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Radio-adaptive Response: An Implication for the Biological Consequences of Low Dose-rate Exposure to X-Ray

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RADIATION induced adaptive response is described as the reduced damaging effect of a challenging radiation dose when induced by a previous low priming dose. To verify the radio-adaptive response that can be induced by occupationally (*in vivo*) received chronic low dose of X-ray, chromosomal aberration (CA) analysis, micronucleus test (MN), interleukin-1β (IL-1β) and nitric oxide (NO) concentrations were investigated for both the occupationally exposed and control groups before and after exposure to 2 Gy γ-rays as a challenge dose.

The results showed that an elevated frequency of CA, MN and nucleoplasmic bridge (NPB) was recorded in radiation workers (exposed group) compared to control group. However, after 2Gy *in vitro* irradiation of lymphocytes of exposed and control groups, the exposed group was found to be lower than that of control group.

On the other hand, IL-1β and NO concentrations in plasma were elevated in exposed group more than in control group. While, after 2Gy irradiation for both groups, there are higher increment in the concentrations of IL-1β and NO in exposed group than the increment difference observed for control group after *in vitro* irradiation as compared to the same group before irradiation.

The present results suggested the existence of an *in vivo* cytogenetic adaptive response in individuals occupationally exposed to low dose of X-ray. In addition, the results showed that NO radicals and IL-1β have a role in the induction of radio-resistance due to *in vivo* exposure that may intermediate this radiation.

Keywords: Adaptive response, occupation exposure, x-ray, chromosome aberration, IL-1β, nitric oxide.

There is much debate concerning the biological effects of low dose ionizing radiation and whether a threshold dose exists for such effects. The International Commission on Radiological Protection (ICRP) **(ICRP, 1990)** has adopted the so-called ‘‘linear-no threshold’’ (LNT) hypothesis to estimate the risks for the stochastic effects of ionizing radiation. Under this hypothesis, the biological effect is assumed to be proportional to the radiation dose received, with no dose threshold for such effects. Hence the ICRP adopted a cautious approach in the light of scientific knowledge at that time and subsequently many national regulatory bodies have based their radiation exposure dose limits for occupationally exposed workers and members of the public on risk estimates derived using this model. However, there have been relatively few data obtained at low doses to support such an important concept (Goldberg *et al.,* 2004 and Prasad *et al.,* 2004).

X-ray machines and radiation emitting sources are used in hospitals for the diagnosis and treatment of diseases. Some of the hospital employees who work in radiology, nuclear medicine, radiation oncology, and some laboratories are specifically trained in the operation of radiation machines and the handling of radioactive materials and sources. Health risks from radiation exposure in such a large occupational segment of the population are clearly of special concern (AAPM report, 1995).

Radiation-induced adaptive response is described as the reduced damaging effect of a challenging radiation dose when induced by a previous low priming dose. Adaptive responses have been observed *in vitro* and *in vivo* using various indicators of cellular damage, such as cell lethality, CAs, mutation induction, radiosensitivity, and DNA repair. In addition, there is general acceptance that mammalian cells carry out repair and control of DNA injury by constitutive processes, that remain available to cope with background damage to DNA, replication and housekeeping, and also by pathways that may be upregulated or induced in response to excessive injury which demands more aggressive repair to retain cell viability (Wolff *et al.,* 1989).

Bio-dosimeter, the biological assessment of radiation dose in individuals through the measure of specific dose-associated effects, provides a measure of the absorbed dose taking into account the individual radiation sensitivity. Cytogenetic biomarkers have been used in occupational setting for over 50 years, especially in the field of radioprotection, where the frequency of CA is currently used as a biological dosimeter of ionizing radiation (Ropolo *et al*.,

2012). On the other hand, there are new investigation methods that allow cytokines to be used as radioprotective or radiosensitizing agents. They can also be used to minimize the effects of irradiation indirectly by neutralizing other harmful cytokines. Evidence from both the laboratory and the clinic indicates a promising future for these molecules as modulators for the radiation response (Laiakis *et al.,* 2007). Several researches have studied the radioprotective effects of IL-1 and tumour necrosis factor (TNF-α) confer radioresistance by promoting repair and restoring the host defenses (Neta *et al.,* 1991). IL-1β has been hypothesized to induce hematopoietic growth factors and endogenous antioxidant mechanisms such as metallothionein, cerruloplasmin, and MnSOD (Weiss and Landauer, 2000). NO is an important cellular signaling molecule that is involved in a variety of signal transduction pathways and possesses unique physiologic or pathologic properties, which have been described as cytoprotective, cytostatic, pro-apoptotic, or anti-apoptotic. NO also plays an important role in the regulation of tumour evolution (Brune, 2003, Li and Wogan, 2005 and Xu *et al*., 2002).

