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Cytogenetic study of dental diagnostic x-ray workers

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***Abstract*---**Background and Objectives: studying chromosome changes for dental x-ray workers in the city of kut and diagnosing those abnormalities resulting from continuous exposure to radiation. Exposure and Methods: The chromosomes of those exposed to dental X-rays were analyzed and studied using cytogenetic analysis to detect chromosomal aberrations caused by ionizing radiation after collecting blood samples from those exposed to Al-Zahraa and Al-Karama Teaching Hospital as well as from the Taiba Specialized Center in addition to the dental clinics in Wasit. Results: Chromosomal analysis of all subjects showed chromosomal aberrations, and most of the aberrations were structural changes that were divided according to the ratio of their frequencies. The largest percentage was for the gap chromosome type (44%), chromosomal-aberration dicentric (16%) and chromosomal-aberration gab and dicentric (8%). Conclusion: Dental diagnostic x-rays have an effect on the chromosomes of workers exposed to a long-term cumulative exposure period, as it leads to the appearance of aberrations of the type chromosomal aberration (gab chromatid) is the most frequent in this stuy , chromosomal aberration (dicentric) The type of deviations that appear when workers are exposed to radiation doses more than the permissible limit The third group had more than one chromosomal change in the same exposure (gab & dicentric) Such deviations hinder the normal division and cycle of cells.

***Keywords*---**X-rays, chromosomal changes, ionizing radiation, X-ray technicians

Introduction

Human beings are continually exposed to low levels of ionising radiation from natural sources. In addition, the human population is increasingly subjected to

manmade sources of radiation. Of the various sources, medical procedures contribute about 14% of the total annual exposure from all sources (Unscear, 2000). While the medical applications of ionising radiation are accepted worldwide as essential tools to improve human health, they also represent by far the largest manmade sources of radiation exposure. Thus, the trend is that population exposure to diagnostic and therapeutic ionising radiation is likely to increase worldwide, which raises concern over radiation overexposure and its associated risks (Tafreshi *et al.,*2019). Measuring the DNA damage chromosomal Aberrations (CAs) in the peripheral blood lymphocytes of exposed individuals is an essential tool in monitoring persons exposed to radiation and as a biological dosimeter. the potential utility of DNA damage analysis for biological monitoring and dose assessment in individuals occupationally exposed to ionising radiation (Hall *et al.*,2017). The difference could be attributed to individual radiosensitivity, differences in the DNA damage repair capacity and variations in the machine parameters used during the procedure (Marcon et al., 2003). The inter-individual variation in damage repair capacity can be inherited or can result from alterations in gene expression induced by other factors such as diet and smoking habits. Further, radiation risks are significantly affected by patient age. Aged individuals have a reduced damage repair capacity, which may lead to a higher amount of damage upon exposure to radiation. It was cautioned that greater radiation risks are attributed to radiation exposure at a young age, the risk of radiation-induced tumours compared to adults (Hall, 2000).

This suggests that the CAs may be eliminated upon cell division, in that the accumulation of CAs may pose a risk for stochastic effects. Furthermore, the induced aberration frequency showed an inter-individual variation and repair ability when the cell cycle progressed (Orrenius *et al.* ,2011). Exposure to medical X-rays induces DNA damage in circulating peripheral lymphocytes. Hence it is presumed that the accumulation of CAs leads to genomic instability, which may further lead to stochastic effects such as carcinogenesis Unrepaired or is-repaired DSB can lead to the formation of chromosome aberrations (CA), a broad class of DNA mutations that are linked to various health risks (Natarajan & Boei,2003). Increased rates of CA in peripheral blood lymphocytes (PBL) have been associated with an increased risk of cancer CA can therefore be considered a potential bioindicator of cancer risk (Boffetta *et al.,* 2007). Numerous types of CA can be broadly categorized into stable (e.g., inversions and translocations) and unstable (e.g., acentric fragments, dicentrics, and rings) aberrations. The former is non-lethal for cells and can persist for years, whereas the latter cause cell death during mitosis, thus are consideredshort-livedd genotoxic events that decline with time after the triggering insult (Bonassi *et al.*,2008). Detection and quantification of unstable CA are simple and employed shortly after IR exposure, e.g., for biodosimetry Persistence of stable CA allows for the evaluation of the long-term effects of single IR exposures or the cumulative effects of protracted chronic exposures (Pascalis *et al.*,2015). Chromosomal aberrations occur when the cell is exposed to x-rays at the beginning of the interphase in the (G1 period) stage of the cell cycle since the break occurs in both chromatids of the chromosome the type of change depends on the site of the break Dicentric and the method of fusion of broken chromosomes (Lobrich & Jeggo, 2017).

