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Determining Uranium Concentration in Soft Tissues of Women with Breast and Uterus Cancer and Healthy in Babylon Governorate, Iraq

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Abstract

A critical public health concern in Iraq is the high prevalence and detrimental effects of breast and uterine cancers among women. These malignancies pose a substantial burden on the national healthcare system. To effectively address this issue, a comprehensive understanding of the factors contributing to these elevated cancer rates is paramount. This study aims to establish baseline levels of uranium within affected tissues and investigate its potential association with the development of these cancers. Sample selection, a vital component of the research design, involved the inclusion of 16 controls and 14 case samples. Nuclear fission track analysis utilizing Columbia Resin-39 (CR-39) solid-state nuclear track detectors was employed to quantify uranium content within the collected tissue samples. Statistical analysis of the results revealed a significant difference in uranium accumulation between healthy and malignant tissue samples. The mean uranium concentration in healthy tissues was 3.080 ± 0.32 µg/kg bodyweight, while malignant tissue samples exhibited a higher concentration of 4.311 ± 0.33 µg/kg body weight. These findings highlight the importance of investigating uranium's role in tissue health, potentially informing the development of diagnostic tools or therapeutic interventions for women's health.

Keywords: Uranium levels; Breast cancer; Uterus cancer; Soft tissue; Environmental pollution

1. Introduction

Environmental radioactive contamination, arising from human activities, poses a significant threat to human and animal health, as well as the environment itself. Sources of such contamination include atomic weapons testing, regulated releases from nuclear facilities, accidental discharges, radioactive waste disposal, and naturally occurring radionuclides (Abu Bakar *et al.* 2019; Duarte *et al.* 2023; Orizaola 2020). In contrast, depleted uranium (DU), a byproduct of nuclear fuel enrichment, offers a unique set of properties that have led to its widespread adoption in various civilian and military applications. These applications capitalize on DU's distinctive chemical and physical characteristics. Notably, DU finds use in diverse areas such as gyroscope and flywheel manufacturing, ship ballasts, cockpit counterweights, medical radiation shielding, and rotor production. Its versatility and cost-effectiveness make DU a valuable resource across multiple industries (Jiang and Aschner 2015; Zhang *et al.* 2020).

This study investigates the detrimental effects of uranium exposure on cellular deoxyribonucleic acid (DNA). Prior research has demonstrated uranium's ability to traverse the placental and blood-brain barriers, raising significant concerns regarding its impact on human health. Exposure to uranium has been linked to alterations in gene expression, thereby disrupting normal cellular processes. Furthermore, uranium exposure can induce DNA damage, potentially leading to mutations and compromising genomic stability. Additionally, uranium exposure has been shown to upregulate reactive oxygen species, consequently causing oxidative stress and cellular damage (Joseph *et al.* 2021; Shankar *et al.* 2021; da Silva *et al.* 2021; Xu *et al.* 2023). An extensive body of literature explores uranium concentrations in human biospecimens, highlighting the importance and depth of research in this field. These studies investigate the presence and potential effects of uranium within human tissues, encompassing diverse aspects such as source, exposure pathways, and health implications. Notably, research has focused on uranium levels in blood samples from various populations, including miners, residents near uranium mines/facilities, and individuals residing in areas with elevated uranium concentrations. This breadth of findings strengthens the existing knowledge base, facilitating a more nuanced understanding of factors influencing human uranium burdens (Abbas *et al.* 2022; Ahmed *et al.* 2022; Al-Hamzawi *et al.* 2014; Hernández-Mendoza *et al.* 2013). Fission track analysis presents a viable technique for quantifying uranium concentrations within soft tissue specimens. This methodology utilizes Columbia Resin-39 (CR-39) detectors, which are instrumental in the analytical process. The protocol involves two distinct stages: sample preparation followed by subsequent analysis. Following analysis, interpretation of the resultant fission track data facilitates the determination of uranium concentration within various tissue samples (Algrifi and Salman 2023; Kohn *et al.* 2019). Limited research has been conducted on the uranium concentration in the soft tissue of cancerous patients. Existing literature lacks comprehensive data and findings on this specific topic. As a result, there are significant gaps in knowledge regarding the presence and effects of uranium. The objective of this study was to determine the uranium levels in the tissue samples of women with breast or uterus cancers. By this analysis, the authors aimed to compare them with the uranium levels found in other Iraqi and global populations. The significance of this research lies in the need to understand the relationship between uranium concentration and the increase in the number of people suffering from cancer. This research also contributes to the existing literature by providing new data on uranium concentration in biological samples, allowing for a better understanding of the overall picture.