Our study aimed to examine the influence of low dose rate of X-ray on the occupationally exposed persons and the health risk in view of the adaptive response theory.

Materials and Methods

Subjects

Ten individuals (6 physician and 4 technicians) had been occupationally exposed to low levels of radiation during their handling with X-ray machine; Shimadzu Corporation, Voltage: 125 V, Current: 450 m.A. During radiography with fixed installations, the radiographer would normally be expected to stand in a control booth that is typically shielded as a secondary barrier against X-ray tube leakage and scattered radiation. None of the workers was exposed to whole body doses of ionizing radiation beyond the annual limit of 20 mSv. Their mean exposure was 0.24 mSv (0-4.66 mSv) during the previous year of blood collection. The reference control group consisted of 10 individuals with no history of exposure to ionizing radiation or chemical compounds. All the subjects of both groups lived in the same urban area.

Subject questionnaire

All the participants were informed about the aim of the study prior to blood collection in the course of a routine occupational medical examination by a specialized physician who filled in a structured questionnaire which covered

standard demographic questions, as well as an occupational, medical and family history. For the organization of the data analysis, each variable was assigned to one of the following categories: demographics, life style, work exposure and medical history.

Blood sampling

Samples of 5 ml of whole blood were collected in heparinized vacationer tubes under sterile conditions by vein puncture in the morning h, between 9 and 10 am, after collection, all blood samples were randomly coded, cooled to 4 ^{0}C and transported to the laboratory. Processing and scoring of samples of the 2 groups were then performed blind and concurrently in the laboratory (usually within 2 h following the sampling). Each peripheral blood sample was divided into two main categories; one as the control (before irradiation) and the other for *in vitro* exposure to 2 Gy γ-rays. Each part was then subdivided into two parts; one was used for MN and CA assay and the other for biochemical analysis.

Irradiation protocol

Heparinized blood samples were exposed to a single dose of 2 Gy at dose rate of 0.492 Gy/min. Irradiation was carried out using the Canadian Gamma Cell-40 biological irradiation belonging to the National Center for Radiation Research and Technology (NCRRT) Cairo, Egypt.

The analysis of structural chromosome aberrations & micronucleus test

The structural CA was performed according to current IAEA guidelines (IAEA, 2001). Four separate culture vials were set up from each individual; 2 for the control (MN & CA) and the other 2 exposed to 2 Gy γ-rays (MN & CA). A short-term culture of lymphocytes for CA and MN was set up.

The MN assay was performed as described by fenech (1993) with some modifications, Fenech (2000).

Biochemical analysis

Blood samples were collected in dry clean centrifuge tubes that contained heparin to obtain plasma and then centrifuged at 2000 rpm for 10 min and the resulting supernatant plasma was collected. Plasma samples were used for the following analysis; IL-1β was determined in plasma according to the method described by Orgenium Laboratories procedure (IL-1β ELISA kit). All reagents

used were purchased from AviBion Human IL-1β ELISA. NO was determined in plasma according to the method described by Miranda *et al*. (2001).

Statistical analysis

Statistical analyses were performed using analysis of variance (ANOVA) according to (Snedecor and Cochran, 1980). Data were presented as mean± S.E. and $P \leq 0.05$ was considered statistically significant.

Results

The personal data of the subjects

As shown in Table 1. the data of exposed group comprises 5 males and 5 females, these persons had been occupationally exposed to X-ray with mean duration of 18.6 years. The mean age of this exposed group was 40.9 years (range 28-52 years). The subjects of control group matched the exposed subjects in age and smoking habits.

