# Application of Oxidative Stress Parameters and Alkaline Comet Assay for Monitoring of DNA Degradation Response in Healthcare Workers Exposed to Chronic Ionizing Radiation at Banha University Hospitals

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#### ABSTRACT

**KEYWORDS** Ionizing radiation,

comet assay, DNA degradation, oxidative stress

Ionizing radiation (IR) has been broadly employed in the last decades, commonly as a diagnostic and therapeutic tool. Thus, medical staff is frequently exposed to IR during work. Persistent introduction to low doses of IR can lead to genotoxicity via stimulating the generation of reactive oxygen species. This study aimed to investigate DNA degradation and evaluate the potential impact of oxidative stress status on DNA damage in healthcare workers (HCWs) working in diagnostic radiology units. The studied groups included 21 HCWs who were occupationally exposed to IR, and 20 healthy office employees selected as unexposed control subjects who were matched with the exposed group with respect to age, sex, and inclusion criteria. Malondoaldehyde (MDA) and superoxide dismutase (SOD), as oxidant and antioxidant biomarkers, were assessed in all participants. Also, DNA degradation was quantified by a comet assay (a genotoxicity marker). The results show significant elevation in MDA, reduction in SOD, and DNA degradation in the exposed group. DNA degradation was higher with increased duration of exposure. Exposed technicians had more DNA degradation than exposed nurses and physicians. A positive correlation between comet parameters (tail length, tail moment, % tailed DNA, and DNA% in the tail) with MDA and duration of exposure was noticed. While negative correlation was demonstrated between these parameters and SOD in the exposed group. IR can lead to increased DNA damage in occupationally exposed health care workers, which is positively correlated with the duration of exposure and oxidant status.

#### Introduction <sup>.</sup>

Ionizing radiation (IR) has currently been a critical concern in various fields due to its potential harmful effects on living organisms and materials. Human beings are subjected to continuous exposure to IR either from natural sources such as cosmic rays or from artificial sources, mostly diagnostic procedures, medical treatment, or occupationally during work (Thomas and Symonds, 2016; Mousavikia et al., 2023). At the present, there has been broad employment of investigational and therapeutic techniques that depend on radiology and nuclear medicine. This has led to a constant increase in the exposure of humans to IR (Stokke et al., 2017; Gaetani et al., 2018).

Ionizing radiation is beneficial in the diagnostic workup and treatment of several diseases. However, the related harmful effect due to the occupational exposure of health care workers (HCWs) cannot be overlooked. Health care workers who use IR for diagnostic and therapeutic reasons, such as those handling X-ray machines and cameras, are the most commonly employed occupational group with a potential risk of

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overexposure (Gao et al., 2020; Mousavikia et al., 2023). Being continuously exposed to low doses of IR, these HCWs are at risk of mutagenicity and carcinogenicity with proven genetic instability and chromosomal aberration in this population despite the application of protection guidelines at hospital radiology units (Feinendegen et al., 2011; Dobrzyn'ska, et al., 2014; Gaetani et al., 2018).

The biological hazards associated with the IR long-standing exposure occur due to direct DNA damage (mitochondrial and nuclear) or indirectly through releasing reactive oxygen species (ROS) that result in oxidative damage of important molecules (Pernot et al., 2012; Loseva et al., 2014; Tričković et al., 2022). It has been demonstrated that the continuous exposure of IR low doses can disturb the oxidantantioxidant balance status in the human body with increasing DNA oxidative damage (Nuszkiewiczet al., 2020).

After being exposed to IR, radiolysis of water occurs and this involves the absorbing radio waves by the molecules of water, leading to their dissociation into reactive products and generation of ROS which cause significant oxidative stress, with damaging effect on essential molecules. Proteins can undergo oxidation, leading to alter their structure and function. Oxidative damage to DNA can result in single and double-strand breaks, base modifications, and crosslinking. which interfere with replication and transcription processes. potentially leading to mutations and genomic instability. Lipid peroxidation is another major consequence of oxidative stress induced by ROS. Cell membrane fatty acids are particularly susceptible to attack by ROS with further decomposition into reactive aldehydes, which can propagate the damage by reacting with other cellular components, including proteins and nucleic acids. The integrity and functionality of cellular membranes are compromised, disrupting membrane fluidity and permeability, and impairing cellular homeostasis (Spitz et al., 2004).