2. Methodology

2.1 Study area

Located within the central territory of Iraq, the Babylon Governorate encompasses an area of roughly 5,119 square kilometers and houses a population of approximately 2,065,042 inhabitants. The provincial capital, Hillah, situated across the Euphrates River from the historical metropolis of Babylon, adds to the region's significant historical background. This central Iraqi province is renowned not only for its rich cultural legacy and its appeal to tourists, but also for its economic development, with primary industries concentrated on agriculture and irrigation systems (Chabuk *et al.* 2019).

2.2 Collection and selection of samples

In this study, a total of 30 tissue samples were procured from female patients with breast or uterine cancer diagnoses. These samples were obtained from Imam Sadiq Hospital and Babylon Oncology Center, both situated within Babylon Province. The samples were subsequently categorized into two groups: 16 normal tissues (3 breast and 13 uterine) and 14 abnormal tissues (9 breast and 5 uterine). Figure 1 shows the proportion of normal and abnormal tissues in the present study. The age distribution of women who were examined by this study was as follows: the mean age of the female patients was 45.57 ± 2.3 years old, ranging from 22 to 70 years old. Comparatively, the mean age of healthy women was 45.5 ± 1.86 years old, with the age range being 35-63 years. The study sample included a diverse age, range spanning from young adults to older females. This information provided valuable insights into the age demographics of the female patients.

The selection was based on the availability of patients who met the inclusion criteria. The criteria were a) a confirmed diagnosis of breast or uterus cancer; b) no previous exposure to radiation therapy or chemotherapy, and c) no known history of occupational uranium exposure. The patients were selected randomly from a list of eligible candidates provided by the local healthcare facilities. This approach ensured a diverse representation of patients from varying age groups. The selection process was carried out blindly, with the researchers unaware of any personal information regarding the patients. Each selected participant provided an informed consent before her inclusion in the study population.

2.3 Methods

Fission Track Analysis: This method was utilized to track and assess the uranium concentrations in the participants' breast or uterine tissue samples. Specifically, we used CR-39 detector, a sensitive plastic nuclear track detector provided by Pershore Moulding Ltd (London, UK). This technique allowed us to examine the uranium content in the normal and malignant tissue samples. By analyzing the fission tracks from the detector, the authors determined the presence and concentration of uranium in the tissue samples, which underwent a series of steps to prepare them for analysis. First, the preserved samples were cleaned using distilled water to remove the remaining formalin solutions. Next, the samples were subjected to high temperatures at 180 °C for 2 hours in an electric oven. Subsequently, a manual mill was used to grind each sample into a fine powder. To ensure uniformity in the grain size, the crushed samples were then filtered using a 75 μm fine mesh. The resultant powder, with its consistent distribution of grain sizes, was mixed with 0.5 g of ashed tissue powder and 0.1 g of methylcellulose. This mixture was compressed into a compact pellet, measuring 1 cm × 1.5 mm in size. Finally, the CR-39 detectors were placed on each side of the compressed tissue pellet. The latent damage to the CR-39 detector was caused by the ^{235}U (n, f) reaction. The pellets were positioned in a paraffin wax dish located 5 cm away from the Am-Be neutron source with a thermal flounce of 3.02×10^9 neutron/cm^2. The irradiation process lasted for seven days (Al-Hamzawi *et al.* 2015; Igarashi *et al.* 1985).