TABLE 1. A) Personal data of exposed group.

A) Exposed										
Indiv. No.	**Sex**	**Age**	**Occup.**	**Rad.**	**Duration**		**Medical history**			**Smok. Status**
					No. of years	**No. of h/day**	**Disease**	**Drug**	**X-ray exposure**	
1	*F	38	Techn.	X-ray	18	6	= =	= =	= =	*NS
2	F	35	Techn.	X-ray	15	6	Anaemia	= =	= =	NS
3	F	35	Techn.	X-ray	15	6	= =	= =	= =	NS
4	*M	52	Physic.	X-ray	26	6	= =	= =	= =	*S
5	M	40	Physic.	X-ray	18	6	= =	= =	= =	NS
6	F	48	Physic.	X-ray	25	6	= =	= =	= =	NS
7	M	45	Physic.	X-ray	23	6	= =	= =	= =	S
8	M	39	Techn.	X-ray	16	6	= =	= =	= =	NS
9	F	49	Physic.	X-ray	24	6	= =	Vitamin	= =	NS
10	M	28	Physic.	X-ray	6	6	= =	= =	= =	S

*__M__: Male. *__F__: Female. *__S__: Smoker. *__NS__: Non smoker.

All subjects of exposed and control groups were not under any prescription or non-prescription medications, vaccinations, and X-ray or another diagnostic proof within the last year before blood sampling. Only one subject (No.9) in exposed group and subject (No.1) in control group received a course of natural vitamins through the last three months.

TABLE 1. B) Personal data of control group.

B) Control										
Indiv. No.	**Sex**	**Age**	**Occup.**	**Rad.**	**Duration**		**Medical history**			**Smok. Status**
					No. of years	**No. of h/day**	**Disease**	**Drug**	**X-ray exposure**	
1	*M	24	Chemist	= =	= =	= =	= =	Vitamin	= =	*NS
2	M	40	Chemist	= =	= =	= =	= =	= =	= =	*S
3	*F	32	Employee	= =	= =	= =	= =	= =	= =	NS
4	M	28	Employee	= =	= =	= =	= =	= =	= =	NS
5	M	49	Engineer	= =	= =	= =	= =	= =	= =	S
6	M	34	Employee	= =	= =	= =	= =	= =	= =	NS
7	F	26	Employee	= =	= =	= =	= =	= =	= =	NS
8	F	29	Employee	= =	= =	= =	= =	= =	= =	NS
9	M	55	Engineer	= =	= =	= =	= =	= =	= =	S
10	F	34	Engineer	= =	= =	= =	= =	= =	= =	NS

*__M__: Male. *__F__: Female. *__S__: Smoker. *__NS__: Non smoker.

Chromosome aberration analysis of exposed and control groups before and after exposure to 2 Gy γ-rays as challenging dose.

Table 2. indicates the mean and standard error of the CA per individual in exposed group before and after irradiation. It reveals that there is a significant increase in chromosome fragments, breaks, dicentrics and total CA after irradiation in comparison with those before irradiation.

The mean value of the total chromosome type aberrations after irradiation was almost 2.4 folds as high as the total chromosome types before irradiation. The frequency of polyploidy and total aberrations revealed a significant increase after irradiation compared to those before irradiation.

The probability values of significance between the means of the different types of aberrations before and after 2 Gy gamma radiation exposures in control group are presented in Table 2. There is a significant increase in chromosome fragments, breaks, dicentrics and total CA in the control group after irradiation when compared with the same group before irradiation. Similarly, a significant increase in chromatid fragments and total chromatid aberrations was observed in the control group after irradiation compared with the same group before irradiation.

TABLE 2. Probability values of chromosome and chromatid aberration frequencies in exposed and control groups before and after exposure to 2 Gy γ-rays.

