NUKLEONIKA 2024;69(3):143–149
doi: 10.2478/nuka-2024-0021

ORIGINAL PAPER

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Determination of uranium concentration in blood samples of women with breast cancer in Babylon Province of Iraq using CR-39 nuclear track detector

Haider O. Essa,
Khalid H. H. Al-Attiyah,
Anees A. Al-Hamzawi

Abstract. The incidence and prevalence of breast cancer in Iraq are alarming. Breast cancer is one of the most common cancers among Iraqi women, and its rates have been steadily increasing over the years. The exact reasons for the high incidence are not yet fully understood, but it is believed to be influenced by a combination of genetic, environmental, and lifestyle factors. The research objectives of this study revolve around two main goals. Firstly, the study aims to establish baseline values for the amount of uranium present in blood samples. Secondly, the study aims to assess the potential relationship between uranium levels in blood and the development of cancer. The investigation includes 16 blood samples from women diagnosed with breast cancer and 20 blood samples from women without breast cancer. The nuclear fission track analysis method using CR-39 solid-state nuclear track detectors will be employed to analyze the uranium contents in women's cancer blood (CB) samples. The methodology adopted for this study involved utilizing the SPSS program to conduct a comprehensive statistical analysis. The results of the study indicate that there is a variation in uranium concentration among both the patient women and healthy women. The uranium concentration among patient women ranged from 3.259 ppb to 1.918 ppb, while among healthy women, it varied from 2.105 ppb to 0.59 ppb. These findings suggest that there may be a correlation between the presence of certain health issues and higher uranium levels.

Keywords: Breast cancer • CR-39 • Environmental pollution • Human blood • Uranium levels

H. O. Essa✉, K. H. H. Al-Attiyah
Department of Physics, College of Science
University of Babylon, Hilla, Iraq
E-mail: headeromran@yahoo.com

A. A. Al-Hamzawi
Medical Physics Department, College of Science
Al-Mustaqbal University, Hilla, Iraq

Received: 6 August 2023
Accepted: 20 February 2024

Introduction

Radioactive elements such as uranium and its daughter nuclides have the potential to harm humans due to their toxicity and emission of ionizing radiation. Uranium, found abundantly in nature, is primarily composed of the uranium-238 isotope. This isotope dominates the uranium present in the natural world and serves as the starting point for a series of radioactive decays, leading to the formation of stable lead-206. This process involves the formation of 13 intermediate radionuclides that emit beta or alpha particles, with some also emitting gamma particles at lower energy levels [1, 2]. Human activities, including phosphate fertilizer application, phosphate mining, atomic testing, uranium mining, and the entire nuclear fuel cycle, have significant consequences on the environment in terms of radionuclide pollution. Radionuclides are radioactive isotopes that result from these activities and can have detrimental effects on ecosystems and human health [3–5]. Rocks and soils contain various naturally occurring radionuclides, albeit in relatively low concentrations [4]. Depleted uranium (DU) is a uranium compound that is generated as a result of uranium enrichment. It is called "depleted" because it contains a lower proportion of the uranium-235 isotope compared to natural uranium. DU is mildly radioactive, with an activity level of approximately 60% of natural uranium. Its production is primarily associated with the nuclear fuel cycle and the manufacture of nuclear weapons. Understanding the background of DU is crucial to comprehending its properties, applications, and potential environmental and health concerns [6].

During the Gulf War and Iraq War 2003, the use of munitions containing DU by the Coalition Forces raised international concerns. The fallout from these conflicts resulted in DU contamination of cars and soil in specific areas. As a result, global communities became worried about the potential health consequences posed by DU residues. The international concerns surrounding DU contamination stem from the fear of long-term health risks for individuals living near these damaged regions. The potential for environmental implications is also a significant factor contributing to these concerns [7]. Uranium exposure can occur through various routes, including inhalation, ingestion, and skin puncture. These routes are the most common pathways for uranium to enter the body [8]. The release of uranium from the bones and other organs into the bloodstream is linked to a wide variety of diseases and conditions [9, 10], including cancer, skin diseases, congenital abnormalities, respiratory disorders, leukemia, kidney failure, and even some diseases for which no cure has yet been discovered. Recent studies reveal that heavy metals such as the radioactive nucleotide uranium cause oxidative stress by increasing the production of reactive oxygen species (ROS) [11, 12]. This oxidative stress is a fundamental contributor to genotoxicity, autophagy, apoptosis, and DNA damage.