The aim of the study is to use cytogenetic chromosomal aberrations as biological indicators in detecting the effect of dental x-rays on the genetic material in blood lymphocytes for a sample of dental x-ray diagnostic workers who were exposed to low doses of Ionizing rays as a result of work because the correlation of this indicator Vital health outcomes for technicians in the long term in their professional lives By studying chromosomal changes and diagnosing those abnormalities that are affected by continuous exposure to X-rays in the city of Wasit.

Exposure and Methods

This study was conducted for a period of one year from November 2021 to November 2022 Complete clinical data and blood samples were taken from the workers On those exposed to X-rays who practice their professional work at Al-Zahraa and Al-Karama Teaching Hospital, Taiba Specialized Dental Center and Al- Taiba private clinics. The study included 60 blood samples, 40 exposed, 33 males and 7 females, and the control group was 20 blood samples for healthy non-exposed subjects for comparison. Cytogenetic analysis of peripheral blood samples was performed using traditional cytogenetic methods according (short time culture), and the use of culture media (LymphoPrime Medium) and colchicine solution, reagents, and stains (KCL, PBS, fixative solution, trypsin solution, and others), many laboratory equipment and tools. Cellular proliferation and chromosomal analyses were performed according to (Geleick, et al. 1990; Yassen, 1990) with some modifications.

Results

The use cytogene of tic analyzes, which is one of the best modern standards used in cell biological standardization to estimate and estimate radiation doses, uses cytogenetic indicators as an indicator in the biological assessment of workers in radiation fields, such as chromosomal changes, mitotic index and frequency of micronuclei in binuclear lymphocytes (Tubbs &Nussenzweig, 2017).so, that technique was chosen in this research.

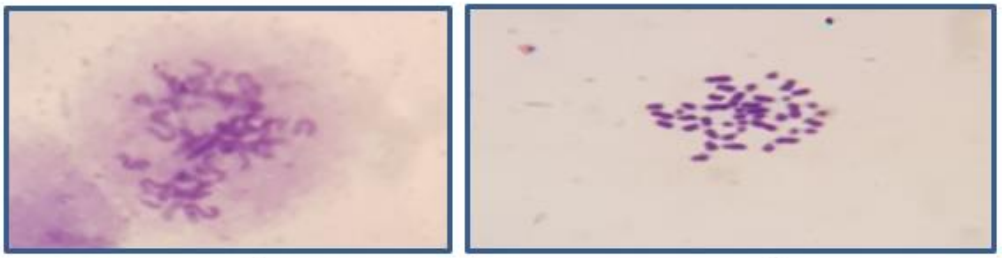
Table (1-1) distribution group samples according dental duration X-ray

Time of duration	Number of case	Age range	Gender M/F
Group(1) 1<year- 5 years	15	22-42	12 : 3
Group (2) 6 years – 10 years	7	28-52	6 : 1
Group (3) 11 year – 15 year	4	32-42	4 : 0
Group (4) 16 year – 20 > year	14	38-78	12 : 2
Control group (0 time duration	20	20-57	10 : 10
Total	60	20-57	10 : 10

The results that were studied showed that the minimum exposure period for dental x-ray technicians was from one year to more than 20 years, exposed within the field of dental diagnosis. Thus, the first research in Wasit studies the exposure period of less than ten years, compared to most research and studies starting from 10 years, as shown in the above table (1-1). In this study, 40 samples of peripheral blood were received from subjects exposed to dental x-rays for cytogenetic preparation (7) cases showed (17.5%) no cell and no metaphase, and 2 cases (5%) clumped metaphases (1-1 B) and 6 cases (0.15%) short chromosomes as well as figure (1-1 A) we can countable but not analyzed, and only (25) cases (62.5%) good growth and good metaphase and the control group consisted of 20 non-exposed normal individuals. Blood samples were taken from them for the purpose of examination to ensure that there is no abnormal condition in their chromosomes, as well as in the number of their cells. Indeed, all (20) cases were metaphase with good enough chromosomes. The obtained chromosomal analysis results are the result of data analysis from (25) cases Two workers exposed to diagnostic dental x-rays and to illustrate the assessment of chromosome number, they are summarized in Table (1-2).