The present study was performed to assess oxidant and antioxidant parameters, DNA degradation, and also to evaluate the association between duration of exposure and oxidative stress status with DNA damaging effects in a population group working in diagnostic radiology units who are occupationally exposed to IR at Banha University Hospitals.

# Subjects and methods:

This is a comparative cross-sectional human study.

# Study setting and subjects:

This study was conducted at the Radiology Department, Faculty of Medicine, Banha University Hospital and analyses of data were performed at Immunology unit, animal reproduction research institute, Egypt.

Data collection and sampling were conducted over a period of one month (from 1st March 2023 till 1st April 2023) after obtaining informed Arabic language written and signed consent letters from all of the participants for sharing in the study.

Convenient sampling was conducted on the studied population. The study included two groups; the exposed group that included 21 participants from the Radiology Department who had exposed to IR for at least one year, and the control group that included 20 age and sex-matched healthy participants those were office employees recruited from academic institutions. Sample size was calculated using OpenEpi, Version 3, open source calculator—cross sectional study was used to calculate the least required sample size at 0.05 alpha error, power of 0.80 and mean difference for SOD antioxidant ( 110.4-116.8/ml) (Mousavikia et al., 2023). The least Number required is 32 patients divided into 2 groups.

**Inclusion criteria:** The studied group was non-alcoholic, had not been subjected to IR for diagnostic or therapeutic purposes, or reported being previously exposed to other genotoxic agents during the year before the beginning of this study (Garaj-Vrhovac and Kopjar, 2003; Martínez et al. , 2010).

**Exclusion criteria:** Individuals who had previously received chemotherapy or radiotherapy and those with an acute illness like respiratory infection or diarrhea were excluded from the study for that day and were included for later sampling when they presented in healthy condition.

All participants completed a designed questionnaire including personal data (age, gender, residence, marital states, occupation, vacation, and special habits), medical history of the participants, including (health status, medical disease, previous surgery, regular drug intake, or chronic diseases). Items related to the IR occupational exposure were also included in the questionnaire, i.e., type of work they do, job duration, duration of work, duration of exposure (hours/week), total duration of exposure (hours), and type of safety used (i.e., using personal protective equipment). Additionally, the checklist also included non-occupational exposure to conditions with mutagenic risks, such as recent vaccinations or contracting risky viral diseases, alcohol or drug intake, and exposure to radiotherapy.

#### Sampling:

Both groups of the study underwent morning peripheral blood sampling via venipuncture. All samples were coded and categorized into two parts:

- 1. One part was put in a heparinized tube that was delivered to the laboratory at once and kept at room temperature with protection from light to perform the comet assays.
- 2. The other part was kept in a plain tube to obtain serum. The sample was left at room temperature for coagulation, and then centrifugation was performed at 986 g for 15 minutes to separate the erythrocytes and plasma. The serum, which was ensured to be clear and nonhemolyzed, was immediately collected and stored at (-80°C) to be used in the oxidant and antioxidant analysis [blood malondialdehyde (MDA) and superoxide dismutase (SOD)].

# Methods:

# I. Oxidant Antioxidant studied parameters:

The assessment of plasma MDA and SOD levels was done using the respective ELISA assay kits in adherence to the manufacturer guidelines. Enzyme Linked Immunosorbent Assay (ELISA) kits from Life Span Biosciences Company (LSBio), North America, and Shanghai Blue Gene Biotech Company were used for the MDA and SOD analyses, respectively.

# II. Genetic study (alkaline comet assay):

The comet assay (alkaline version, Sigma-Aldrich) was used to determine the initial damage response of DNA in peripheral blood leucocytes. The assay was explained by Singh et al. (1988) and modified by Kruszewski et al. (1994).

For analysis free comet scoring software by Autocomet.com was used to acquire images, calculate the integrated intensity profile for each cell, estimate the comet cells components and evaluates the range derived parameters.