On the other hand, a few circumstantial indications indicate that uranium may impede the DNA repair process. Exposure to uranium has been linked to DNA damage induction and repair disruption [13, 14]. When determining the uranium content in a patient's blood, it is common practice to use a solid-state nuclear track detector [15]. According to Fleischer *et al*. [16] hypotheses, solid-state track recording materials that were squeezed together and in contact with both films were exposed to thermal neutron irradiation, known as the fission-track technique. This method seems ideal for analyzing uranium levels in human blood for precise quantification. In this investigation, the CR-39 nuclear track detector will be used to compare the amounts of uranium in the blood of women diagnosed with breast cancer and a healthy control group.

Materials and procedures

Study area

The governorate of Babil, also referred to as Babylon Province, is situated in the central part of Iraq. It covers a total area of 5119 km^2. This region is geographically positioned in a central location within the country, providing convenient access to other major cities and regions [17].

Sample selection

The blood sample-selection criteria for this study involved obtaining 16 blood samples from women diagnosed with breast cancer and 20 blood samples from women who do not have breast cancer. The samples were obtained from Imam Sadiq Hospital and the Babylon Oncology Center in Babylon Province. The age distribution of female patients in this study revealed a mean age of 40.94 ± 3.28 years, ranging from 25 years to 70 years. Comparatively, the mean age of healthy women was found to be slightly lower at 37.6 ± 2.72 years and with an age range of 22–70 years. The study involved a carefully selected group of female breast cancer patients who formed the basis of the research. The participants were chosen based on predetermined criteria to ensure the validity and reliability of the study findings. These criteria took into account factors such as age, confirmed diagnosis of breast cancer, absence of previous exposure to radiation therapy or chemotherapy, and no known history of occupational uranium exposure. The patient-selection process for this study involved randomly choosing individuals from a list of eligible candidates provided by healthcare facilities. This method ensured that a diverse range of patients representing different age groups was included in the study. The study followed the Declaration of Helsinki's [18] tenets of responsible research practice. The operation was performed with the patient's informed verbal and analytical permission. A local ethics committee had a chance to review the research protocol, subject information, and permission form, and they gave their stamp of approval (Babil Health Directorate).

Methodology of the experiment

Fission track analysis is a well-established technique used to examine the fission tracks produced by the spontaneous fission of uranium atoms in a material. The CR-39 detector is a sensitive plastic nuclear track detector used in fission track analysis. Pershore Moulding Ltd supplies it in the United Kingdom. This detector can record and retain fission tracks produced by the spontaneous fission of uranium atoms. These fission tracks can be visualized and analyzed, allowing us to determine the presence and concentration of uranium in blood samples. The CR-39 detector offers a reliable and precise method for assessing uranium content in various materials. The preparation of blood samples involved several steps to ensure their suitability for analysis. First, the samples were collected from the participants. Next, the blood samples were carefully transferred into Petri dishes and labeled with numbers assigned to the participants in the study, as shown in Fig. 1.

