

Evaluation of Haematological Variation in Wistar Rat (*Rattus Norvegicus*) Associated with Radon Ingestion from Groundwater

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Abstract

Numerous studies have been conducted on radon inhalation and its effects on oxidative stress parameters, but research involving radon ingestion and its haematological effects has not been reported. A total of one hundred and ten (110) albino rats weighing between 150 – 200 g each, consisting of fifty (50) non-pregnant females and 50 males, were used in this study. The rats were randomly assigned to four groups according to the study design. Exposed group rats were fed with newly collected radon-laden water (RLW) with an average radon concentration of 44333 Bq/m³ every day, while rats in Control Groups were given stream water, called control water (CW), with an average radon concentration of 1030 Bq/m³, throughout the experiment. After eight (8), sixteen (16) and twenty-four (24) weeks of radon exposure, respectively, six (6) rats were euthanized from Groups 1, 2, 3 and 4, after which the cervical dislocation was done to collect the blood samples. Blood samples were collected in an Ethylenediamine tetraacetic acid (EDTA) bottle for a haematological assay. The collected samples were taken to the laboratory to determine the values of WBC, ABS Platelet Count, Haemoglobin, and PCV using an automated haematology analyzer. Results with a t-test analysis at $\alpha = 0.05$ show significant differences in weeks 8 and 16, but more significantly in week 24. Therefore, the study highlights radon ingestion's potential to disrupt haematological parameters, emphasising the importance of mitigating radon exposure. Thus, it is crucial to supply drinking water that meets safety standards for radon levels to protect public health.

Keywords: Radon; Haematology; Blood; Hand-pumped water; Portable water.

I. INTRODUCTION

Availability of portable water in Nigeria and in the world at large is of great concern in this century. In search of

potable water, people try different available sources to get access to water [1] regardless of any form of possible contamination or threat posed to their health. Sources of radiation are present everywhere around us in soil, water and

air; some are natural and some are artificial [2]. Some radiations are non-ionizing, while others, like radon, are highly ionizing. Radon is present in all three states of matter, it has many isotopes of which Radon 222, the most abundant of them is a product of the disintegration of primordial radionuclide U-238 and Th-234 in the Uranium and Thorium decay series [1].

Predominantly, Radon (radon-222) is a water-soluble, odourless, colourless, tasteless and geogenic gas produced by the radioactive decay of uranium-bearing rock formations. It is the second cause of lung cancer according to World Health Organization (WHO) [3]. Research has shown that the groundwater used for consumption every day consists of high levels of radon-222 which is carcinogenic [4].

Radon exposure has been reported to bear carcinogenic risk, not only to miners but also from indoor exposure and lately from groundwater [5]. It has a half-life of 3.8 days, and once released in the air, it decays into short-lived radioactive solid particles (radon progeny), including polonium, lead, and bismuth.

Breathing radon in the indoor air of homes is the primary public health risk from radon, which is the second leading cause of lung cancer in the general population in the world. Radon in drinking water causes about 89% of lung cancers resulting from breathing radon released from water, and 11% of stomach cancers resulting from radon in drinking water [6]. When radon an alpha particle emitter, which belongs to the group of ionizing radiation is swallowed in drinking water, it

proceeds through the gut to the stomach and intestines, then the bloodstream, and lastly, the lungs, where most of it is exhaled, while the remnants are decayed. Blood is the fundamental of all living organisms. It is a liquid which flows in all living cells [7]. The study of blood properties, its functions and diseases related to it is called haematology. In this study, haematological properties such as packed cell volume (PCV), haemoglobin concentration, total white blood cell counts (TWBC), and ABS Platelet are observed in relation to their exposure to radon.

