

Histological study for Effecting of Radon Gas on Uterus of Female Rats

Sajad A. Algazali¹, Adhraa Baqir Hassan², and Ali Abid Abojassim^{3*}

¹University of Kufa, Faculty of Pharmacy, Najaf, Iraq

²University of Kufa, Faculty of Science, Department of Biology, Najaf, Iraq

³University of Kufa, Faculty of Science, Department of Physics, Najaf, Iraq

*Corresponding author email: ali.alhameedawi@uokufa.edu.iq

ABSTRACT

The present study was aimed to determine the Histological effect of radon gas (radium-226 source) on the females' rats that include study histological change in uterus. The study was done on 25 animals of aged (8-12) weeks and the weight of (132-208)gm. The first group rats were no exposed to radon gas which it is as control group. While other groups of rats (4 groups) were exposed to radon gas at doses 3063.05 Bq, 4546.86 Bq, 5265.65 Bq, and 613.85 Bq, respectively. The result of body weight shows no significant ($P>0.05$) in body weight of rats in groups exposed by radon gas in compare with control group. Also, the result of muscle thick uterus shows significant decrease ($P<0.05$) in groups exposed by radon gas in compare with control group, while the endometrial thickness is significant increased ($P<0.05$) in groups exposed by radon gas in compare with control group. So, it may be concluded the radon gas at high time exposure cause infertility in female rats.

Keyword: histological parameters, uterus, female rats, and radon gas.

1. INTRODUCTION

The noble gas radon is a naturally occurring radioactive substance. It greatly contributes to the exposure from naturally occurring sources of ionizing radiation. It is also widely used in the treatment of benign inflammatory and non-inflammatory diseases, in many countries. Today, it is widely accepted that the emitting alpha-particles from radon in these springs are responsible for the main dose and thus for several beneficial (Rühle et al., 2017, Gaisberger et al., 2021; Maier et al., 2021). When radon and its short-lived decay products are exhaled, the dose of radiation to tissue of the lung is dominated by the alpha particles emitted by the deposited decay products, which, especially those attached to small aerosols or are in unattached form, cause damage to sensitive lung cells, thereby increasing the probability of cancer developing. Therefore, radon acts mainly as the source of its decay outcome, which actually delivers the dosage to the lungs; however, as a convenient abbreviation, the health effects of radon decay products are often referred to as the health effects of radon (Abojassim *et al.*, 2021). There are several causes of infertility in female and according to the Center for Disease Control, female infertility can be caused by defective ovulation, transport (ovum and sperm), and implantation (zygote). Defective ovulation occurs due to the dysfunction of the hypothalamus and pituitary gland, which may prevent ovulation through the excessive production of prolactin (CDC. Infertility FAQs. 2013). Several known causes for female infertility are premature ovarian insufficiency, polycystic ovarian syndrome (PCOS), and endometriosis (a condition, in which endometrial tissue grows on ovaries). Numerous types of non-ionizing and ionizing radiations and both have recognized causative influences on infertility. Other health effects have been studied but there is no conclusive evidence at present concerning radon-induced health effects other than lung cancer (Abojassim *et al.*, 2021). The aim of this

research is to study the effect of four proid time exposure (4, 8, 12, and 16 day) of radon gas on the histological sections of uteruses for female Albino rats.

2. MATERIALS AND METHODS

2.1. Preparing laboratory animals

The study included (25) female albino Swiss Rat belonging to the (Bulb/C) breed, whose ages ranged between (8-12) weeks, and their average weight was between (132-208) grams. Animals were used after confirming their fertility for the purpose of conducting fertility testing and the animals were obtained from the animal house in the Faculty of Science / University of Kufa. The animals were introduced into the animal house at the Faculty of Science / University of Kufa and placed in plastic cages with their own mesh covers and the floor of the cages were furnished with sawdust and were changed weekly and the animals were placed under appropriate laboratory conditions where the temperature ranged between (21 - 30) C and at a fixed lighting system at the rate of (13) hours of light and (11) hours of darkness. The animals were constantly given water and food in blackberry form and according to need, and were obtained from specialized agricultural shops in Baghdad.

2.2. Irradiation Source and Radon detector

Radium-226 has been used for samples irradiation of the female rates samples. Radium-226 source was produced in 2009 and has activity 6600 Bq. This source consists of a container containing a rod, where the radioactive nucleus releases the radon gases.