The setup for irradiating the samples and detectors to the neutron source can be observed in Figure 2. After irradiation, the detectors were etched in NaOH solution under controlled conditions. The fission track density, which is an important parameter, is then recorded using an optical microscope at 400x magnification as shown in Figure 3. The density of the fission tracks was determined based on the following equation:

$$\text{Track density } (\rho) = \frac{\text{Average of total tracks}}{\text{Area of field view}} \quad (1)$$

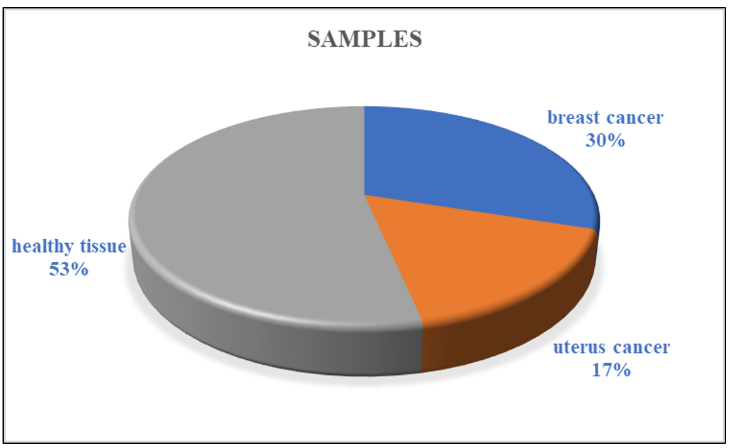
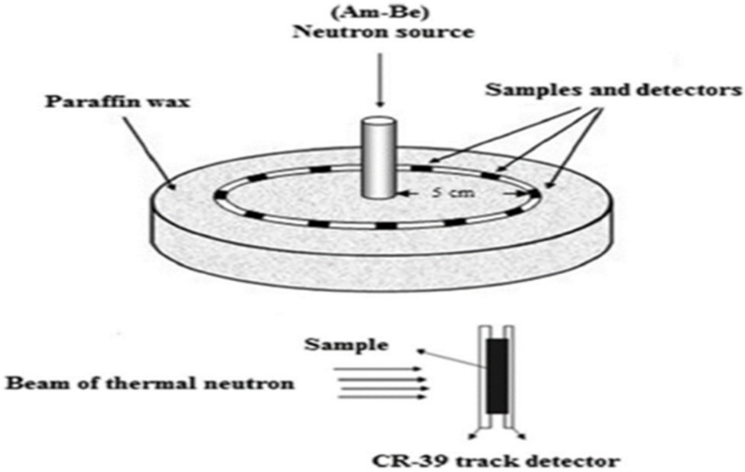
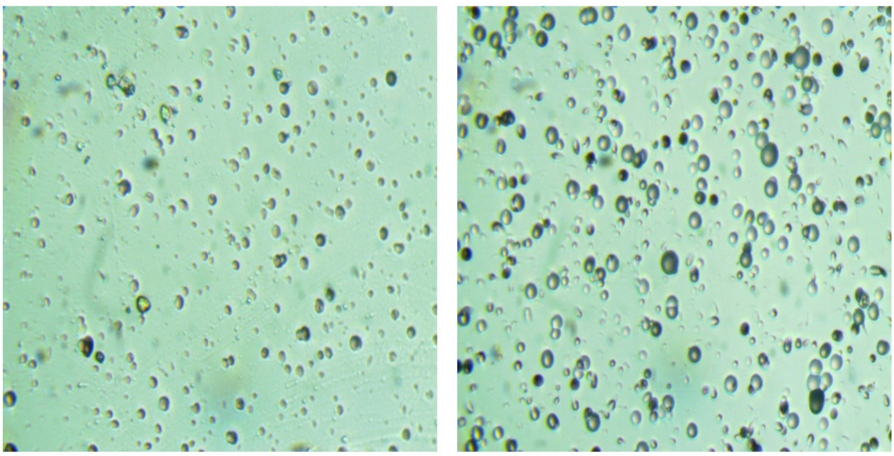