Following the sample-preparation phase, the samples were placed in an electric oven and heated

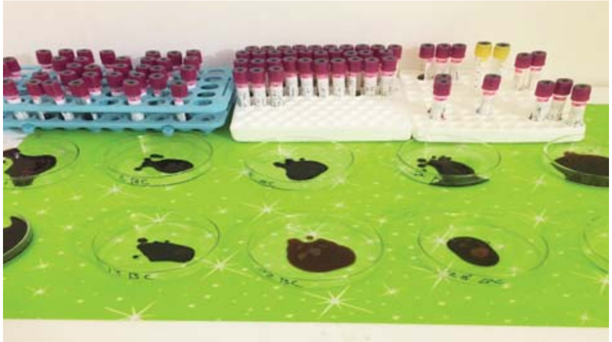

Fig. 1. The preparation of blood samples.

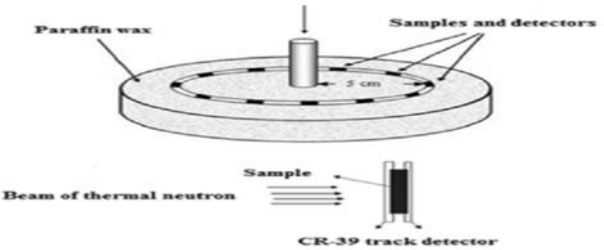

Fig. 2. Samples and detectors are exposed to a neutron flux [19].

to a temperature of 100°C. This temperature was sustained for approximately 13 min. Once the samples are heated, a hand mill is utilized to grind them into a fine powder. After the heating and grinding process, the resulting samples were carefully filtered to remove any larger particles and to ensure the desired grain-size uniformity. This was achieved by employing a fine mesh with a pore size of 75 μm. The blood powder obtained was mixed with 0.5 g of blood powder and 0.1 g of methylcellulose. Next, the mixture was compressed into a compact pellet measuring 1 cm × 1.5 mm in size. This compression process ensured the stability and integrity of the pellet. In the final stage of preparation, CR-39 detectors were strategically placed on each side of the compressed blood pellet. These detectors would capture and record the latent damage caused by the uranium-235 (n,f) reaction. The pellets were then positioned in a paraffin wax dish, placed at a distance of 5 cm from the Am–Be neutron source. The neutron source had a thermal flounce of 3.02×10^9 n·cm^{-2}. Finally, the irradiation process was conducted for 7 days. The setup for irradiating the samples and detectors to the neutron source is shown in Fig. 2. The experimental method for determining the uranium content is identical to the one previously described [2, 15, 20].

After the detectors have been irradiated and etched in NaOH-etchant solution, the fission track density is recorded using an optical microscope at a magnification of 400×, as shown in Fig. 3. The density of the fission tracks is then determined using Eq. (1). This equation takes into account the number of observed fission tracks and the area of the detector that has been analyzed. By applying this equation, the fission track density can be accurately calculated, allowing for a quantitative assessment of the irradiated detectors.

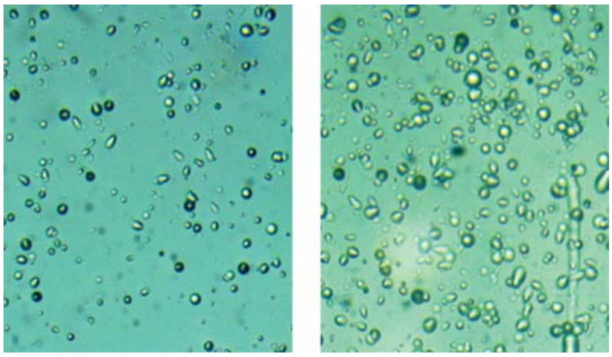

Fig. 3. (a) The nuclear tracks on the CR-39 detector captured under a microscope for healthy women. (b) The nuclear tracks on the CR-39 detector captured under a microscope for patient women.

$$\text{track density } (\rho) = \frac{\text{average of total tracks}}{\text{area of field view}} \tag{1}$$