One of the more sensitive polymeric plastics, the CR-39 nuclear track detector sheets is the most commonly used in radionuclides measurement that producing α -particles such as radon gases, and has a small size, simplicity, and durability (Hamzah *et al.*, 2022). The CR-39 abbreviation for the Columbia Resin No. 39 detector. Chemical structure for CR-39 is C₁₂ H₁₈ O₇. Each detector was assigned a unique code in order to differentiate them. TASTRAK Analysis System, Ltd., UK: TASTRACK sold the detector CR-39. A CR-39 detector sheet had dimensions of 2.5cm×2.5cm, a thickness of 1mm, and a code for each sheet that suited the TASL image system. The sheet has a density of almost 1.32 grams per cubic meter.

2.3. Experimental Design

The rats kept in animal house for acclimation to laboratory condition for two weeks before they were used for the experiment. Each group was formed 5 Rats and the Rats exposed by radon gas (²²²Rn) at different doses which depend on the variable period time.

- Group 1: Rats exposed by radon gas at concentration 588.51 Bq/m³ of 4 days for 3 groups (radon gas).
- Group2: Rats exposed by radon gas at concentration 714.62 Bq/m³ of 8 days for 3 groups (radon gas).
- Group 3: Rats exposed by radon gas at concentration 756.66 Bq/m³ of 12 day for 3 groups (radon gas).
- Group 4: Rats exposed by radon gas at concentration 840.73 Bq/m³ of 16 day for 3 groups (radon gas).
- Group5: Rats administrated normal saline only as control for 3 groups (radon gas).

The females Rat was explained after being anesthetized with chloroform on the end of each group. The abdominal cavity was opened and the various organs of the female reproductive system that included the Uterus were removed, placed in a solution of formalin (10%), after removing the fatty substances attached to it. It was dried by filter paper and then weighed using a sensitive scale type (Sartorius, Germany) after which the textile and physiological manifestations were studied according to the experiments designed in this study.

3. RESULT

The results of Table (1) indicate that there is no significant difference ($p>0.05$) in radiation groups and control groups (before and after weights) at different weights (186.67±32.32, 186.33±19.0, 208±35.76, 174.33±10.07, 163±13.11, 180.67±42.15, 177±28.05, 198.33±20.21, 178.33±15.89, 172.33±15.01) respectively.

Table (1): Demographic of treated periods and weights (before and after) radiation.

Treated groups	Treated Periods	Weight before (g)	Weight after (g)	Weight gain (g)	Weight gain %
Control	D.W	163±13.11	172.33±15.01	9.33±2.31	5.7±1.08
RAD	4 days	186.67±32.32	180.67±42.15	-6±9.85	-3.97±6.46
	8 days	186.33±19.01	177±28.05	-9.33±26.5	-4.73±14.52
	12 days	208±35.76	198.33±20.21	-9.67±17.1	-3.77±8.37
	16 days	174.33±10.07	178.33±15.89	4±10.15	2.27±5.94
Univariate	LSD	44.250	47.784	28.184	15.418
	p-value	0.296	0.782	0.492	0.501

The results of Table (2) indicate that there is significant decrease ($p>0.05$) in radiation groups and control groups for muscle thick of uterus (226.67±32.15, 172.0±33.05, 115.0±5.0, 76.67±15.28, 625.33±5.51, 625.33±5.51, 625.33±5.51, 625.33±5.51) respectively.

Table (2): Demographic of Muscle thick uterus in control groups, radiation groups and treatment radiation groups

Muscle thick uterus (µm)			
Treated groups Periods	Control	RAD	Multivariate p-value, LSD
4 days	625.33±5.51	226.67±32.15	0.0001 * 28.644
8 days	625.33±5.51	172.0±33.05	
12 days	625.33±5.51	115.0±5.0	
16 days	625.33±5.51	76.67±15.28	
p-value	1.000	0.0001 *	
LSD	10.370	45.963	

The results of Table (3) indicate that there is significant increase ($p>0.05$) in radiation groups and control groups for endometrial thickness (1166.67±51.32, 1183.33±66.58, 1210±26.46, 1370±101.49, 534.0±4.0, 534.0±4.0, 534.0±4.0, 534.0±4.0) respectively.

Table (3): Demographic of Endometrial thickness in control groups, radiation groups and treatment radiation groups

Endometrial thickness (µm)

Treated groups Periods	Control	RAD	Multivariate p-value, LSD
4 days	534.0±4.0	1166.67±51.32	0.0001 * 85.663
8 days	534.0±4.0	1183.33±66.58	
12 days	534.0±4.0	1210±26.46	
16 days	534.0±4.0	1370±101.49	
p-value	1.000	0.021 *	
LSD	7.531	126.538	