Figure 1. Proportion of normal and abnormal tissues



Source: Al-Hamzawi *et al.* 2015

Figure 2. Samples and detectors are exposed to a neutron flux



(a) (b)

Figure 3. (a) Analysis of nuclear tracks on CR-39 detectors captured under magnification for healthy women, (b) Analysis of nuclear tracks on CR-39 detectors captured under magnification for patients' women.

2.4 Quantification of the uranium content

The detectors were calibrated, using standard samples with known uranium concentrations (provided by the International Atomic Energy Agency (IAEA). To determine the concentration of uranium in soft tissue samples using CR-39 detectors, the track densities recorded around the sample pellets were compared to those around a standard pellet. Based on this comparison, it is possible to calculate the uranium concentration in each of the soft tissue samples. The process involves analyzing the recorded track densities and applying the appropriate mathematical relation (Equation-2). This method provides a reliable and efficient way to determine the uranium concentration in soft tissue samples, allowing for accurate assessment and analysis. The use of CR-39 detectors ensures precise measurements and contributes to the overall reliability and accuracy of the results (Khan and Qureshi 1994; Tommasino 1987).

$$C_x = C_s \frac{\rho_x}{\rho_s} \qquad (2)$$

C_s and C_x in Equation (2) represent the uranium concentrations in standard and unknown samples. The density of fission tracks, denoted as ρ_s and ρ_x, in the standard and unknown samples, respectively (Singh *et al.* 1986).

2.5 Statistical processing and analysis

The SPSS software, version 26.0 was utilized to process and statistically analyze the study data. A significant aspect of the analyses involved conducting an independent *t*-test on data from both study groups. The aim was to determine if there was a notable difference in the probability level (*P*). Based on these statistical treatments, we obtained valuable insights into the significant differences among the data groups under investigation. The results of the analyses provided meaningful information that enabled us to draw valid conclusions and make informed decisions.

3. Results and Discussion

3.1 Results

The study examined the uranium concentrations in the tissue samples from women with breast or uterus cancer. The investigation included 14 abnormal tissues samples which were collected and analyzed for the uranium concentration. Table 1 presents a summary of the study findings. It reveals that the highest uranium concentration in breast cancer tissues was 5.242 ± 0.36 μg/kg bodyweight, found in sample (CT 07, age 65), while the lowest concentration was 3.477 ± 0.34 μg/kg bodyweight, found in sample (CT 08, age 45). In uterine cancer tissue samples, the highest uranium concentration observed was 4.779 ± 0.26 μg/kg bodyweight, found in sample (CT14, age 22), and the lowest concentration was 4.155 ± 0.31 μg/kg bodyweight, found in sample (CT10, age 27).

Uranium concentrations have been assessed in 16 normal tissue samples as seen in Table 2. The recorded uranium concentrations ranged from 1.872 ± 0.26 μg/kg bodyweight to 3.905 ± 0.39 μg/kg bodyweight. The mean uranium concentration in these tissues was found to be 3.080 ± 0.32 μg/kg bodyweight. These results provide valuable insight into the presence and distribution of uranium in soft tissues. As mentioned earlier, no observed relation was found between the age of women with breast and uterus cancer and uranium concentrations.