II. MATERIALS AND METHODS

A. Geology of the Study Area

The research work was carried out in Ogbomoso, Oyo State, Nigeria, which lies between latitude $8^{\circ} 2' - 8^{\circ} 11' N$ and longitude $4^{\circ} 7' - 4^{\circ} 22' E$ [8]. It is geologically located within the southwestern Nigeria basement complex made up of ancient gneiss-migmatite series and meta-sedimentary series. The rock contains biotite, hornblende, quartz, plagioclase, microcline and rarely pyroxene. Those gneisses with a high content of mafic minerals may yield clayey soils, while the coarse-grained, more granitic components may account for soils with varying textures with less clay [9]. The major geological settings in Nigeria divided in equal proportion include the sedimentary Basins [10], [11] and the crystalline Basement Complex [12]. Fig. 1 depicts the map of Nigeria showing the Ogbomoso Precambrian basement complex.

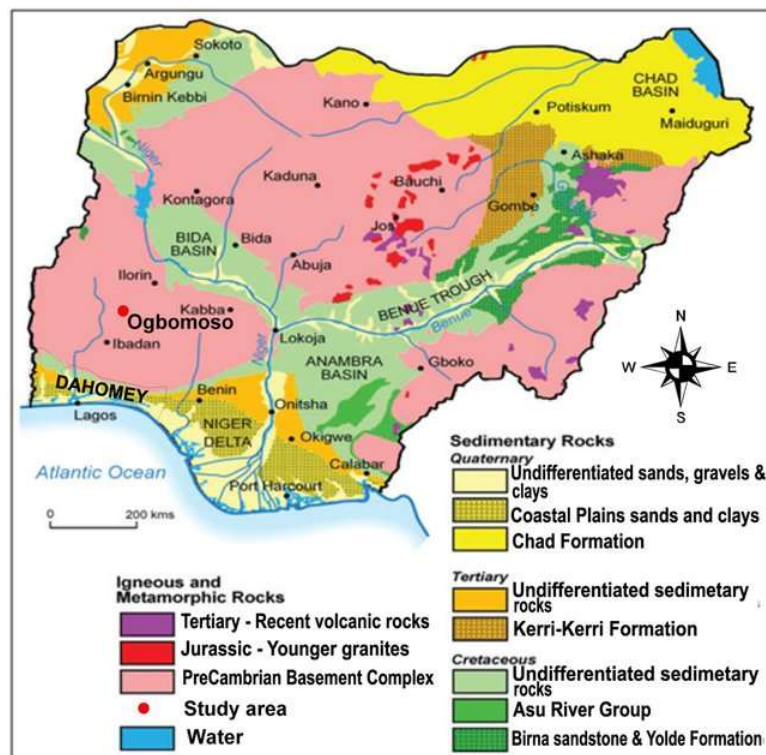


Fig. 1. Map of Nigeria showing Ogbomoso [13]

B. Collection of water samples

Water samples from six selected hand-pumped boreholes and a stream from the Ogbomoso metropolis were analyzed using a RAD 7 radon detector [14]. With daily measurements to ensure accuracy of results, borehole water with the highest radon concentration (44.333 Bq/L) was designated as Radon Laden Water (RLW), while stream water with the lowest

concentration (10.30 Bq/L) served as Control Water (CW). The samples of Stream water were collected around Water Corporation, Ogbomoso, labelled and measured accordingly. Conscious effort was taken to ensure that water was collected directly from the pump into the container, which was securely tightened to prevent the escape of radon gas [15]. Fig. 2 shows the arrangement for measuring radon in water with RAD 7.