Estimations of uranium content

The calibration of the detectors was carried out using standard samples with known uranium concentrations (0.5, 1.5, 3, and 4 ppb) provided by the International Atomic Energy Agency (IAEA). This calibration process is crucial in determining the concentration of uranium in blood samples using CR-39 detectors. Calculation of the uranium concentration in blood samples using CR-39 detectors involves analyzing the recorded track densities around the sample pellet and comparing them to those around a standard pellet. By utilizing this comparison, the uranium concentration can be determined. This is achieved by applying the appropriate mathematical relation, as described in Eq. (2). The recorded track densities serve as a direct measure of the uranium concentration in the blood sample. This method provides a reliable and efficient way to quantify the amount of uranium present, allowing for accurate assessment and analysis of uranium contamination in blood samples. The use of CR-39 detectors ensures precise measurements, enhancing the overall reliability and accuracy of the results obtained [21, 22]:

$$C_x = C_s \frac{\rho_x}{\rho_s} \frac{I_s}{I_x} \frac{R_s}{R_x} \tag{2}$$

C_s and C_x represent the uranium concentrations (ppb) in the standard and unknown samples, respectively. Similarly, the density of fission tracks (tracks·mm^{-2}) in the standard and unknown samples is denoted as ρ_s and ρ_x, respectively. Calculation of isotopic abundances involves determining the relative amounts of uranium-238 and uranium-235 in standard and unknown samples. The isotopic abundances are represented by I_s and I_x for uranium-238 and uranium-235, respectively. The range depths of fission fragments are denoted as R_s and R_x for standard and unknown sample detectors, respectively. These values represent the depth at which the fragments penetrate the detector material and provide insights into the energy deposition and behavior of

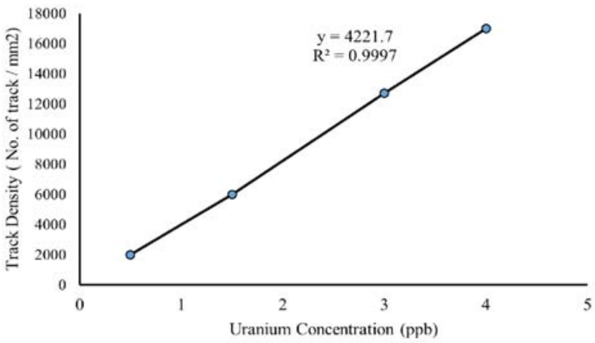

Fig. 4. The relationship between the uranium concentration of the standard blood samples and track density [24].

the fragments. The factors R_s/R_x and I_s/I_x are taken as unity [23]; hence, Eq. (2) turns into:

$$C_x = C_s \frac{\rho_x}{\rho_s} \quad (3)$$

The calculation of the (ρ_s/C_s) value involves determining the ratio between the track density and the uranium concentration in the standard samples as shown in Fig. 4.

Analysis of statistics

Version 26.0 of the IBM SPSS Statistcs program was used for the statistical processing and analysis. An independent t-test was carried out using the data from both groups to determine whether there was a statistically significant difference in the probability level (P).

Results and discussion

The study investigated the uranium concentrations in blood samples from 16 women diagnosed with breast cancer. The findings, summarized in Table 1, indicate that the highest recorded uranium concentration was 3.259 ± 0.20 ppb, detected in sample CB02 from a woman aged 36 years. Conversely, the lowest recorded uranium concentration was 1.918 ± 0.21 ppb, observed in sample CB03 from a woman aged 46 years. In analyzing the data from Table 1, it is evident that there are notable differences in uranium concentrations between younger and older women. Young women display elevated levels of uranium compared to their older counterparts. This observation suggests that age might have an influence on uranium accumulation within the body. The exact reasons for this disparity require further investigation, but it is worth noting that uranium exposure can occur through various pathways, including ingestion, inhalation, and external contact. Therefore, it is possible that the variations in uranium concentrations could be attributed to differences in lifestyle, occupational exposure, and dietary habits [25].