Table 1. Uranium concentrations in abnormal soft tissues of women with breast and uterus cancer

Sample code	Age	Cancer type	Uranium concentration in (μg/kg bodyweight) ± S.D.
CT 01	55	Breast	4.012 ± 0.272
CT 02	50	Breast	3.905 ± 0.345
CT03	45	Breast	4.190 ± 0.382
CT 04	40	Breast	4.119 ± 0.326
CT 05	30	Breast	4.315 ± 0.351
CT 06	53	Breast	4.582 ± 0.393
CT 07	65	Breast	5.242 ± 0.365
CT 08	45	Breast	3.477 ± 0.346
CT 09	50	Breast	4.583 ± 0.357
CT 10	27	Uterus	4.155 ± 0.311
CT 11	51	Uterus	4.298 ± 0.392
CT 12	35	Uterus	4.226 ± 0.278
CT 13	70	Uterus	4.476 ± 0.376
CT 14	22	Uterus	4.779 ± 0.268
Mean ± Std Error			4.311 ± 0.340

Figure 4 illustrates that the mean uranium concentration in the tissue of cancer patients is higher compared to healthy individuals. This difference was statistically significant ($P < 0.001$) according to an independent sample *t*-test. These findings provide concrete evidence of a potential link between uranium levels and the development of cancer.

Table 2. Uranium concentrations in normal soft tissues of women.

Sample code	Age	Healthy tissue	Uranium concentration in (μg/kg bodyweight) ± S.D.
HT 01	53	Breast	3.085 ± 0.275
HT 02	40	Breast	2.728 ± 0.342
HT03	50	Breast	2.996 ± 0.386
HT 04	37	Uterus	3.620 ± 0.343
HT 05	36	Uterus	2.336 ± 0.321
HT 06	35	Uterus	3.228 ± 0.327
HT 07	40	Uterus	3.656 ± 0.328
HT 08	43	Uterus	3.513 ± 0.359
HT 09	45	Uterus	1.872 ± 0.265
HT 10	47	Uterus	2.764 ± 0.332
HT 11	50	Uterus	2.960 ± 0.248
HT 12	50	Uterus	3.085 ± 0.323
HT 13	43	Uterus	3.905 ± 0.391
HT 14	53	Uterus	1.944 ± 0.325
HT 15	43	Uterus	3.691 ± 0.357
HT 16	63	Uterus	3.905 ± 0.364
Mean ± Std Error			3.080 ± 0.330

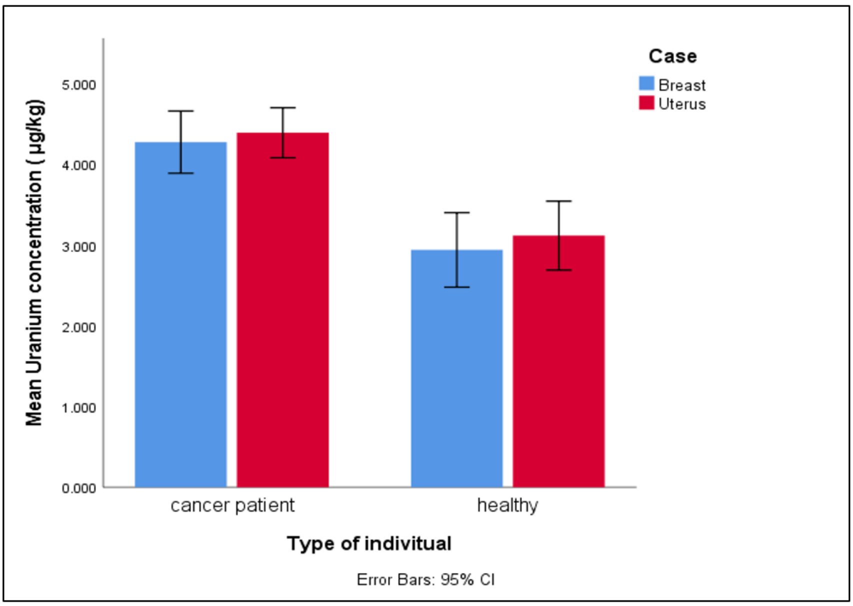


Figure 4. Mean uranium contents in cancer patients and healthy groups

The age groups used in the study provide a clear categorization of participants based on their age range, as shown in Figure 6. The first age group consists of individuals aged 20-29 years, followed by those aged 30-39 years, 40-49 years, 50-59 years, and finally, 60-70 years. By analyzing the soft tissue samples, the study determined the uranium concentrations in each age group. The results indicate an increase in uranium concentrations as participants' age increases. Table 3 presents a comprehensive comparison between the results of this investigation and those documented in prior international studies.