To quantify the damage to DNA, the comet analysis included the following; Tail length (TL) i.e. the length of DNA migration is directly related to the size of the DNA fragment and shown in micrometers. It was measured from the center of the cell to the end of the tail and calculated as (tail length percentage of DNA in tail)/1.), Tail moment (TM) which was expressed in arbitrary units and calculated as: tail length X percentage of migrated DNA / 100, the percentage of tailed cells (% tailed DNA) and the percentage of DNA in the tail).

#### **Ethical approval:**

This research was commenced after review and approval by the Research Ethics Committee of Banha Faculty of Medicine (RC. 20.11.2023).

#### Statistical analysis:

The collected data were statistically analyzed using IBM SPSS statistics (V. 26.0,

IBM Corporation, USA, 2019). Categorical data were presented as numbers and percentages. Normally distributed variables (quantitative parametric measures) were expressed as mean  $\pm$  SD. After ensuring the data normality, an independent t-test was used to compare two groups, and an Analysis of Variance (ANOVA) test was used to compare more than two groups. After ANOVA, a posthoc test was used to assess the possible statistical significance between groups. Assessment of the potential association between numerical variables was done using the Pearson test. The significance levels were deemed at *a p* value of  $\leq 0.05$ .

#### **Results:**

Forty-one (41) persons were enrolled the present study; 21 persons are in occuptionally exposed to IR (study cases) and 20 who are not (control cases). Of these, 10 (47.6%) of cases and 9 (45%) of control subjects were male. The mean age was 37.9 vears for cases and 36.75 years for control subjects. Regarding occupation, 17 (81%) of cases were radiology technicians, while 14 (70%) of control subjects were office employees. The total duration of occupation was  $(132.95 \pm 114.076)$  months for cases and  $(173 \pm 123.54)$  months for control subjects, with no significant difference between the total duration of work between the two groups (Table 1).

Charac	Characteristics		rols (n=20)	Study ca	ases (n=21	
		n	%	n	%	
Sex	Female	11	55 %	11	52.4 %	
Sex	Male	9	45 %	10	47.6 %	
	Min- Max	21-	-57	2	2-56	
Age (years)	Mean ± SD	36.75 =	± 11.83	37.9	$37.9 \pm 10.12$	
	p- value		0.37	7		
	Banha	11	55%	8	38%	
Residence	Other Qalyobia cities	5	25%	8	38%	
Keshuenee	Other governates	4	20%	5	24%	
Occupation	Technician	14	70%	8	38%	
	Physician	0	0.0%	6	28.5%	
	Nurse	0	0.0%	7	33.5%	
	Employee	6	30%	0	0.0%	
	Married	11	55%	17	81%	
Marital status	Divorced	1	5%	0	0.00%	
Iviai itai status	Widow	1	5%	0	0.00%	
	Single	7	35%	4	19%	
Medical disease or	present	5	25%	4	19%	
surgery	Absent	15	75%	17	81%	
Dogular drug intaka	present	5	25%	3	14.3 %	
Regular drug intake	Absent	15	25%	18	85.7%	
Total duration of	Mean ± SD	173 ±	123.54	132.95	± 114.076	
occuption (months)	p- value		0.14	Ļ		

Table (1). Statistica	l comparison between	n the studied	population regarding	ng scio-demograph	nic data
and asso	ciated medical diseas	es (n=41).			

These characteristics are recorded at time of entry to the study. n: number of studied population, %:precentage, SD:standard deviation, Min- Max: minimum – maximum. The result is statistically significant at  $p \le 0.05$ 

Regarding the distribution of screened subjects according to the nature of occupational radiation exposure. Nine (42.85%) of them were exposed to CT, only 3 (14%) were exposed to MRI, and only 2 (9.5%) were exposed to X-ray. Seven cases (33.65%) were exposed to multiple radiation sources (CT, X-ray, MRI). The average mean  $\pm$  standard deviation (SD) of the hours of exposure per week was  $54.62 \pm 23.38$  hours, while total hours of exposure to radiation at all years of work were  $36337.52 \pm 38399.17$ hours, while there was no significant difference in duration of exposure between subjects exposed to each source of radiation (Table 2).