Table (1-2) Distribution of cytogenetic analysis results of 60 cases

Samples	Number of case	Result obtained
Control group	20	Each cases metaphase with good enough chromosomes
Dental diagnostic X-ray Workers	7	No cell (no cell in division(No metaphase
	2	Un spread (clumped metaphases
	6	Metaphases with short chromosomes countable but not analyzed
	25	Metaphases with good enough chromosomes
Total	60	



A B

Figure (1-1) A- Metaphase with short chromosomes of case No. (6) B-clumped metaphase of case No. (2)

Table (1-3): chromosomal abnormality of dental duration X-ray

No. of patient	Age	Gender	dental duration X-ray (years)	Chromosomal aberration
5	28	F	6	46,XX normal chromosomes
6	29	M	5	46,XY normal chromosomes
7	29	M	5	46,XY normal chromosomes
10	29	M	13	Chromosome gape 46,XY
12	32	M	12	46,XY chromosome gape
13	32	M	6	46,XY normal chromosomes
16	32	M	10	46,XY normal chromosomes
19	38	F	16	46,XX chromosome gape
22	37	M	13	Chromosome gape 46,XY
23	39	M	11	46,XY Chromosome gape
24	38	M	20	46, XY dicentric chromosome
25	38	M	18	46, XY dicentric chromosome
26	40	M	19	46, XY dicentric chromosome
27	41	M	10	46,XY normal chromosomes
30	44	M	16	46,XY chromosome gape
31	38	M	10	46,XY normal chromosomes
32	47	F	26	46,XX chromosome gape
33	47	M	20	46,XY chromosome gape
34	45	M	18	46,XY chromosome gape
35	45	M	20	46,XY chromosome gape
36	52	M	10	46,XY normal chromosomes
37	53	M	20	46,XY chromosome gape
38	59	M	25	46, XY dicentric chromosome
39	63	M	35	46, XY dicentric chromosome & gap
40	78	M	50	46, XY dicentric chromosome & gap

Table (1-4) Summary of Cytogenetic analysis of dental duration X-ray

groups	Number of case	Frequency %	Gender M:F	Median age
Chromosomal- aberration chromosome gape	11	44	9:2	41.5
Chromosomal – aberration dicentric	4	16	4:0	43.7
Chromosomal – aberrations chromosome gap and dicentric	2	8	2:0	70.5
Normal chromosomes	8	32	8:1	35

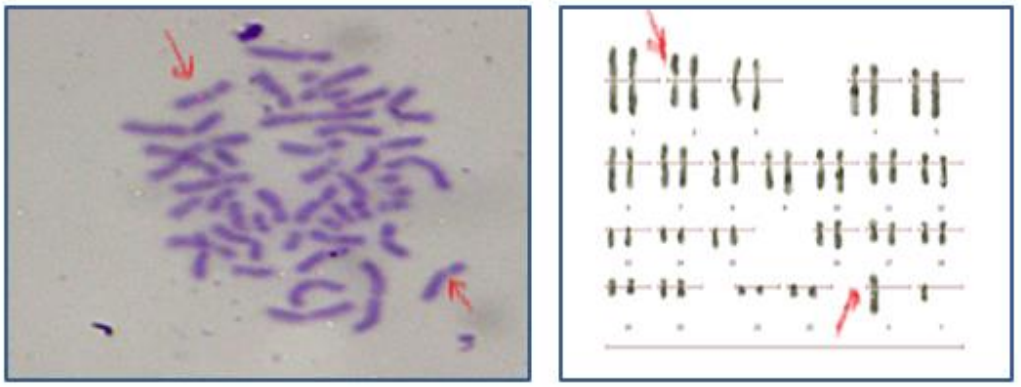


Fig. (1- 2) karyotype of the case (11) 46 XY chromosome Gapes (1000X).

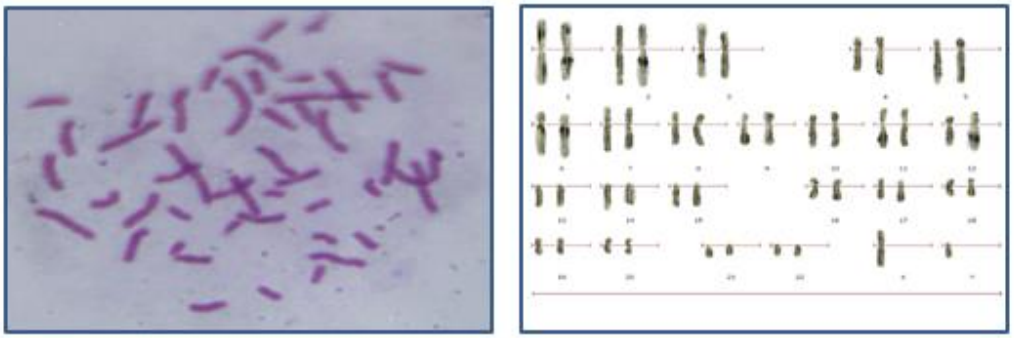


Fig. (1-3) karyotype of case (8) 46 XY normal chromosomes (1000X)