3.2 Discussion

The study data demonstrated that there was a possibility of elevated uranium concentrations in young women compared to those found in the older women. Various factors can influence the concentrations of uranium in women's tissue. One significant factor is the contamination of water and food that women consume daily. If the water and food sources contain high levels of uranium, it can result in higher concentrations of uranium in their tissue. Also, environmental factors play a crucial role in uranium concentrations. When soil or building materials in the environment are contaminated with uranium, women may come into contact with it, leading to its accumulation in their bodies. Inhalation of polluted air in areas with uranium contamination can also contribute to higher concentrations of uranium in women's tissue (Handley-Sidhu *et al.* 2010). The investigation of war remnants, particularly

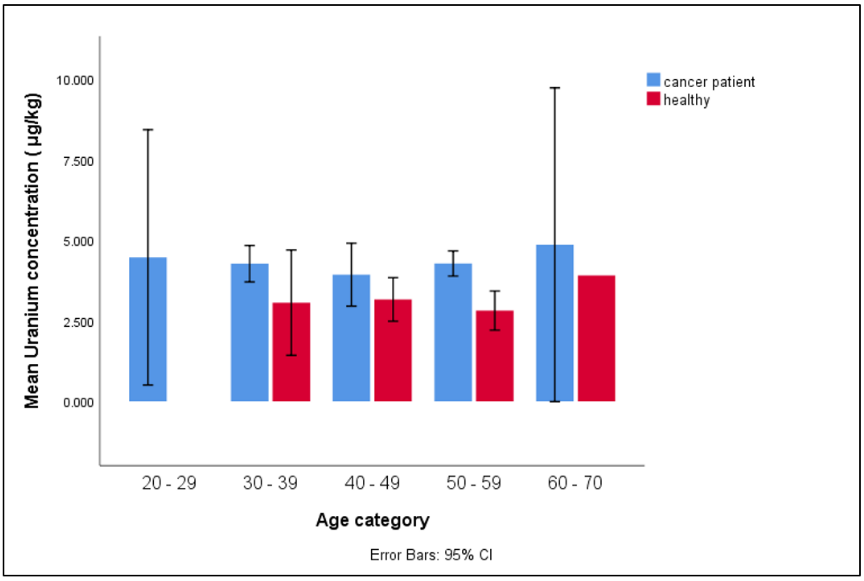


Figure 5. Mean uranium concentrations in tissue samples of the study groups as a function of age

Table 3. Uranium concentration in tissues for different countries.

Country	Mean uranium concentration in (µg/kg bodyweight)		References
Belarus		5.50 ± 0.6	(Malenchenko et al.,1972)
Japan		0.34	(Igarashi et al., 1985)
Russia		5.30 ± 0.9	(Drutman et al., 1985)
USA		0.43 ± 0.26	(Singh et al., 1986)
Iraq	Abnormal Normal	5.242 ± 0.36 3.905 ± 0.39	Present work

depleted uranium ammunition, has revealed its significant contribution to groundwater contamination. Studies conducted, such as the research by Schimmack, et al (Schimmack *et al.* 2005), have shown that leached uranium-238 from corroded ammunition can enter the groundwater, posing potential health hazards to residents dependent on contaminated water sources. The movement of uranium-238 through the soil, driven by rainwater, contributes to the elevated uranium concentration in the groundwater (Schimmack *et al.* 2007). The presence of uranium corrosion products in contaminated environments constitutes a potential ecological hazard, demonstrably impacting plant life. Plants and trees can take up these products through their roots and subsequently transported throughout different plant tissues. This uptake of uranium-corroded products by plants can lead to various consequences, including impacts on plant growth, development, and overall health. The accumulation of these corroded products in plants may pose potential health risks for humans and animals that consume the contaminated plant materials (Handley-Sidhu *et al.* 2010).

