4	0.1	-	-	-
Control(+)	1.2	0.5	0.1	0.3	0.3	-	-	-
5 gray cyst radiation	1.3	0.4	0.2	0.3	0.4	-	-	-
5gray fluid Radiation	1	0.3	0.1	0.4	0.2	-	-	-
25 gray cyst Radiation	1.6	0.3	0.2	0.6	0.5	-	-	-
25 gray fluid Radiation	1.5	0.2	0.2	0.7	0.4	-	-	-

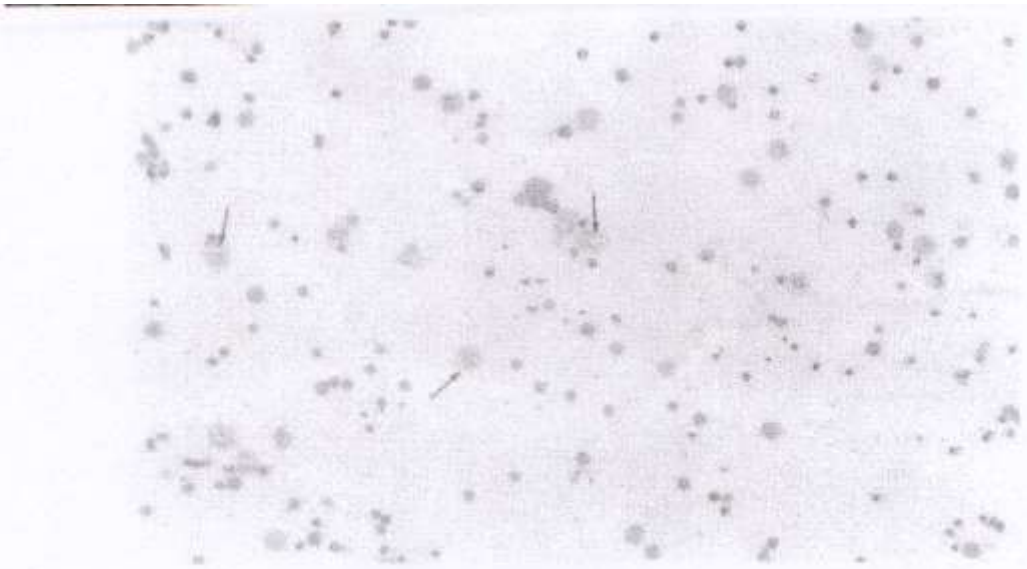

Figure (1) mitotic index bone morrow control positive.

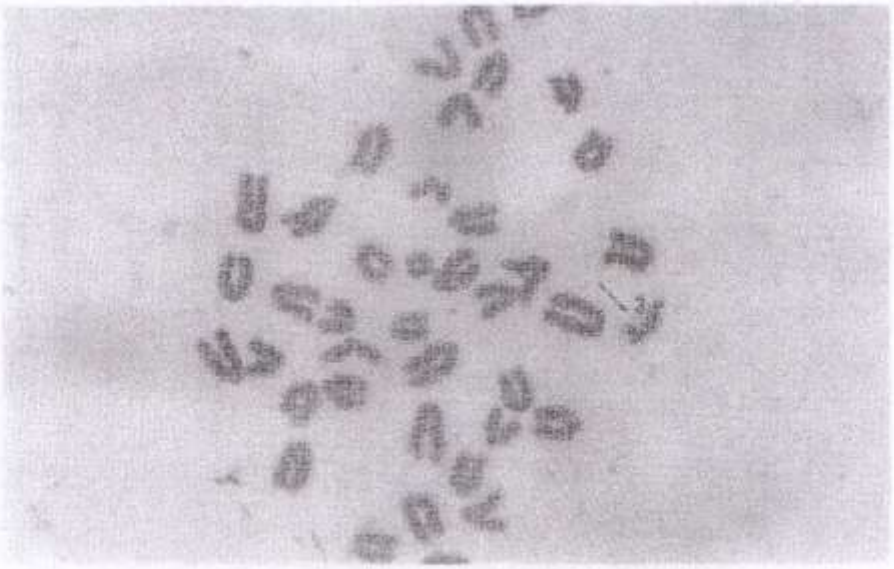

Figure (2) chromosome aberration type Gap chromatid in the bone morrow of experimental mice 5 gray cyst.