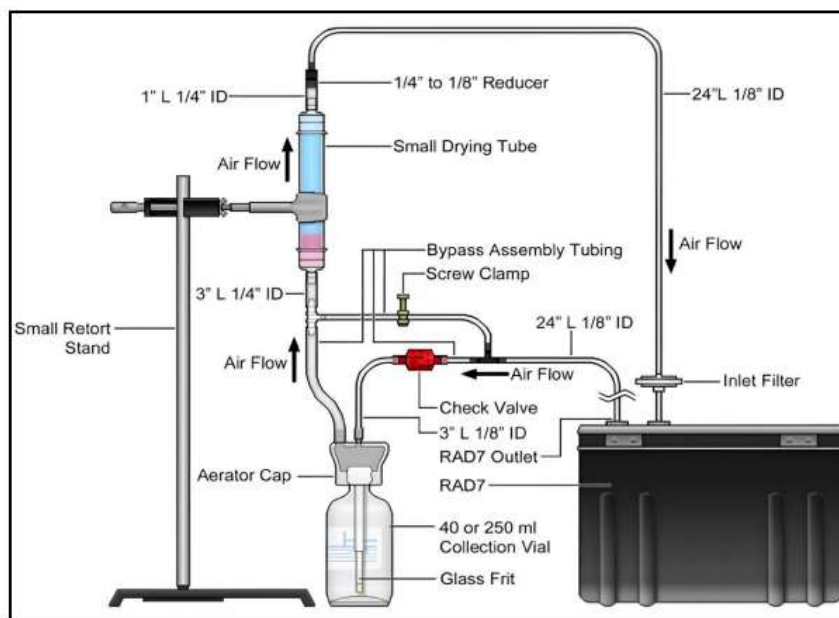


Fig. 2. Measurement of Radon concentration in Water with RAD [16].

C. Experimental Animal and Design

The choice of animal and design varies [17] depending on the research in view. In this study, a total of one hundred and ten (110) albino rats weighing between 150 – 200 g, consisting of 50 non-pregnant females and 50 males were used. The rats were randomly assigned to four groups according to the study design. Group 1 (25 female rats) and Group 2 (25 male rats) were labelled as Female Control Group (CF) and Male Control Group (CM), respectively. Group 3 (25 female rats) and Group 4 (25 male rats) were labelled as Female Exposed Group (EF) and Male Exposed Group (EM), respectively. Animals in each group were kept in different cages, designed according to the standard cage size recommended by the Animal Research Review Panel (ARRP) Guideline 20: Guidelines for the Housing of Rats in Scientific Institutions. To adapt to the new environment, all rats were allowed to acclimatize for two weeks before entering the trial. Standard animal feed was given to all animals, and all moral principles on the use and treatment of animals were considered [18].

D. Animal exposure

Rats in Groups 1 and 2 (Control Groups) were supplied with stream water called control water (CW), while rats in Groups 3 and 4 (Exposed Groups) were supplied with radon-laden

water (RLW) from a hand pump borehole (water sample RLW) throughout the experiment. Exposed group rats were fed with a freshly collected water sample, RLW, every day. Therefore, all exposed rats ingested radon from the water sample RLW throughout the experiment, whereas control group rats were fed with stream water from control water (CW) throughout the experiment. The animals drank the water directly from water troughs, varying in values based on the concentration of radon ingested.

E. Blood sample collection

After eight (8) weeks of radon exposure, six (6) rats were euthanized from Groups 1, 2, 3 and 4, and their blood samples were taken by cardiac puncture. Blood samples were collected into an anticoagulant (EDTA) bottle for haematological assay [19]. Haematological parameters investigated in this work are: Total White Cell Count (TWCC), Haemoglobin, ABS Platelet Count and Packed Cell Volume (PCV). Similar procedures in Groups 1, 2, 3 and 4 were repeated after sixteen (16) weeks and twenty-four (24) weeks of radon exposure.

III. RESULTS AND DISCUSSION

This research has been carried out for health risk management related to radon ingested in water [20]. Tables I

- VI are the results of radon concentration for stream water and hand-pumped borehole water for the twenty-four weeks of radon exposure. Tables I - III represent the data obtained from stream water, while Tables IV-VI are the results of radon concentration from a hand-pumped borehole with a very high

mean concentration value of 44.333 Bq/L, far higher than the recommended safe value of 11.1 Bq/L [21]. Tables VII – IX depict the results of haematological indices of the Wistar rats, for control and exposed animals, which showed variation in values based on the concentration of radon ingested.

