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Assessment of DNA Damage in Blood of Iraqi Museum Workers

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Abstract: This study was done to evaluate the risk of natural alpha emitter's concentrations in blood of museum employees and any potential DNA damage that may be linked to it. The current research was conducted on employees in State board of antiquities and heritage, Abd al-karim qasim museum and Iraqi museum. Blood samples were collected from 30 participants in employees group and 30 participants in controls group that was collected from general population. Alkaline comet assay has been used to assess DNA damage in lymphocytes of participants. The results showed higher DNA damage and high comet L/W value in employees (1.56) compared with control group (1.1).The smokers have increased average L/W value compared to participants who never smoked in employee group. From this study concluded based on these results, exposure of museum employees and archaeologists to natural ionizing radiation may have increased risk to DNA damage and cancer compared to non-occupational employees.

Keywords: Comet assay, DNA damage, Lymphocytes, Museum, Radon gas

Radiation is known for causing DNA damage (Yusuf et al 2020, Surniyantoro et al 2020). It is also a known carcinogen Some studies has also suggested that DNA damage can also be caused by chronic low level natural radiation (Sinitsky and Druzhinin, 2014, Walczak et al 2020). In addition to that, a study has found adoptive response in people who live in chronic low dose natural radiation (Kumar et al 2015, Mohammadi et al 2006). High radon concentrations has been measured previously in old heritage buildings (Nastro et al 2018, Mansor and Rashid 2020), the state board of antiquities and heritage and the Iraqi museum. The radon levels were measured and an average of 702 Bq/m^3 has been obtained very similar to the study mentioned above. Likewise, the study was carried in Abd al-karim qasim antique museum building employees. In the current study, the risk of high natural alpha emitter's concentration was assessed by the help of comet assay to detect any genotoxic effects of high alpha radiation compared to the controls of general population. Comet assay is selected based on many researches that suggested it to be suitable for DNA damage and genotoxic effects caused by ionizing radiation and more importantly due to the effects of ionization that form ions when interacting with the target materials and these ions is attracted by applying voltage to reveal the comet tail that are composed of damaged DNA fragments (Ostling and Johanson 1984) and even used to detect environmental radiation damage of low doses (Azqueta et al 2009, Druzhinin et al 2015). Alkaline comet assay was used to detect more DNA damage types than the neutral comet (Singh et al 1988). The aim of this study is to find a link between the type of occupation and DNA damage.

MATERIAL AND METHODS

The current research was approved by the institutional review board (IRB) in Al-Nahrain school of medicine. Experiment was in accordance with Helsinki Declaration of 1975. Written Informed consents were signed from each participant to be enrolled in the research. This research enrolled 60 participants in total divided into two groups: 30 people were in the first group collected from general populations. 30 participants were in the second group from the sites. All participants answered detailed questionnaire involving general personal information and also included years of work, smoking habits, if diagnosed or treated from cancer, recent radiation exposure, current infections, family history or genetic diseases and chronic diseases. This study was carried out as mentioned previously in State board of antiquities and heritage, Iraqi Museum and Abd al-karim qasim museum all located in Baghdad, Iraq.

The protocol that is used in this study is similar to a previous protocol (Dhawan et al 2021). In short, after preparation of solutions as described in the previous reference, comet assay slides were prepared by dipping them in methanol and slides wiped clean and left to completely dry. To cover slides, normal melting agarose (NMA) used to cover the slides. The slides were kept overnight to adhere and solidify. Using disposable syringe and needle, 3 ml of vein blood were drawn and placed

immediately into a 3 ml EDTA tubes. The blood was saved in cold container and transported to refrigerator to preserve it. Next, either lymphocytes are isolated, or can use whole blood both with good results (Singh et al 1988). After the first layer was solidified, low melting agarose (LMA) and blood mixture were added in second layer and LMA for the third layer and allow the agarose to solidify between layers and coverslips were replaced and now the slides are prepared. Afterwards, lysing solution were used to dip slides in it and kept it for 2 hours in refrigerator at 4°C. When time is done, slides were placed on the box arranging them together with no spaces between them. Allow them for 20 minutes in electrophoresis solution before performing electrophoresis at24 volts, 300 mill amperes for 30 minutes. After the electrophoresis is performed, the slides were placed in neutral solution to end the reactions for 5 minutes and repeated for 3 times. Lastly, stained the slides with ethidium bromide stain and kept the slides in the stain for 5 minutes then the slides were immersed in distilled water and coverslip was placed then observed under fluorescent microscope. The images were taken and calculations are made on these images using Image J program.

RESULTS AND DISCUSSION

Results were calculated based on the circular shape of cells. L/W value represent L for length of the circular cell and divided that number by width represent the width of circle with 90° angle to each other representing a + sigh. Considering these facts, the L/W value of 1 is the normal preferable value. When this value increases, there is more DNA damage and a comet tail was formed.Average L/W values of comet assay were observed in the blood of employees equal to (1.56) compared to control group (1.1). Employees on average have more DNA damage therefore longer comet tails and L/W value > 1. The L/W that is represented here is also representing the average of multiple random cells selected in the slide for each participant. Table 1 represents each participant and information about them and their resulted L/W e in employee group and Table 2 for control group. Numbering for participant is done according to the time of blood donation.

Refer to Figures 1 and 2; represent the L/W value for each participant. Figure 1 represent employees group and Figure 2 represent controls group. The employees group has more DNA damage than the control group. There was no value that is less than 1. There was an association between smoking and increased DNA damage observed in employees group.The smokers have increased average L/W value compared to never smokers in employee group (Fig. 3).