Types of aberrations	Exposed gp. before γ-rays	Exposed gp. after γ-rays	Control gp. before γ-rays	Control gp. after γ-rays
Chs. Frg.	2.3± 0.37	4.0± 0.39[b]	1.6± 0.31	4.7± 0.42[a]
Chs. Brk.	1.7± 0.30	3.3± 0.47[b]	0.8± 0.25	4.7± 0.30[a]
Dicentric	0.0± 0.00	1.1± 0.35[b]	0.0± 0.00	1.7± 0.30[a]
Ring	0.0± 0.00	0.2± 0.13	0.0± 0.00	0.3± 0.15
Total Chs. Ab.	4.3± 0.67	10.4± 1.17[b]	2.5± 0.52	13.9± 1.15[a]
Chd. Frg.	1.5± 0.34	2.1± 0.28	1.2± 0.33	3.1± 0.23 [a]
Chd. Brk.	0.6± 0.22	1.0± 0.26	0.6± 0.34	1.4± 0.48
Chd.exchange	0.1± 0.10	0.1± 0.10	0.0± 0.00	0.2± 0.13
Total Chd. Ab.	2.2± 0.51	3.2± 0.407	1.8± 0.55	4.7± 0.54[a]
Polyp.	0.2± 0.13	1.2± 0.20[b]	0.0± 0.00	1.3± 0.30[a]
Total Ab.	6.7± 1.21	14.8± 1.45[b]	4.3± 1.02	19.9± 1.70[a]

Gp.: group, **Chs:** Chromosome, **Chd:** Chromatid, **Frg:** Feagment, **Brk:** Break, **Ab:** Aberration. **Polp:** Polyploidy, **[a]:** Significantly different from control group before irradiation, **[b]:** Significantly different from exposed group before irradiation, Data (Mean ± S.E.) are considered significant at $P< 0.05$, n= 10.

Table 3. shows that the statistical differences between the means of chromosome fragments, chromatid fragments, chromatid breaks, chromatid exchanges, total chromatid aberrations, polyploidy and total aberrations were insignificantly increased, before γ-rays, in the exposed group when compared with control group. Whereas, the differences between the means of chromosome breaks and total CA were significant ($P< 0.05$) before γ-rays in the exposed group as compared with control group.

TABLE 3. Probability values of chromosome and chromatid aberration frequencies in exposed and control groups before exposure to 2 Gy γ-rays.

Types of aberrations	Control group before γ-rays	Exposed group before γ-rays
Chs. Frg.	1.6± 0.31	2.3± 0.37
Chs. Brk.	0.8± 0.25	1.7± 0.3[a]
Dicentric	0.0± 0.00	0.0± 0.00
Ring	0.0± 0.00	0.0± 0.00
Total Chs. Ab.	2.5± 0.52	4.3± 0.67[a]
Chd. Frg.	1.2± 0.33	1.5± 0.34
Chd. Brk.	0.6± 0.34	0.6± 0.22
Chd.exchange	0.0± 0.00	0.1± 0.1
Total Chd. Ab.	1.8± 0.55	2.2± 0.51
Polyp.	0.0± 0.00	0.2± 0.13
Total Ab.	4.3± 1.02	6.7± 1.21

Legend as in Table 2.

The results in Table 4. reveals that a significant decrease in chromosome breaks, total CA, chromatid fragments, total chromatid aberrations and total aberrations were recorded after irradiation in the exposed group when compared to those of control group. On the other hand, this decrease was found to be insignificant in chromosome fragments, dicentrics, rings, chromatid breaks, chromatid exchanges and polyploidy in exposed group when compared to those of control group.

TABLE 4. Probability values of chromosome and chromatid aberration frequencies in exposed and control groups after exposure to 2 Gy γ-rays.

Types of aberrations	Control group before γ-rays	Exposed group before γ-rays
Chs. Frg.	4.7± 0.42	4.0± 0.39
Chs. Brk.	4.7± 0.3	3.3± 0.47[d]
Dicentric	1.7± 0.3	1.1± 0.35
Ring	0.3± 0.15	0.2± 0.13
Total Chs. Ab.	13.9± 1.15	10.4± 1.1[d]
Chd. Frg.	3.1± 0.23	2.1± 0.28[d]
Chd. Brk.	1.4± 0.48	1.0± 0.26
Chd.exchange	0.2± 0.13	0.1± 0.1
Total Chd. Ab.	4.7± 0.54	3.2± 0.47[d]
Polyp.	1.3± 0.3	1.2± 0.2
Total Ab.	19.9± 1.70	14.8± 1.45[d]

[d]: Significantly different from control group after irradiation.