4. DISCUSSION

The study reveals no significant difference in radiation groups and control groups (before and after weights) at different weights as present in table (1). Which female rats exposure to radon gas source for different periods, the outcome not found significant change in body weight of female rats. Our study agreement with previous study such as Xuexian Pei et al, (2015) that showed no difference was found for the change of weight in mice that exposed radiation. Figure (1) shows the uterine tissue for control group of female white rats appears: a. The thickness of the two layers of myometrium b. Endometrial. The study reveals decrease significant in RAD groups as presented in table (2). From the obtained results, it could be observed that female rat’s radiation markedly decreases in muscle thick of uterus, which radiation effect on female rats was at all periods but was highest effect in 16 days as showed in Figure (2) which effect of radiation causes uterus cancer due to change muscle thick, myometrial fibrosis and vascular decrease. These results agreement with Rodriguez-Wallberg et al., 2014 which showed 25–50 Gy of radiation would produce infertility in a third of young women and in almost all women over 40 years of age. Exposure to radiation during childhood leads to altered uterine vascularization decreased uterine volume and elasticity, myometrial fibrosis and necrosis, endometrial atrophy and insufficiency. Moreover, ulceration and necrosis last several months, and the damaged tissue may be replaced by dense collagen deposition. The cervix gets quite atrophic and loses its elasticity, especially in older patients (Dehghan *et al.*, 2016, Tehet *et al.*, 2014). The study reveals decrease significant in RAD groups compared with control groups as presented in table (3). From the obtained results, it could be observed that female rat’s radiation markedly increased in endometrial thick of uterus, which radiation effect on female rats was at all periods but was highest effect in 16 days as showed in Figure (2). This agreement with Marci et al., (2018) that show effects of IR go beyond the gonadotoxic affect and also include (in the case of irradiation of the abdominal cavity in childhood) altered uterine vascularization, decreased uterine volume and elasticity, myometrial fibrosis and necrosis, as well as endometrial atrophy and insufficiency. These changes may cause complications during the course of pregnancy, including placental disorder, foetal malposition, preterm delivery, low birth weight, as well as a high risk of uterine rupture. This study agreement with Mahajan, 2015 in adults, an exposure to Total Body Irradiation (TBI) of 12 Gy is associated with significant uterine damage. During childhood, radiation doses of > 25 Gy focused directly to the uterus appear to induce irreversible damage and so far, there is no consensus on the dose of radiation to the uterus, above which a pregnancy would not be sustainable. Irradiation may lead to placental disorders (e.g. placenta accreta or placenta percreta), fetal malposition, preterm labor and premature delivery (Tang and Webber, 2018). Also, this study agreement with Milgrom et al., (2013) in pre-menopausal patients the volume transfer constant (Ktrans) and the extracellular extravascular volume fraction (Ve) were significantly.

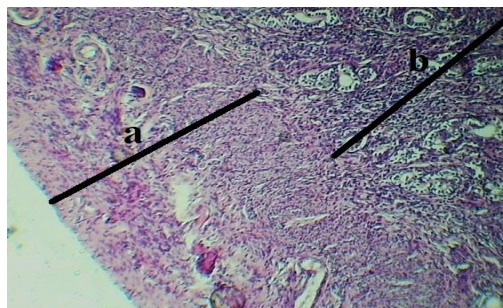


Figure (1): Uterine tissue in the control group of female white rats appears: a. The thickness of the two layers of myometrium b. Endometrial (Hematoxelin - Eosin × 50).

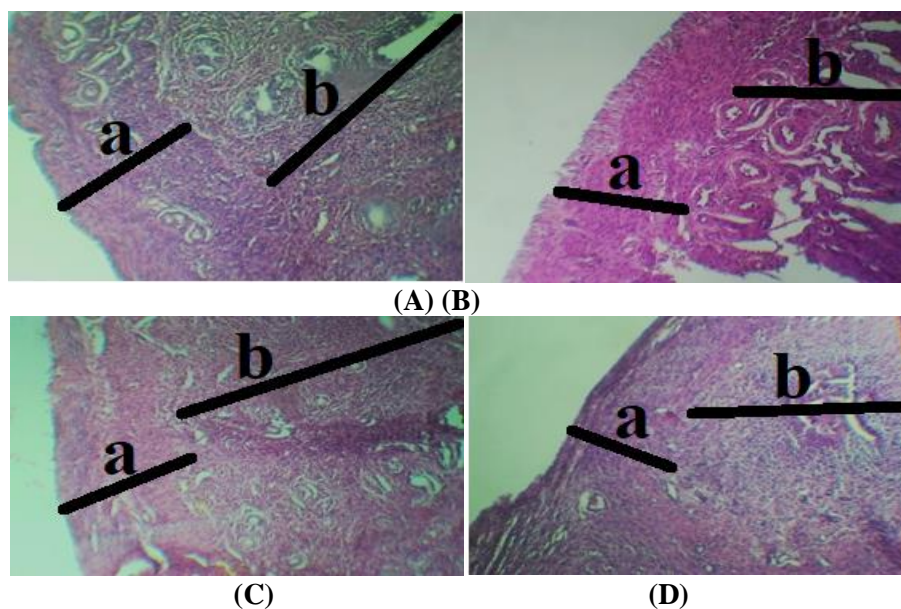


Figure (2): A, B, C & D: Uterine tissue in the treated group with Radon gas a period of irradiation (4, 8, 12 &16) days of female white rats appears: A. An increase thickness of the two layers of myometrium B. An increase endometrial (Hematoxelin - Eosin × 50).