High alpha emitters concentrations like indoor radon gas, together with high radionuclides and other chemicals d in cigarettes increase the DNA damage that a person is exposed to and increases the chance of developing lung cancer by folds for smoking personnel (Who Handbook bn Indoor Radon A Public Health Perspective 2009). Workers in museums are constantly in contact with dusty old antiques or investigate ancient buildings. These antiques are usually made of natural earthy materials that contain natural

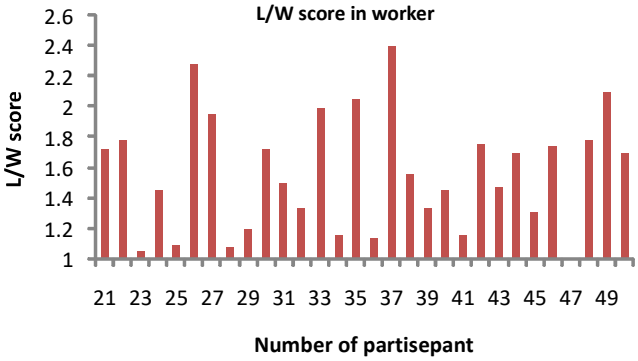

Fig. 1. L/W value for each employee

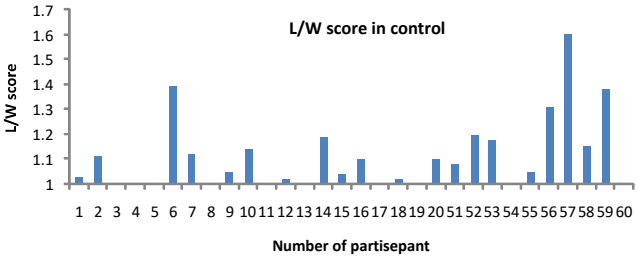

Fig. 2. L/W value for control

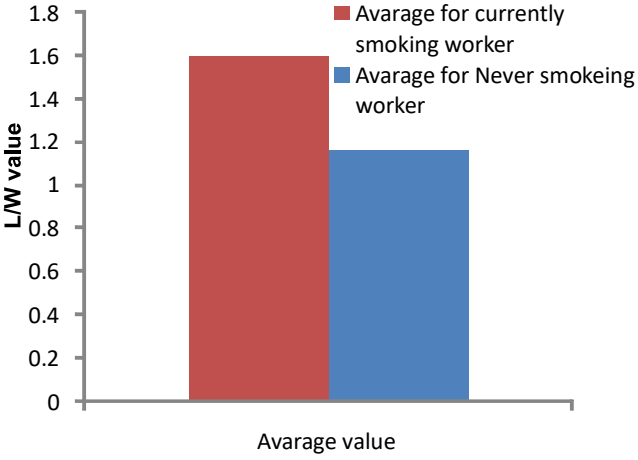

Fig. 3. Average L/W value for smokers compared to average value for non-smokers in employees group

Table 1. Number of participants and their age, duration of employment and L/W for each participant in employee group

No. of participant	Age (years)	Duration of employment (Years)	L/W	No. of participant	Age (years)	Duration of employment (Years)	L/W
21	32	1	1.72	36	29	1	1.14
22	30	1	1.78	37	26	1	2.4
23	32	1	1.05	38	30	1	1.56
24	29	1	1.45	39	28	5	1.33
25	53	28	1.09	40	41	7	1.45
26	50	1	2.28	41	29	1	1.16
27	24	1	1.96	42	55	25	1.76
28	30	1	1.08	43	55	34	1.48
29	45	20	1.19	44	31	9	1.7
30	34	6	1.72	45	49	20	1.31
31	47	13	1.5	46	34	1	1.75
32	59	28	1.34	47	38	8	1
33	59	18	1.99	48	56	2	1.78
34	27	1	1.16	49	34	7	2.1
35	26	1	2.05	50	22	1	1.7
Average	-	-	-	-	37.8	8.16	1.56

Table 2. Number of participant age and L/W value for control group

No. of sample	Age (years)	L/W	No. of sample	Age (years)	L/W
1	32	1.03	16	36	1.1
2	43	1.11	17	35	1
3	31	1	18	17	1.02
4	49	0.94	19	21	1
5	48	1	20	17	1.1
6	52	1.39	51	62	1.08
7	57	1.12	52	65	1.2
8	44	1	53	34	1.18
9	25	1.05	54	38	1
10	65	1.14	55	45	1.05
11	40	1	56	34	1.31
12	23	1.02	57	41	1.6
13	20	1	58	40	1.15
14	52	1.19	59	45	1.38
15	23	1.04	60	47	1
Average	-	-	-	39.36	1.1

radioactivity and may have dangerous materials that was unknown in old times to be of damage to human). The ignorance of safety and regulation laws and environmental contaminations often may increase contaminant in air that may damage health and cause the detected DNA damage and may become an opportunity for developing cancer. In general, soldiery lifestyle that are common in middle eastern countries, with a non-balanced diet and common health misconceptions can also be a significant factors for increased DNA damage observed (Ames 2001, Badran and Laher 2011, Mohammad 2021).

CONCLUSION

The outcome of this study demonstrated that DNA damage in employees working in museums has more DNA damage than other occupations. These participants have no signs or

symptoms due to current disease, and have no genetic diseases that causes impaired DNA repair, not diagnosed or treated from cancer and no recent radiation exposure. The exposure to high natural radioactivity and type of occupation may contribute to the increased DNA damage indicated in this current research.

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