Various factors can influence the concentration of uranium in human organs and tissues. They include lifestyle choices, dietary habits, contamination exposure levels, and other potential factors (Grison *et al.* 2022; Stojsavljević *et al.* 2019). The invasion of Iraq in 2003 resulted in the spread of radiation contamination across the affected areas. The destruction of institutions and research centers, such as the Al-Tuwaitha nuclear site, led to the dispersion of various radiation sources. These sources, including radioactive materials and isotopes, became uncontrolled and posed a significant threat to the environment and the population (Harada *et al.* 2022; Zhiltsov *et al.* 2023).

Military interventions in Iraq, including the 1991 Gulf War and the 2003 invasion, involved the deployment of depleted uranium (DU) munitions by US forces. The use of these weapons has raised concerns about potential long-term health consequences for the Iraqi population (Palczewska 2022; Savabieasfahani *et al.* 2020; Surdyk *et al.* 2021). Following inhalation or ingestion, uranium oxides undergo metabolic conversion to uranyl ions. These uranyl ions then form stable complexes with biomolecules such as proteins and ligands, facilitating their systemic dissemination throughout the body. Elimination of uranium primarily occurs via urinary excretion, with minor deposition in skeletal tissues, kidneys, and the liver (Kaur and Mehra 2019; Nordberg and Costa 2021; Ran *et al.* 2020).

The comparison of uranium concentration in women with breast and uterus cancer to those without cancer, as shown in Tables 1 and 2, clearly indicates a significant increase in uranium levels in the cancerous tissues, as shown in Figure 5. This finding suggests a potential link between uranium toxicity and the development of breast and uterus cancer. Within the human body, uranium is metabolized into different compounds, with tetravalent uranium being oxidized to hexavalent uranium and eventually forming uranyl ions. Furthermore, uranium tends to form complexes with proteins, plasma, citrate, and bicarbonates. These processes highlight the intricate interaction between uranium and the human body, potentially contributing to the toxic effects observed in women with breast and uterus cancer (Cooper *et al.* 1982; Rahman *et al.* 2023; Wils *et al.* 2023). The binding and distribution of uranium in the body play a crucial role in its effects on DNA and cancer development. When uranium enters the body, it binds tightly to proteins and other molecules, making it difficult to filter out. Once in the bloodstream, uranium binds to circulating transferrin, a protein involved in iron transport. Additionally, uranium also binds to proteins and phospholipids in the proximal tubule, a part of the kidney responsible for filtering blood and regulating electrolyte balance. These binding mechanisms highlight the potential for uranium to accumulate and persist within the body, increasing the likelihood of adverse effects (Garai and Delangle 2020; Zhou *et al.* 2021).

The mean values of uranium concentrations show a clear upward trend across the age groups, suggesting a positive correlation between age and uranium content. These findings are consistent with the predictions made by the International

Commission on Radiological Protection (ICRP) uranium model, which indicates an expected rise in uranium content with age under a continuous level of uranium intake (Jackson 1996).

4. Conclusion

Higher values of uranium in the tissue samples of female patients compared to healthy females, which raises major concerns about the potential health risks. The study findings warrant the need for further investigation into the causes, implications, and long-term effects of elevated uranium concentrations. Understanding the risks associated with uranium exposure is crucial for developing effective strategies to safeguard the public health. The presence of elevated uranium concentrations in female patients is associated with an increased risk of developing cancer. Uranium is a known carcinogen, and exposure to its high levels can lead to the development of various types of neoplasias, including lung, bone, or kidney cancer. The mechanism by which uranium increases the risk of cancer is believed to involve its ability to damage DNA strand, disrupt cellular processes, and induce genetic mutations.

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