	Characteristics		Study cases (n=21) n (%)	Duration of exposure/week (hours /week) Mean ± SD	Total duration of exposure (hours) Mean ± SD
	CT scan	n %	9 42.85%	$54 \pm 23$	26822.2 ± 27290.2
Nature of	MRI	n %	3 14%	57.3 ± 2	$43392 \pm 24616.6$
radiation		n %	2 9.5%	40 ± 21.3	$13640 \pm 13384.4$
Multiple sources	n %	7 33.65 %	67 ± 21.8	52033.1 ± 54594.6	
Number of ho p-value	ours in all cases			$54.62 \pm 23.38$ 0.066	36337.52 ± 38399.17 0.108

Table (2	:): Radiation	related cha	racteristics	ofexposed	population	(n=21).
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CT: computed tomography, MRI: magnetic resonance imaging. The result is significant (\*) at  $p \le 0.05$ .

n: number of studied population, %:precentage, SD:standard deviation

The comet assay showed that in most control participants' cells, the DNA was compressed and maintained in normal rounded nuclei. While DNA degradation occured in radiation-exposed cases, causing nuclear DNA streaks extending from the nucleus head-forming tail region. Estimation of tailing illustrated a significant increase in exposed cases than the control participants' measured values (Tables 3) and (Figure 1).

 Table (3): Comparison between the studied groups (n=41) regarding DNA degradation by alkaline Comet assay

Parameters	Exposed cases (n=21) Mean ± SD	Control (n=20) Mean ± SD	t test	P value
Percentage of tailed cells	$23.3\pm6.57$	$10.14\pm0.833$	8.672	< 0.001**
The percentage of DNA in the tail	$9.07\pm2.95$	$4.25\pm1.374$	6.48	< 0.001**
Tail length (px)	$16.43\pm9.85$	$10.39 \pm 1.75$	2.63	0.0059*
Tail moment (units)	$1.01 \pm 0.26$	$0.48\pm0.13$	7.95	< 0.001**

n: number of studied population, SD:standard deviation, t: Student t-test.

The result is significant (\*) at  $p \le 0.05$ .

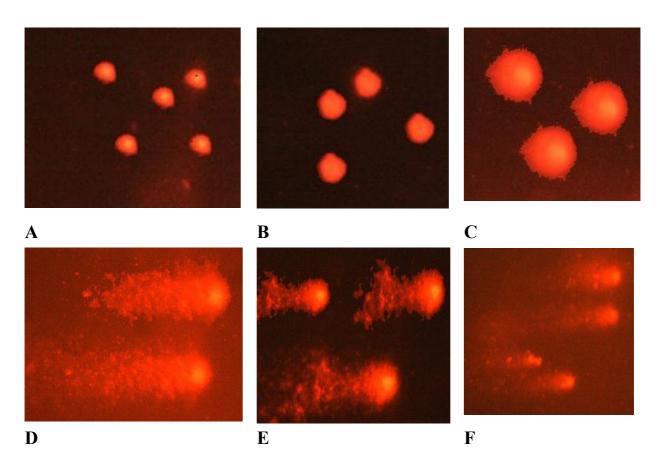


Fig. (1): shows difference of alkaline comet between control subjects (upper panel) and exposed cases (lower panels). In control subjects (A, B & C) alkaline comet showed normal nucleus. In exposed cases (D, E & F) alkaline comet showed increase in the length and intensity of the comet tail associated with a decrease in the nuclear DNA content.

Regarding oxidative stress markers, the mean levels of MDA and SOD measured in the exposed subjects were  $47.33 \pm 31.03$ nmol/ml and  $124.07 \pm 56.07$  U/L, respectively. The observed values differed significantly from values measured in control subjects, which were  $4.22 \pm 2.237$  nmol/ml and  $275.38 \pm 33.24$  U/L, respectively (Table 4).

Table (4): Comparison be	etween the studied groups	regarding oxidative stress	parameters (n=41).