Legend as in Table 2.

Micronucleus test of exposed and control groups before and after exposure to 2 Gy γ-rays as challenging dose

1000 cytokinesis blocked cells were scored (cells which have complete nuclear but not cytoplasmic division) for each individual of the exposed and control groups. Table 5. shows the means, standard errors and probability values of cells with one, two, and three MN in the exposed group before and after irradiation.

A significant increase in the mean values of cells with one, two, and three MN, and total number of MN was observed in the exposed group after irradiation when compared with those before irradiation. In addition, the probability value of cells with nucleoplasmic bridge in the exposed group after irradiation is statistically significant ($P< 0.05$) when compared with those of the same group before irradiation.

TABLE 5. Means and standard errors of 1, 2, 3 MN/ individual in the exposed and control groups before and after exposure to 2 Gy γ-rays as a challenge dose.

Means± S.E. No. of individual	Exposed gp. before γ-rays	Exposed gp. after γ-rays	Control gp. before γ-rays	Control gp. after γ-rays
Cell with one MN	15.8± 0.87	28.6± 1.53[b]	11.6± 0.72	35.6± 1.87[a]
Cell with two MN	0.9± 0.23	2.3± 0.3[b]	0.7± 0.21	2.4± 0.34[a]
Cell with three MN	0.0± 0.00	0.4± 0.16[b]	0.0± 0.00	0.4± 0.16[a]
Total no. of MN	17.6± 1.19	34.4± 2.1[b]	13.0± 1.02	41.6± 2.63[a]
No. of NPB	0.6± 0.31	2.0± 0.47[b]	0.3± 0.15	3.6± 0.48[a]

Legend as in Table 2.

Table 5. shows the frequency of the total number of MN in the control group after irradiation was increased 3.2 folds compared with those of the control group before γ-rays. A highly significant increase in the mean values of 1, 2, 3 MN, total number of MN and NPB was observed in the control group after γ-rays compared to those of the control group before gamma irradiation.

TABLE 6. Means and standard errors of 1, 2, 3 MN/ individual in the exposed group and control group before exposure to 2 Gy γ-rays as a challenge dose.

Means± S.E. No. of individual	Control gp. before γ-rays	Exposed gp. before γ-rays
Cell with one MN	11.6± 0.72	15.8± 0.87[a]
Cell with two MN	0.7± 0.21	0.9± 0.233
Cell with three MN	0.0± 0.00	0.0± 0.00
Total no. of MN	13.0± 1.02	17.6± 1.19[a]
No. of NPB	0.3± 0.15	0.6± 0.31

Legend as in Table 2.

Data collected in Table 6. illustrates that, before irradiation, the probability values of cells with one MN and total number of MN were significantly increased ($P< 0.05$) in the exposed group as compared to the control group, while, the probability values of cells with two MN and NPB were not significant compared with control group.

It could be seen from Table 7. irradiation of samples caused a significant decline in the probability value of cells with one MN and total number of MN in the exposed group than those of the control group. In same concern, the probability value of cells with NPB was significantly decreased in the exposed group than that of the control group after exposure to 2 Gy γ-rays.

TABLE 7. Means and standard errors of 1, 2, 3 MN/ individual in the exposed group and control group after exposure to 2 Gy γ-rays as a challenge dose.

Means± S.E. No. of individual	Control gp. after γ-rays	Exposed gp. after γ-rays
Cell with one MN	35.6± 1.87	28.6± 1.53[d]
Cell with two MN	2.4± 0.34	2.3± 0.30
Cell with three MN	0.4± 0.16	0.4± 0.106
Total no. of MN	41.6± 2.63	34.4± 2.10[d]
No. of NPB	3.6± 0.48	2.0± 0.47[d]

Legend as in Table 4.