Table 1. Levels of uranium in female patients' blood samples

Sample code	Age (years)	Cancer type	Uranium concentration (ppb) ± SD
CB01	33	Breast	2.681 ± 0.20
CB02	36	Breast	3.259 ± 0.20
CB03	46	Breast	1.918 ± 0.21
CB04	42	Breast	2.024 ± 0.30
CB05	70	Breast	2.541 ± 0.48
CB06	34	Breast	2.444 ± 0.49
CB07	42	Breast	2.260 ± 0.31
CB08	45	Breast	2.628 ± 0.36
CB09	45	Breast	2.374 ± 0.23
CB10	44	Breast	2.199 ± 0.30
CB11	25	Breast	2.742 ± 0.37
CB12	45	Breast	2.812 ± 0.34
CB13	67	Breast	2.462 ± 0.28
CB14	26	Breast	2.216 ± 0.29
CB15	27	Breast	2.345 ± 0.24
CB16	28	Breast	2.516 ± 0.27
Mean ± Std Error			2.463 ± 0.30

CB, cancer blood.

Table 2. Uranium levels in healthy women's blood

Sample code	Age (years)	Uranium concentration (ppb) ± SD
HB01	53	1.080 ± 0.34
HB02	45	1.501 ± 0.28
HB03	40	0.905 ± 0.33
HB04	50	1.773 ± 0.43
HB05	23	1.045 ± 0.28
HB06	38	0.590 ± 0.22
HB07	36	0.712 ± 0.27
HB08	40	1.816 ± 0.43
HB09	42	1.036 ± 0.37
HB10	22	0.958 ± 0.28
HB11	23	1.010 ± 0.28
HB12	24	1.387 ± 0.27
HB13	30	2.105 ± 0.27
HB14	46	0.817 ± 0.24
HB15	38	1.510 ± 0.24
HB16	40	1.308 ± 0.28
HB17	29	0.940 ± 0.26
HB18	70	0.782 ± 0.30
HB19	23	0.993 ± 0.31
HB20	40	1.851 ± 0.35
Mean ± Std Error		1.200 ± 0.30

HB, healthy blood.

Additionally, the uranium-concentration analysis was conducted in 20 blood samples collected from women without breast cancer, as presented in Table 2. The recorded uranium concentrations in Table 2 varied between 0.59 ± 0.22 ppb and 2.105 ± 0.27 ppb. The mean uranium concentration across all the samples was found to be 1.205 ± 0.30 ppb. These results provide valuable insight into the presence and distribution of uranium in the human body. As mentioned earlier, no significant correlation was observed between the age of women and the levels of uranium present. Similarly, this lack of observed relation was also present in women without a diagnosis of breast cancer. Various factors can influence the concentration of uranium in human blood. These factors include lifestyle choices, dietary habits,

Table 3. The comparison of the present work results with those from previous studies

Country	Mean uranium concentration (ppb)	Reference
Iraq/Babylon	1.69 ± 0.31 ppb (leukemia patients) 0.83 ppb (healthy group)	[33]
Iraq/Babylon	1.02 ± 0.26 ppb (blood smokers) 0.86 ± 0.17 ppb (non- smokers)	[34]
Iraq/Babylon	1.24 ± 0.29 ppb	[35]
Russia	0.04 mg/l (occupationally exposed workers)	[36]
United States	0.025 mg/l	[37]
Iraq/Babylon	2.463 ± 0.30 ppb (women with breast cancer) 1.205 ± 0.30 ppb (women without breast cancer)	Present work

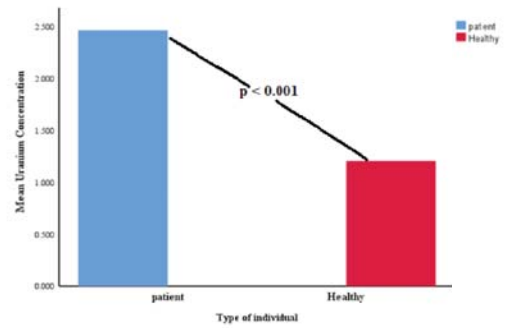

Fig. 5. Mean uranium concentrations in healthy and cancer patients groups.

contamination exposure levels, and other potential influencing factors [26, 27]. The spread of radiation contamination in Iraq after the 2003 invasion was a significant consequence of the military intervention. The invasion resulted in the destruction of institutions and research centers, including the Al-Tuwaitha nuclear site, which played a crucial role in the dispersal of radioactive materials and isotopes. These sources of radiation became uncontrolled and posed a serious threat to both the environment and the population [28, 29].