Parameters	Exposed cases (n=21) Mean ± SD	Control (n=20) Mean ± SD	t test	P value
MDA (nmol/ml)	$47.33 \pm 31.03$	$4.22 \pm 2.237$	6.04	< 0.0001**
SOD (U/L)	$124.07 \pm 56.07$	$275.38\pm33.24$	-10.19	< 0.0001**

MDA: Malondialdehyde, SOD: Superoxide dismutase, n: number of studied population, SD:standard deviation, t: Student t-test. The result is significant (\*) at p ≤ 0.05. physicians and nurses. Exposure to different radiation sources (CT, MRI, X-ray) induces significant differences in the comet assay parameters among groups. Higher levels of DNA degradation are reported in cases exposed to X-rays and multiple radiation sources than those exposed to CT (Table 5).

As regard the duration of exposure among workers, the comet assay parameters were significantly increased in HCWs exposed to radiation for more than 5 years than those exposed to radiation for less than 5 years. Technicians show a significant increase in the comet assay parameters compared to

Study	% tailed cells	% of DNA in the tail	Tail length (px)	Tail moment (units)
population	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
Exposed < years (n:15)	5 19.75 ± 4.16	$7.84 \pm 2.56$	10.67± 2.56	$0.88 \pm 0.15$
Exposed > years (n:6)	<b>5</b> 32.18 ± 0.63	$12.15 \pm 0.86$	$30.85 \pm 6.3$	$1.33 \pm 0.24$
t-test	-7.16	-3.82	-10.69	-5.13
P value	< 0.00001**	0.0005**	<0.0001**	0.00002**
T (n 8) P (n 6)	$30.6625 \pm 2.877$ $21.52 \pm 1.99$	$\begin{array}{c} 11.91 {\pm}~ 0.92 \\ 9.5 {\pm}~ 0.97 \end{array}$	$26.88 \pm 9.06$ $11.55 \pm 11.29$	$\begin{array}{c} 1.26 \pm 0.24 \\ 0.99 \pm 0.05 \end{array}$
N (n 7)	$16.42 \pm 2.77$	$5.46 \pm 1.6$	$8.66 \pm 1.45$	$0.74 \pm 0.06$
f-test	56.8	54	21.7	21.4
P value	< 0.001**	< 0.001**	< 0.001 **	< 0.001 **
Post	<u>T:P</u> T:N P:N		T:P T:N P:N	T:P T:N P:N
Hoc P test valu	e <0.01 <0.01 <0.0	1 <0.01 <0.01 <0.01	<0.01 <0.01 0.6	0.012 <0.01 0.017
E1 (n 7)	$27 \pm 8.7$	$9.8 \pm 4.08$	$25.25 \pm 12$	$1.17 \pm 0.37$
E2 (n. 9)	$18.9 \pm 2.29$	$7.4 \pm 1.67$	$9.8 \pm 1.6$	$0.85 \pm 0.11$
E3 (n 3)	$24.3 \pm 1.97$	$   \begin{array}{r}     10.7 \pm 0.23 \\     11.39 \pm 0.005 \\     6.8   \end{array} $	$13.6 \pm 1.59$	$1.04 \pm 0.002$
E4 (n 2)	$28.5 \pm 2.7$		$19.6 \pm 5.8$	$1.097 \pm 0.019$
f- test	11.04		18.48	7.8
P- value	<0.001 **	0.0005*	<0.001**	0.0002*
Post	E1:E2 E4:E2	E2:E4 E2:E3	E1:E2 E1:E3 E2:E4	E1:E2
Hoc P test valu	0.048* 0.006**		E1.E2         E1.E3         E2.E4           0.0004*         0.002*         0.01*	0.01*

Table (5): Comparison of mean comet parameters among exposed population (n=21).

n: number of studied population, SD:standard deviation, t: Student t-test, f: Fisher test. The result is significant (\*) at p ≤ 0.05.