Biochemical effects

Table 8. clearly shows that there is a significant increase in the mean values of IL-1β in exposed group before irradiation when compared to those of control group, while, a significant decrease in IL-1β levels was recorded after exposure to 2 Gy compared to those of control samples. On the other hand, a significant increase in the IL-1β levels was noticed in exposed group after irradiation compared to those of the corresponding groups before irradiation. A similar pattern of a significant increase in the IL-1β concentration was recorded in the control group after irradiation when compared with the same group before irradiation. Also, there is a significant increase in NO concentrations in plasma of individuals of exposed group compared with those of the control group before irradiation.

TABLE 8. Means and standard errors of IL-1β (pgm/ ml) and NO (µmol/ L) in plasma of control and exposed group before and after exposure to 2 Gy γ-rays as a challenge dose.

Groups	IL-1β (pgm/ ml)		NO (µmol/ L)	
	Before γ-rays	After γ-rays	Before γ-rays	After γ-rays
Control group	3.1± 0.13	25.7± 1.67[a]	25.8± 1.39	39.93± 2.24[a]
Exposed group	3.7± 0.11[a]	18.8± 0.38[bd]	33.68± 1.49[a]	41.99± 1.84[b]

Legend as in Table 4.

This increment was non significant in both exposed group as compared with the control group after irradiation. Meanwhile, NO concentrations were significantly increased in exposed group after irradiation when compared to those of the corresponding groups before irradiation.

Discussion

The exact risk of radiation-induced cancer at very low doses is not totally understood and is further complicated by many factors, such as the magnitude

of the dose, the time span over which the dose was delivered, the general state of the health of the individual, the type of radiation to which the individual was exposed, the energy of the radiation and the area of the body to which the dose was delivered, among others (Thompson, 2001).

Initial exposure to small doses of radiations is known to condition the cells to adaptive response. It enhance DNA repair ability, produce protective proteins to minimize the indirect damaging effect of subsequent high doses of radiations (Cai, 1999), stimulate proliferation as well as immune response (UNSCEAR, 1994) and induce delay in the passage of cells through the cell cycle (Filippovich *et al.*, 1998).

The induction of the adaptive response appears to be influenced by several factors, including the quality of radiation, the time interval between adaptation and challenge doses, the stage of the cell-cycle at which the adaptation dose was delivered, the dose and dose rate used for adaptation and challenge, and the number of adaptation doses used (Bai and Chen, 1993). Furthermore, once induced, the adaptive response was shown to persist in human blood lymphocytes for at least three cell cycles (Coates *et al.*, 2004).

Our results indicated that the overall frequency of aberrant cells was higher in exposed workers (5.5%) than in controls (3.7%). These results are in agreement with those reported by Maffei *et al.* (2004), and the overall frequency of CA (chromosome break and total CA) before exposing to 2 Gy is significantly higher in exposed group than in the matched controls. The present results for exposed group are in full agreement with the results of previous reports (Kubelka *et al.*, 1992).

We compared induction of cytogenetic radio adaptive response in X-ray occupationally exposed workers to non-exposed controls. We considered X- ray as conditioning doses in radiation workers including X-ray workers. When samples were irradiated to 2 Gy, it was found that the frequency of aberrant cells for X-ray workers 12.7 was lower than exposed control (16.4). Most of the chromosome and chromatid type aberrations decreased in X-ray workers than exposed controls. We scored 12 and 13 polypoidy metaphases in X -ray worker and exposed control, respectively.

In this experiment, the percentages of dicentrics showed that there are no significant changes between controls and exposed workers. Absence of dicentrics

recorded in the present study or low frequency of dicentrics recorded by many authors (Cardoso *et al.*, 2001 and Maffei *et al.*, 2004) may be due to the low levels of ionizing radiation experienced by the workers (Barquinero *et al.*, 1993).

The present results showed that irradiation of samples resulted in significant difference in the frequency of chromosome break and chromatid fragment (3.3% and 2.1%, respectively) for X-ray workers when compared with control (4.7% and 3.1%), respectively. Results also showed that the frequency of total CA (10.4%) and total chromatid aberrations (3.2%) were significantly lower than exposed controls (13.9% and 4.7%, respectively). These results are in agreement with previous reports of (Mitchell and Boreham, 2000 and Mozdarani and samavat, 1996).