The comparison of uranium concentration in women with breast cancer to those without cancer, as shown in Tables 1 and 2, clearly indicates a significant increase in uranium levels in women with breast cancer as shown in Fig. 5. This finding suggests a potential link between uranium toxicity and the development of breast cancer. The data presented in Table 1 show the measured uranium concentration in women diagnosed with breast cancer, providing a baseline for comparison. Table 2, on the other hand, presents the uranium concentration in women without breast cancer, serving as a control. By examining both tables, it becomes evident that women with breast cancer exhibit higher levels of uranium compared to those without the disease. This comparison provides valuable evidence supporting the notion that elevated uranium levels may play a role in the development of breast cancer. Figure 5 demonstrates a significant finding that the average uranium concentration in the blood samples of cancer patients is notably higher in comparison to that of healthy individuals. This statistical distinction ($P < 0.001$) has been determined using an independent sample t-test, providing compelling evidence of a clear connection between uranium levels and the development of cancer. Within the human body, uranium undergoes metabolic processes, including the conversion of tetravalent uranium to hexavalent uranium, resulting in the formation of uranyl ions. Moreover, uranium forms complexes with proteins, plasma, citrate, and bicarbonates, indicating the intricate interaction between uranium and the human body. These interactions may contribute to the toxic effects observed in women with breast cancer [30–32].

Table 3 highlights the comparison of the research results obtained in this work with those from previous studies conducted in Iraq–Babylon and different countries. According to the results, the mean uranium concentrations identified in the present research and in previous studies conducted in Iraq–Babylon are greater than those reported in different countries. Military actions in Iraq, especially depleted uranium munitions, may have contributed to this elevated element level.

Conclusion

Noticeably higher concentrations of uranium have been detected in the blood samples of female patients, raising concerns about associated health risks. This discovery highlights the importance of understanding the causes, effects, and impacts of elevated uranium levels. Further research is essential to comprehensively examine the potential toxicity of uranium, increased risk of cancer, and reproductive effects. By identifying the underlying factors and exploring gender-specific impacts, proactive measures can be developed to minimize uranium exposure and safeguard public health.

Acknowledgments. First and foremost, we would like to express our gratitude to the doctors and nurses at the Imam Sadiq Hospital and the Babylon Cancer Center in Babylon province. In addition, we would like to use this opportunity to convey our appreciation to the individuals who donated blood to make this project a success.

Funding. The authors did not get any financial assistance or other support from any organization for the work that was submitted.

Conflict of interest. We declare that the authors have no competing interests.

Ethical approval. The principles mentioned in the Declaration of Helsinki were adhered to during this inquiry. The Ethics Committee of the Babil Health Directorate gave its go-ahead for the project.

Informed consent. A Local Ethics Committee looked through the research protocol, as well as the details on the subjects and the consent form, and gave their stamp of approval (Babil Health Directorate).

Author contribution. Each author made a significant contribution to the study's idea and design. Haider O. Essa, Khalid H. H. Al-Attiyah, and Anees A. Al-Hamzawi were the ones in charge of the material preparation, data collection, and analysis, respectively. The first version of the article was written by Haider O. Essa, and all of the writers provided feedback on earlier drafts of the article. Each contributor was responsible for reading and approving the final draft.

ORCID

K. H. H. Al-Attiyah http://orcid.org/0009-0001-3063-8596
H. O. Essa http://orcid.org/0000-0002-3547-8712
A. A. Al-Hamzawi http://orcid.org/0000-0002-0910-0505

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