- Exposed Technician (T), Exposed physician (P), Exposed nurse (N), Multiple exposure cases (E1), CT exposed cases (E2), MRI exposed cases (E3) and X-ray exposed cases (E4)

Regarding the duration of exposure among workers, there was a significant increase in MDA and decrease in SOD in workers exposed to IR above 5 years compared to those who had less than 5 years of exposure duration. Technicicians show a significant increase in MDA and decrease in SOD than physicians and nurses. Exposure to different radiation sources (CT, MRI, X-ray) induces significant differences in MDA and SOD among groups. Higher levels of oxidative stress are reported in cases exposed to X-rays and multiple radiation sources than in those exposed to CT only (Table 6).

	Table (6): Comparison	of mean oxidative stress	parameters among ex-	posed population (n=21).
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Expo	sed population	MDA (nmol/ml)         SOD (U/L)           Mean ± SD         Mean ± SD					
Exposed < 5	years (n: 15)	$32.23 \pm 22.26$ $152.77 \pm 36.46$		-6			
Exposed $> 5$	•		$85.07 \pm 16.24$	4		$52.3\pm28.8$	
•	t-test		-5.24			6	
P value			<0.001**		<0.001**		
T (n 8)			$80.53 \pm 16.09$	9	$64.9 \pm 33.9$		
P (n 6)		42.11±17.39 132.1±18.7					
N (n 7)		$13.84 \pm 3.2$ $184.7 \pm 18.6$		- )			
	f-test	44.72 41.03					
	P value	<0.001** <0.001**					
Post Hoc te	est	T:P	T:N	P:N	T:P	T:N	P:N
	P value	<0.01*	<0.01*	< 0.01*	<0.01*	<0.01*	< 0.01*
E1 (n 7)			$66.72 \pm 37.40$	6	9	$91.16 \pm 78.52$	2
E2 (n9)			$23.28 \pm 13.47$	7	$160 \pm 20.3$		
E3 (n 3)			$61.33 \pm 6.7$		$111.7 \pm 3$		
E4 (n 2)			$66.62 \pm 0.49$	)	$95.82 \pm 1.65$		
	f- test		16.3		9.35		
	P- value		<0.001**			0.0003**	
Post		E1:E2	E4:E2	E3:E4	E1:E2		E4:E2
Hoc test	P value	0.0003**	0.0004**	0.002**	0.006**		0.012**

 MDA: Malondialdehyde, SOD: Superoxide dismutase, n: number of studied population, SD:standard deviation, t: Student t-test, f: Fisher test. The result is significant (\*) at p ≤ 0.05.

- Exposed Technician (T), Exposed physician (P), Exposed nurse (N), Multiple exposure cases (E1), CT exposed cases (E2), MRI exposed cases (E3) and X-ray exposed cases (E4)

According to the correlation between comet assay parameters and oxidative stress parameters, a positive correlation was found between comet parameters (tail length, moment, % tailed DNA, and DNA % in tail) and MDA. While a negative correlation between them and SOD was noticed, as shown in (Table 7).

Table (7): Correlation between comet assay parameters and oxidative stress parameters

	MDA (nmol/ml)	SOD (U/L)
% tailed cells	r=0.94 (P<0.001*)	r= - 0.96 (P<0.001*)
% DNA in tail	r = 0.91(P < 0.001*)	r= - 0.95 (P<0.001*)
Tail length (px)	r=0.89(P<0.001*)	r = -0.91 (P < 0.001*)
Tail moment (unit)	r = 0.91(P < 0.001*)	r = -0.95 (P < 0.001*)

Pearson correlation test is used. MDA: Malondialdehyde, SOD: Superoxide dismutase. The result is significant (\*) at p < 0.05.

Studying the effect of exposure duration on comet assay and oxidative stress parameters revealed the presence of a strong statistically significant positive correlation between the duration of being exposed to IR and the degree of DNA degradation in terms of % tailed cells, % DNA in tail, TL, and TM, and a statistically significant strong positive correlation between the duration of being exposed to IR and the degree of oxidative stress illustrated by increased oxidative stress product MDA and decreased antioxidant activity of SOD (Tables 8 & 9).