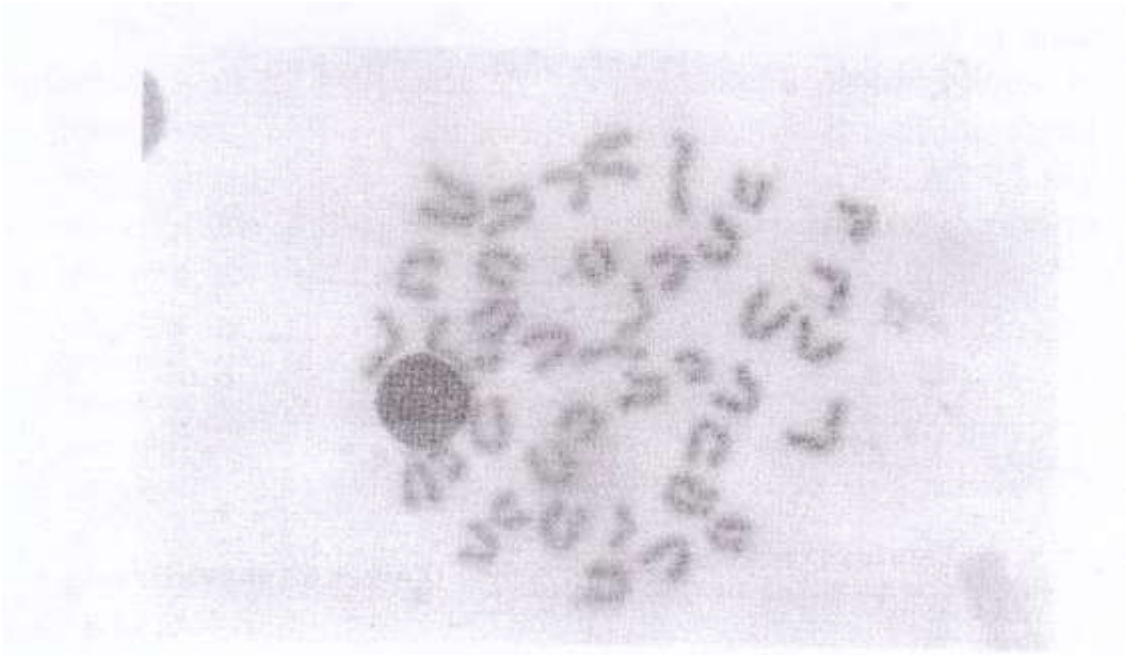

Figure (3) chromosome in the meta phase stage in bone morrow of mice which control negative.

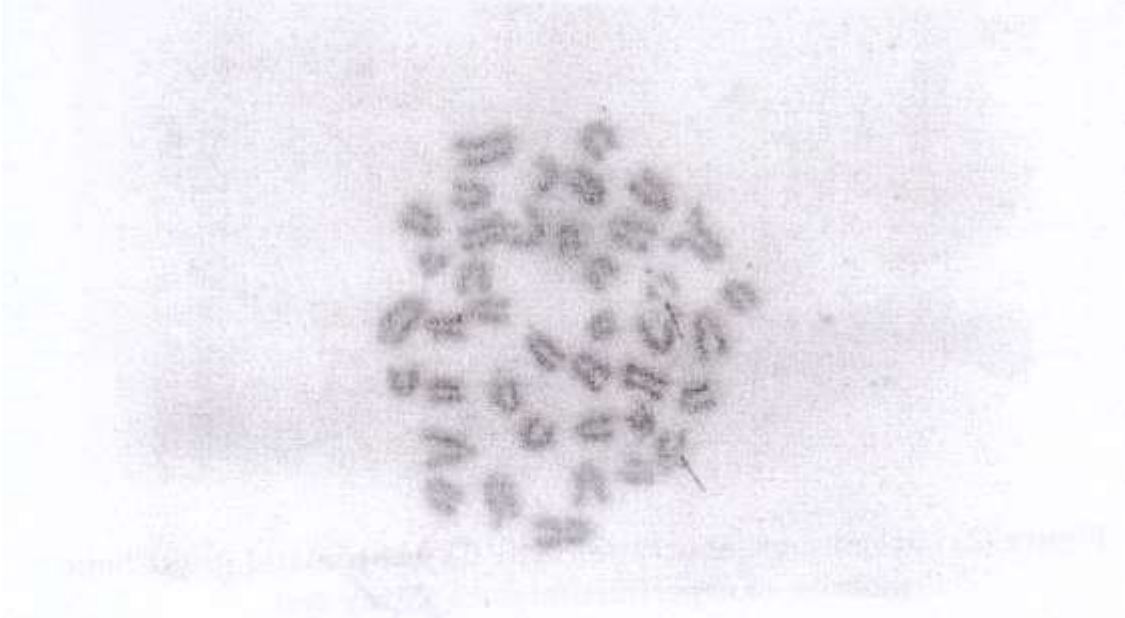

Figure (4) chromosomal aberration type Gap chromatide in bone morrow in the mice in group 25 gray fluid.