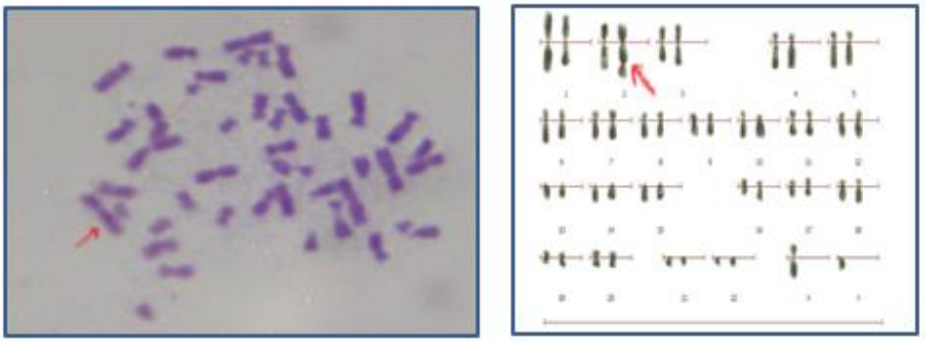


Fig. (1- 4) karyotype of case (4) 46, XY, dis2 chromosome dicentric (1000X).

Discussion

Chromosomal aberrations occur when the cell is exposed to radiation at the beginning of the prophase of the primary growth stage G1 period of the cell cycle, where the break occurs in the chromatids of both chromosomes. The type of change depends on the site of the break and method of fusion of the broken chromosomes (Ballardin *et al.*,2007).

A cell commonly used to monitor occupationally exposed populations to radiation is lymphocytes from human peripheral blood. Whenever there is chronic exposure to X-rays, it will cause leukocytes to accumulate in lymphocytes in the G0 stage of the cell cycle, Therefore it is indexed and cytotoxicity of radiation to x-rays (Tommasino & Durante, 2015) Chromosomal abnormalities have been correlated with genetic alterations that can trigger genomic instability and development of cancer Therefore biomonitoring based on chromosome aberration analysis make it possible to estimate the cancer risk (verdorfer *et al.* , 2001).

Table (1-3) represents the average of the types of chromosomal aberrations studied in lymphocytes after examining (25) samples taken from a group of dental diagnostic workers, x-rays. The percentage of chromosomal changes in age groups is (41 .5 %) year the highest in this study, as it was a gap chromatid type, where it was 11 cases and its frequency was (44%) The ratio of males to females was (9 : 2) The consistent with most previous research and studies, which indicate that this age group is most affected by low radiation for diagnostic X-rays High frequencies of chromosomal damage from Cape diversity in peripheral blood lymphocytes of dental X-ray technicians who were exposed to a high exposure period, suggesting the possibility of For years of working in the medical imaging field long-term adverse health effects due to genetic instability as in In this study, the effects of x-rays on chromosomes were evaluated, and e cases showed a chromatid gap, which is formed, The chromatid aberrations scored included chromatid gaps and breaks (Jain *et al.*,2017).As for the exposed numbers (10,12,19,22,23,32,33,34,35,37), One case was female and the rest was male After examining it, Their ages ranged between (29-53) years, and they were within the exposure periods of (11-20) years It was (11) cases From a total of (25) exhibitors as 1n the figure (1-2). it was found that there are chromatic aberrations of the gap type in the worker's group compared with the control group, agreed where they noticed a significant increase in chromatid gap-type chromosomal aberrations when compared with the control group. (kazhal,2017).

Where a dicentric type appeared in four exposed to X-rays, and their sample number was 24,25,26,38 They were between the ages of (38-59) All of them were males and their exposure period ranged from 18-to 25 in the field of dental x-rays as in the figure (1-4) , The current study confirmed, The emergence of this dicentric type in four subjects, the reason for this may be due to their advanced age and exposure for many years to X-rays in their field of work, in addition to the fact that most of them are smokers due to the fact that the latter is an important factor in showing chromosomal aberrations (Umal, 2014). According to studies and research of the chromosomal aberrations High frequency of type of chromosomal aberrations, dicentric and chromosome were observed in the peripheral blood lymphocytes of medical staff who were exposed to x- ray This

agrees with (Lee *et al.*,2010). concluded that chromosomal aberrations, specially dicentric chromosomes can be used as a good indicator of exposure to radiation This is because it is the most sensitive indicator of radiation exposure Dicentrics are proved to be excellent markers of radiation exposure and have been widely used as a key marker in the radiation dosimetry (Ameerunnisa *et al.*,2011). This study agreed with all of the (Stuart &white, 1967; Rozgaj *et al.*,1999; Abolfazl Movafagh *et al.*,2007).