At low doses, the body's natural repair mechanisms usually perfectly repair any damage incurred (Thompson, 2001). Many of induced DNA lesions are successfully repaired from a few min (4-15 min) (Tice, 1995) to a couple of h (2-3 h) (Singh *et al.*, 1988) after exposure. Chromosome and chromatid breaks arise from double strand breaks that have been incompletely repaired or unrepaired. The repair of double strand breaks can also produce double fragments, giving rise to polycentric chromosomes or centric ring chromosomes (Pfeiffer *et al.*, 2000) which are visible on metaphase preparations.

In this test, we observed also increase in the values of the total number of MN in the exposed group before exposure to 2 Gy (176 MN) scored in (167 binucleated cell) compared with (130 MN) scored in (123 binucleated cell) in the control group. The probability values of differences between means of binucleated cells with one MN, and the total number of MN were higher significantly in the exposed group than control. Our observations are in agreement with the studies of Joseph et al. (2004).

The results obtained by Thomas *et al.* (2003) validate the use of NBP frequency in binucleated cells as a biomarker of DNA damage and chromosome rearrangement. So, we use the measurement of NPB to evaluate in vivo radiation exposure of occupational or medical exposed individuals. Two folds difference in NPB for X-ray workers before exposure to 2 Gy as compared with control group.

We observed that after irradiation the values of total number of the MN in the X-ray workers (344 MN) scored in (313 Binucleated cell) whereas, in the control group were (416 MN) scored in (384 binucleated cell). Our results also showed that the frequency of cells with one MN and total number of MN for X-ray workers (28.6 and 34.3 respectively) were significantly lower than non-exposed individuals (35.6, 41.6 respectively).This observation is in agreement with previous findings of other authors, who found that the mean MN/cell for radiation workers' lymphocytes after 1 and 2 Gy irradiation was significantly lower than the control group (Gourabi and Mozdarani, 1998).

The results also showed that the frequency of NPB for X-ray workers (2.0%) were significantly lower than exposed individuals (3.6%). The inclusion of NPB may also enhance the capacity of the cytokinesis block MN assay to identify those individuals with higher DNA repair capacity after radiation.

The present study has been assessed the cytogenetic changes as manifested by 2 techniques, the rate of CA, and MN in peripheral blood lymphocytes, which are considered to be ideal systems for studying the chromosome changes of human subjects occupationally exposed to X-ray. We depend mainly on wide range of cell counts; genetic pool for each individual due to the limited size of the samples. In fact, previous evidence has been obtained that the cellular response of people exposed to low dose radiation is surprisingly stronger than would be expected in view of the amount of energy deposed and/or the fraction of cells actually traversed (Mothersill and Seymour, 2004).

In the present investigation, before exposure to 2 Gy γ-radiation, IL-1β concentration in examined X-ray medicine workers showed slightly higher significance than that in the control group. However, their levels remained within normal limits. This slight increase in IL-1β recorded in our study demonstrates the induction of limited inflammation under the effect of occupational exposure to ionizing radiation. Our results are in line with earlier studies of Klusiniski *et al.* (2006[a]).

As well, previous studies showed that serum concentration of C-reactive protein (CRP), interleukin-1β, TNF-α, IL-6 showed only slightly higher increase in X-ray department workers compared with control group **(**Hrycek *et al.*,2002 and Klusiniski *et al.*, 2005&2006[b]).

The investigation revealed that 2 Gy γ-irradiation (challenge dose) caused higher significant increase in the concentration of IL-1β in exposed group (5.1 folds) than that of the corresponding groups before irradiation. Whereas, there are higher increment difference observed for control group after irradiation as compared to the same group before irradiation (8.3 folds). The present results are in agreement with Linard *et al.* (2004) and O'Brien-Ladner *et al.* (1993). From the present results, we can suggest that IL-1 β in occupationally exposed workers may have a protective role against exposure to the challenge dose.

Cytokine-induced priming leads to increase NO production in the macrophage, so it is not surprising that radiation induced priming has the same effect. As well, the present results revealed that the NO concentration in examined X-ray medicine workers showed higher significance than that in the control group. These results are in broad agreement with previous studies of Han *et al.* (2010).