 Table (8): Correlation between measured comet assay parameters and total duration of occupational exposure to IR

Comet assay parameters	Total occupational exposure duration
% tailed cells	r=0.89 (P<0.001*)
% DNA in tail	r= 0.79 (P<0.001*)
Tail length	r=0.97 (P<0.001*)
Tail moment	r=0.87 (P<0.001*)

 Table (9): Correlation between measured oxidative stress parameters and total duration of occupational exposure to IR

Oxidative stress parameters	Total occupational exposure duration
MDA	r=0.9 (P<0.001*)
SOD	r= - 0.92 (P<0.001*)

MDA: Malondialdehyde, SOD: Superoxide dismutase.

#### **Discussion:**

Ionizing radiation (IR) is widely used, and its impact on human health has been an issue of continuous discussion since it was discovered. Sources of IR are frequently utilized in medicine for diagnostic and purposes. The long-standing therapeutic exposure of the HCWs to low doses of IR is a matter of concern. Currently, continuous investigations have been done to minimize its health-related effects and the necessity of protection against IR sources is progressively stressed to keep safe occupational conditions (Fazel and Einstein 2020).

In the current study, there was a notable inter-individual variation regarding scio-demographic data and medical diseases among the exposed and the control. This was also described previously by the studies of Speit et al. (2003) and Kopjar and Garaj-Vrhovae (2005). The explanation for such results could be attributed to variation in the genomic sensitivity or a combination of nongenetic and genetic factors (Fachin et al., 2009).

With respect to oxidative stress markers, this work showed significantly increased MDA levels and significantly reduced levels of SOD levels in the exposed group compared to the control group. These findings were consistent with the study of Malekirad et al. (2005), who reported an increase in the MDA levels with continuous exposure to low doses of IR. Similarly, El-Benhawy et al. (2016) observed significant elevation in the levels of oxygen free radicals in the exposed workers compared to the nonexposed ones. Additionally, Kłucinski et al. (2008) demonstrated that long-standing exposure to low levels of IR was associated with the reduction of antioxidant levels. Kłucinski et al. (2008) and Russo et al. (2012) stated reduced activity of SOD in exposed individuals. In the same context, El-Benhawy et al. (2020) found that the total antioxidant

status was significantly reduced in the exposed HCWs.

On the other hand, other studies revealed significant elevation in the MDA and SOD levels in exposed HCWs compared to the non-exposed (Kumar et al., 2016; Ahmed et al., 2019).

The present study demonstrated that IR occupational exposure had remarkable DNA-damaging effects illustrated by significant increased TL, TM, % tailed DNA, and percentage of tail DNA in peripheral blood leukocytes of the exposed group compared to the control participants.

These findings aligned with the study of Dobrzyn'ska et al. (2014), who stated that there is a high development of DNA damage in HCWs with occupational exposure to IR. Also, Maffei et al. (2004) revealed an elevated rate of chromosomal aberration in peripheral lymphocytes of HCWs with occupational exposure to IR low doses.

The current study revealed a positive correlation between the oxidative stress (MDA) parameter and DNA degradation, as illustrated by comet parameters. But a negative correlation was detected between them and SOD.

In healthy individuals, there is a balance between the generation of ROS and the antioxidant activity. This balance is disturbed by the long-standing exposure to IR since it can elicit indirect effects upon cells by generating ROS and triggering oxidative stress (Chakraborty et al., 2009), which is implicated in several pathological processes encompassing inflammation, necrosis, fibrosis, DNA damage, and carcinogenesis (Spitz et al., 2004; Zakeri and Hirobe, 2010).

The current results revealed that MDA and DNA damage levels were positively correlated with the duration of exposure to IR. In accordance, Gao et al. (2020) in their linear regression models determined that large exposure duration and dose of exposure to IR increased oxidative stress. Furthermore, Ahmed et al. (2016) reported the positive correlation between the levels of antioxidants and the duration of occupational exposure as well as the lifetime IR dose. However, this correlation was not found for MDA. On the contrary, Dobrzyn'ska et al. (2014) found that there was no significant association between the DNA damage levels and the duration of IR exposure, with a significant increase in the DNA in both who worked for more or less than 10 years.