In another study, the absence of dicentric chromosomal aberration in Brazilian dentists who are chronically exposed to X-rays, take adequate precautions during exposure in their work environment or that the level of exposure was not sufficient to cause damage (Cintia & Ilce,2002). As for the exposures whose numbers were (39-40), their ages were (63-78), respectively, and the duration of their occupational exposure to chromosomes was (35) and (50) years in the field of dental diagnosis by x-ray, and they were males.

Their chromosomes were described after examining them under the power of magnification 1000X, as shown in the figure above (1-5) , in the type of gap and dicentric chromosome, This is inconsistent with what was stated by (Sivankaev, 2000) . Where the emergence of the type of dicentric deviation was not observed in the group of workers in the field of radiation.

The gape and dicentric aberrations type was detected in samples of the last group due to the long exposure periods, which are known as chronic, as well as age and smoking led to the emergence of these chromosomal differences, As for the ring chromosomal aberrations, they are few and rare in comparison with the chromosomal aberrations that were not observed in the blood samples of the worker's group and the control groups are because the occurrence of ring chromosomes requires a double break in the same chromosome and the fusion of their ends compared to the chromosomal pieces that form when a single break in the chromosome occurs (Hall & Giaccia, 2011).

This is consistent with what he found (Balasem & Ali,1992). In the absence of cyclic chromosomal aberrations in human blood samples exposed to low radiation doses Because the amount of dose used in dental radiography, is less compared to the dose used in other fields of medicine such as diagnostic and therapeutic medical radiation (Wouters *et al.*,2009).

This leads to the accumulation of cytotoxicity due to reduced oxidative stress and accumulation of free radicals, and the inability to remove the cancer-causing reactive oxygen stress (ROS) and turn it into Accumulation genotoxicity This leads to many age-related diseases such as Alzheimer's and Parkinson's disease. (Feinendegen, 2005). It was concluded that chronic exposure to x-ray can ionizing radiation shows a significant genotoxic capacity show radiation-induced chromosomal damage increasing the capacity of several antioxidant compounds against x-ray radiation. It was seen how some pure flavonoids (diosmin and rutin) and polyphenolic extracts show greater protective capacity than traditional radioprotectors, for example, sulfhydryl compounds and even vitamin C, against X-rays in vivo (Benavente-Garci *et al.*, 2005). The numbers of workers (5,6,7,13,16,27,31,36), All of them were males, except for one case females and

the duration of exposure were about (5-10) years, and their ages were between (28-52) years old, The number of cases was 8 cases of a total of (25) samples Their tests showed that they did not have any chromosomal changes, they were normal as in the figure above (1- 3). This result was identical to what it came up with Identical to what came from a cell study on Brazilian dentists who were occupationally exposed to a low dose of ionizing radiation, where there was no significant difference between dentists and non-exposed controls (Cintia & Ilce,2002).

In this study, dentists who were exposed for at least ten years to x-rays, and where the average lifespan of long-lived lymphocytes is seven years and It may be the reason for the absence of chromosomal changes, in addition to the fact that it is possible to assess the potential of physical and chemical factors to cause many dangerous effects on all cells (Maddleti et al.,2002).In conclusion, my studies were compatible with both (Amal *et al.*,2015 ; Kazhal,2017 ; Ameerunnisa *et al.,*2011; Alihossein Saberia *et al.*,2013; Gadhia *et al.*,2004), and You will not agree with (Cintia & Ilce Mara,2002).

Conclusion

This first study in Wasit included a study of the chromosomes exposed to medical diagnostic dental x-rays in a period of exposure of fewer than 10 years in the field of their professional exposure, The occupational hazard of the x-ray technician increases from the cumulative effects of exposure over time (stochastic effects). Exposure to daily procedures over a long time may cause serious, long-term and possibly fatal adverse. The results of the chromosomal analysis were :the chromosome's most common changes are structural changes, The largest proportion of these differences was the Chromosomal - gap type followed by Chromosomal - dicentric As shown in the figure above (1-4). This finding may be useful in the conclusion that chromosomal aberrations, specially dicentric chromosomes can be used as a good indicator of exposure to radiation This is because it is the most sensitive indicator of radiation exposure and has been widely used as a key marker in the radiation dosimetry biomonitoring based on chromosome aberration analysis make it possible to estimate the cancer risk.

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