The investigation revealed that 2 Gy irradiation caused higher increase in the concentration of NO in exposed group (1.2 folds) than that of the corresponding group before irradiation. Whereas, there was higher increment observed for control group after exposure to *in vitro* 2 Gy γ-ray as compared to the same group before irradiation. These results are consistent with Ibuki *et al.* (2003). Other authors reported that NO modulates cell radiosensitivity (Verovski *et al.*, 1996), and ionizing radiation increases NO production by inducing NO synthesis (NOS) expression and stimulating constitutive NOS (Leach *et al.*, 2002). Matsumoto *et al.* (2004) and Shankar *et al.* (2006) suggested that reactive nitrogen species (RNS), particularly NO secreted from irradiated cells may initiate a signaling pathway to induce the radioadaptive response. Matsumoto *et al.* (2007) also stated that radio-adaptive responses have been observed using various end points, such as CA, mutations, and clonogenic survival. They found that radioresistance observed in the radioadaptive response can be induced by NO at extremely low concentrations, which are endogenously generated after exposure to radiation.

Conclusion

The present results pointed to the presence of cytogenetic adaptive response in medicinal workers occupationally exposed to low dose of X-ray. In addition, the results explained that NO radicals and IL -1β have a role in the induction of radio-resistance due to *in vivo* exposure that may intermediate this radiation exposure.

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الاستجابة الإشعاعية التكيفية: دلالة للتأثيرات البيولوجية من التعرض لمعدل جرعة منخفض من الأشعة السينية

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قسم الكيمياء ، كلية العلوم ، جامعة القاهرة و *قسم الأمان الاشعاعى ، هيئة الرقابة الإشعاعية و النووية و **قسم البيولوجيا الإشعاعية ، المركز القومي لبحوث وتكنولوجيا الإشعاع ، ص. ب: ٢٩ مدينة نصر ، مصر.

تعرف الاستجابة الإشعاعية التكيفية على أنها " انخفاض التأثير الضار للتعرض لجرعات الإشعاع المرتفعة نتيجة التعرض السابق لجرعات أولية منخفضة".

للتحقق من حدوث الاستجابة الإشعاعية التكيفية للعاملين المعرضين لجرعات منخفضة من الأشعة السينية ، تم دراسة الاستجابة الوراثية الخلوية و المناعية و الكيميائية الحيوية للعاملين بالأشعة السينية بعد التعرض إلى جرعة قدرها ٢ جراى من الإشعاع الجامى المعملي و مقارنتها مع أفراد المجموعة الضابطة.

تم دراسة تحليل الانحراف الكروموسومى و اختبار النويات و قياس الانترلوكين ١-بيتا و تركيزات أكسيد النيتريك قبل و بعد التعرض لجرعة ٢ جراى من الإشعاع الجامى لكلا من المجموعة المعرضة مهنيا و المجموعة الضابطة. و قد كشفت النتائج ارتفاعا في معدل الانحراف الكروموسومى و النويات و جسر النيكلوبلازم لدى العاملين بمجال الإشعاع أكثر من المجموعة الضابطة و لكن عند التعرض الى جرعة ٢ جراى من التشعيع الجامى المعملي للخلايا الليمفاوية للعاملين بالإشعاع ، سجلت النتائج انخفاضا في معدلاتها عند مقارنتها بالمجموعة الضابطة و من ناحية أخرى أظهرت النتائج ارتفاعا في تركيز الانترلوكين ١-بيتا و أكسيد النيتريك في البلازما لدى الأفراد العاملين بالإشعاع عند مقارنتها بالمجموعة الضابطة بينما كانت النتائج بعد التعرض الى جرعة ٢ جراى من أشعة جاما مرتفعة ولكنها أقل من مستوى الارتفاع في المجموعة الضابطة بعد التشعيع المعملي بالمقارنة مع نفس المجموعة قبل التشعيع.

نستنتج من النتائج السابقة وجود استجابة للأفراد المعرضين مهنيا لمعدل منخفض من الإشعاع السيني لحدوث تكيف وراثي خلوي بالإضافة إلى أن الانترلوكين ١-بيتا و أكسيد النيتريك يحدثان دورا في إحداث المقاومة الإشعاعية المستحثة بسبب التعرض المهني للعاملين.