5. CONCLUSION

Radon gas does not cause any effected in the weights of body in female rats, while cause large effect on histological change in uterus such as muscle thick and endometrial thickness. It is found that muscle thick was decreased with increasing of radon concentrations, whilethe endometrial thickness was increased with the period time of radiation. Very high levels of radon gas exposure can affect on histological change in uterus for many years or permanently. So, it is clear from this study the exposure to high levels of radon gas affect fertility and may be causing infertility.

REFERENCES

1. Gaisberger, M.; Fuchs, J.; Riedl, M.; Edtinger, S.; Reischl, R.; Grasmann, G.; Hölzl, B.; Landauer, F.; Dobias, H.; Eckstein, F.; et al.(2021). Endogenous anandamide and self-reported

- pain are significantly reduced after a 2-week multimodal treatment with and without radon therapy in patients with knee osteoarthritis: A pilot study. *Int. J. Biometeorol.*, 65, 1151–1160.
2. Maier, A.; Wiedemann, J.; Rapp, F.; Papenfuß, F.; Rödel, F.; Hehlhans, S.; Gaipl, U.S.; Kraft, G.; Fournier, C.; Frey, B.(2021). Radon Exposure—Therapeutic Effect and Cancer Risk. *Int. J. Mol. Sci.* 2021, 22, 316.
 3. Rühle, P.F.; Wunderlich, R.; Deloch, L.; Fournier, C.; Maier, A.; Klein, G.; Fietkau, R.; Gaipl, U.S.; Frey, B.(2017). Modulation of the peripheral immune system after low-dose radon spa therapy: Detailed longitudinal immune monitoring of patients within the RAD-ON01 study. *Autoimmunity*, 50, 133–140.
 4. Tang M, Webber K.(2018). Fertility and pregnancy in cancer survivors. *Obstet Med.*;11:110–15.
 5. Milgrom SA, Vargas HA, Sala E, Kelvin JF, Hricak H, Goodman KA.(2013). Acute effects of pelvic irradiation on the adult uterus revealed by dynamic contrast-enhanced MRI. *Br J Radiol.*;86:20130334.
 6. Mahajan N.(2015). Fertility preservation in female cancer patients: an overview. *J Hum Reprod Sci.*;8(1):3–13.
 7. Marci R, Mallozzi M, Di Benedetto L, Schimberni M, Mossa S, Soave I, et al.(2018). Radiations and female fertility. *Reprod Biol Endocrinol.*; 16(1): 112.
 8. Xuexian Pei, Qijun Gu, Dongdong Ye, Yang Wang, Xu Zou, Lianping He, Yuelong Jin and Yingshui Yao (2015). Effect of computer radiation on weight and oxidant-antioxidant status of mice, Original / Investigación animal, *Nutr Hosp.*31(3):1183-1186.
 9. Rodriguez-Wallberg KA, Oktay K.(2014). Fertility preservation during cancer treatment: clinical guidelines. *Cancer Manag Res.*;6:105–17.
 10. Teh WT, Stern C, Chander S, Hickey M.(2014). The impact of uterine radiation on subsequent fertility and pregnancy outcomes. *Biomed Res Int.*;2014:482968.
 11. Dehghan T, Mozdarani H, Khoradmehr A, Kalantar SM.(2016) Effects of gamma radiation on fetal development in mice. *Int J Reprod Biomed (Yazd)*.14(4):247–54.
 12. Hamzah, Z. S., Hashim, A. K., & Abojassim, A. A. (2022). Assessment of Annual Effective Dose and Excess Lifetime Cancer Risk in Grain Samples Collected from Kerbala Governorate, Iraq. *Iranian Journal of Science and Technology, Transactions A: Science*, 1-10.
 13. Abojassim, A. A. (2021). Radiological risk assessment of radon gas in bricks samples in Iraq. *Journal of Nuclear Engineering and Radiation Science*, 7(3).
 14. CDC. Infertility FAQs. 2013. Available online: [https://www.jognn.org/article/S0884-2175\(15\)31517-3/fulltext](https://www.jognn.org/article/S0884-2175(15)31517-3/fulltext) (accessed on 30 December 2020).