The current study revealed that technicians had more exposed DNA degradation, increased MDA, and reduced SOD activity than exposed nurses and physicians. In the same context, the same findings were reported by Dobrzyn'ska et al. (2014) who observed that the highest levels of DNA degradation in the nuclear medicine technicians compared to the other exposed HCWs. The highest DNA degradation levels found in technicians could be related to the fact that they are working very near the patients and the IR sources, nearly for all of their shift time.

# **Conclusion:**

Occupational exposure to IR can produce an increase in DNA degradation in peripheral blood leukocytes of HCWs who are occupationally exposed to IR in their diagnostic radiology units. Also, we conclude that there is an imbalance in oxidant and antioxidant status, leading to increased DNA-damaging oxidative effects as illustrated by increased MDA and decreased SOD. Additionally, our study concluded the presence of a positive correlation between the DNA damaging effect and the exposure duration.

#### **Recommendations:**

To ensure occupational HCWs safety, we recommend;

- 1- Workshops and awareness campaigns directed to HCWs especially nurses and technical staff to explain the dangers of exposure to IR with confirmation on the necessity of commitment to strict application of radioprotection procedures and follow safety instruction to restrict the health hazards during occupational exposure to IR.
- 2- Rotation of workers through all tasks in order to distribute occupational exposures more uniformly over the time and prompt for radiological workers not to exceed their annual limit of exposure.
- 3- Continuous bio-monitoring of HCWs.

Conflicts of interest: No conflict of interest.

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تطبيق معاملات الإجهاد التأكسدي وفحص المذنب القلوي لرصد إستجابة تكسير الحمض النووي لدى العاملين في مجال الرعاية الصحية المعرضين للإشعاع الأيوني المزمن في مستشفيات جامعة بنها

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تم استخدام الإشعاع الأيوني (IR) على نطاق واسع في العقود الماضية، كأداة تشخيصية وعلاجية. وبالتالي، يتعرض الطاقم الطبى في كثير من الأحيان للأشعة الأيونيية أثناء العمل يمكن أن يؤدي التعرض المستمر لجرعات منخفضة من الأشعة الأيونيية إلى السمية الجينية من خلال تحفيز توليد أنواع الأكسجين التفاعلية. تهدف هذه الدراسة إلى دراسة تكسير الحمض النووي وتقييم التأثير المحتمل لحالة الإجهاد التأكسدي على تلف الحمض النووي لدى العاملين في مجال الرعاية الصحية (HCWs) العاملين في وحدات الأشعة التشخيصية. شملت المجموعات التي تمت در استها ٢١ من العاملين في مجال الرعاية الصحية الذين تعرضوا مهنيًا للأشعة الأيونيية ، وتم اختيار ٢٠ من موظفي المكاتب الأصحاء كمجموعة مراقبة غير معرضة تمت مطابقتها مع المجموعة المعرضة فيما يتعلق بالعمر والجنس ومعايير الاشتمال. تم تقييم مالونولدهايد (MDA) و وسوبر أكسيد ديسميوتاز الأكسيد (SOD)، كمؤشرات حيوية مؤكسدة ومضادة للأكسدة، في جميع المشاركين. أيضا، تم قياس كمية تكسير الحمض النووى عن طريق فحص المذنب (علامة السمية الجينية). أظهرت النتائج ارتفاعًا في MDA، وانخفاضًا في SOD، وتكسير الحمض النووي في المجموعة المعرضة. وكان تكسير الحمض النووي أعلى مع زيادة مدة التعرض. كان لدى الفنيين المعرضين تكسير الحمض النووي أكثر من الممرضات والأطباء المعرضين. ولوحظ وجود علاقة إيجابية بين معالمات المذنب (طول الذيل، وعزم الذيل، ونسبة الحمض النووى الذيل، ونسبة الحمض النووي في الذيل) مع MDA ومدة التعرض. بينما ظهر ارتباط سلبي بين هذه معالمات و SOD في المجموعة المعرضة. يمكن أن يؤدي الأشعة الأيونية إلى زيادة تلف الحمض النووي لدى العاملين في مجال الريحاية الصحية المعرضين مهنيًا، وهو ما يرتبط بشكل إيجابي بمدة التعرض وحالة الأكسدة