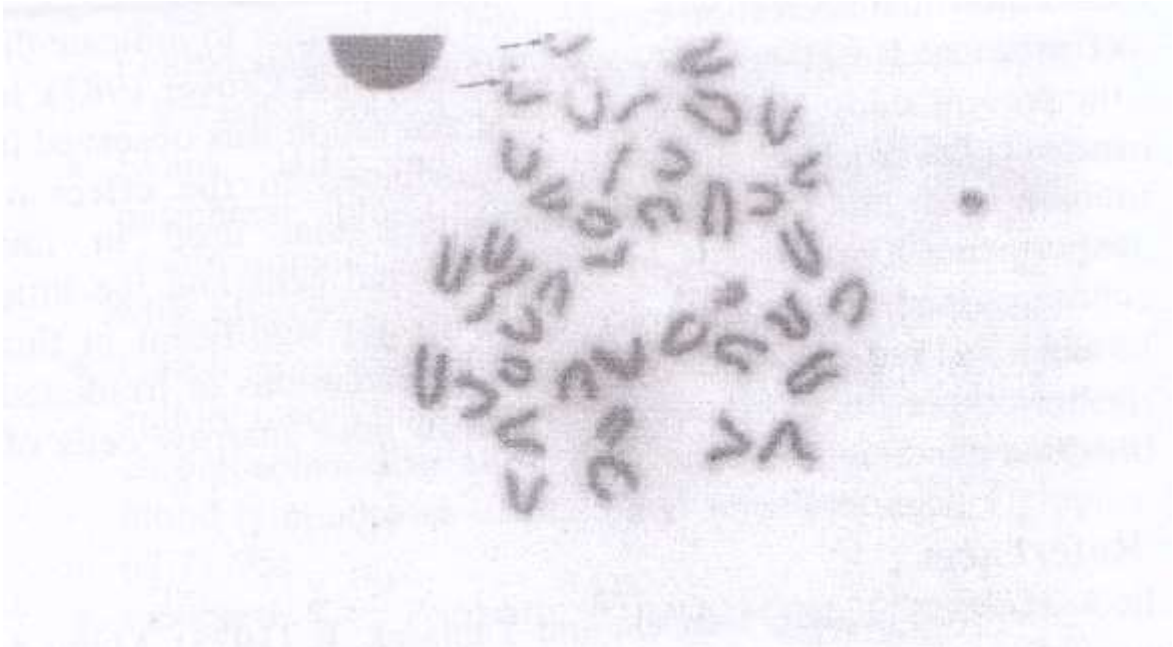

Figure (5) chromosomal aberration type Gap chromatid in the bone morrow in the mice in group 5 gray fluid.

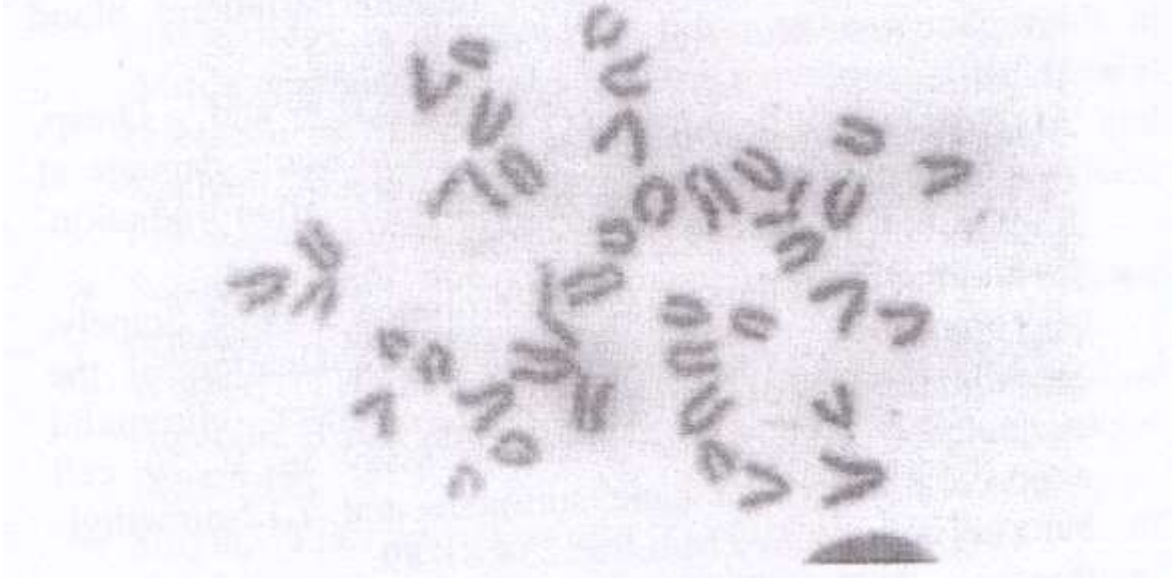

Figure (6) chromosome in the meta phase stage in the bone morrow of the mice which control positive.

The M1 test was used by many investigators to evaluate the chromosomal changes in cells subjected to irradiation. Chemical and physical factors like irradiation can effect the time required for cell division to normal and not irradiated cells.

Chromosomal aberration can be used as a marker to indicate the effect of such factors on normal cells (Mahli & Crover,1987). In the present study, little chromosomal aberration was observed in mice cells. Such change can be attributed to the effect of radiation from irradiated protoscolices that used in this experiment. The changes in M1 of affected cells and the little chromosomal aberration were found to not significant in this study. This means that 5 Gy and 25 Gy radiations of irradiated protoscolices have little or no effect on bone marrow cells of infested mice.

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