Table I. Radon Concentration– Stream Water (Week 1 to 8)

Day	Radon (Bq/L) Mean	Day	Radon (Bq/L) Mean	Day	Radon (Bq/L) Mean
1	10.570±2.13	20	10.326±0.17	39	10.082±0.16
2	10.300±1.15	21	9.576±0.23	40	10.332±0.15
3	10.300±1.80	22	9.826±0.21	41	10.583±0.16
4	9.650±2.13	23	10.075±0.21	42	9.833±0.34
5	9.850 ±0.82	24	10.078±0.23	43	10.083±0.24
6	9.850 ±0.24	25	10.328±0.21	44	10.334±0.44
7	9.950 ±0.12	26	9.578±0.16	45	10.584±0.18
8	10.250±3.82	27	9.820±0.14	46	9.834±0.20
9	9.850 ±2.82	28	10.329±0.25	47	10.085±0.23
10	10.470±0.85	29	10.580±0.15	48	10.335±0.24
11	10.440 ± 1.42	30	9.830±0.34	49	9.585±0.12
12	10.323±0.16	31	10.080±0.25	50	9.835±0.23
13	10.574±0.21	32	10.330±0.15	51	10.086±0.14
14	9.820±0.16	33	10.580±0.25	52	10.336±0.24
15	10.072±0.36	34	9.831±0.17	53	10.333±0.13
16	10.325±0.27	35	10.081±0.15	54	10.071±0.16
17	10.575±0.18	36	10.331±0.15	55	10.073±0.18
18	9.825±0.26	37	10.582±0.14	56	10.322±0.21
19	10.025±0.18	38	10.832±0.24		

Table II. Radon Concentration – Stream Water (Week 9 to 16)

Day	Radon (Bq/L) Mean	Day	Radon (Bq/L) Mean	Day	Radon (Bq/L) Mean
1	10.500±2.13	20	10.326±0.17	39	10.082±0.14
2	10.500±2.15	21	10.576±0.16	40	10.332±0.18
3	10.200±1.80	22	10.826±0.16	41	9.583±0.16
4	10.500±2.13	23	10.075±0.18	42	9.833±0.14
5	10.500±0.12	24	10.078±0.15	43	10.083±0.34
6	10.350 ±0.82	25	10.328±0.25	44	10.334±0.44
7	10.250 ±0.45	26	10.578±0.16	45	10.584±0.41
8	10.250 ±3.43	27	10.320±0.18	46	9.834±0.31
9	10.150 ±2.44	28	10.329±0.45	47	10.085±0.14
10	10.470 ±0.85	29	10.580±0.15	48	10.335±0.24
11	10.440 ± 1.42	30	10.330±0.22	49	9.585±0.16
12	10.323±0.16	31	10.080±0.18	50	9.835±0.17
13	10.457±0.16	32	10.330±0.15	51	10.086±0.18
14	10.320±0.26	33	10.580±0.17	52	10.336±0.21
15	10.072±1.15	34	9.831±0.16	53	10.333±0.20
16	10.325±0.17	35	10.081±0.25	54	10.071±0.16
17	10.475±0.18	36	10.331±0.55	55	10.073±0.22
18	10.425±0.16	37	10.582±0.16	56	10.322±0.24
19	10.025±0.19	38	10.832±0.24		

Table III. Radon Concentration – Stream Water (Week 17 to 24)

Day	Radon (Bq/L) Mean	Day	Radon (Bq/L) Mean	Day	Radon (Bq/L) Mean
1	10.570±2.13	20	10.326±0.17	39	10.082±0.24
2	10.500±1.15	21	10.576±0.26	40	10.332±0.24
3	10.900±1.80	22	9.826±0.21	41	10.583±0.34
4	10.650±2.13	23	10.075±0.16	42	9.833±0.14
5	9.850 ±0.82	24	10.078±0.15	43	10.083±0.24
6	9.850 ±0.92	25	10.328±0.36	44	10.334±0.16
7	9.950±0.72	26	10.578±0.17	45	9.584±0.15
8	10.250±3.82	27	9.820±0.16	46	9.834±0.18
9	9.850 ±2.82	28	10.329±0.15	47	10.085±0.14
10	10.470±0.85	29	9.580±0.23	48	10.335±0.16
11	10.440 ± 1.42	30	9.830±0.15	49	10.585±1.21
12	10.323±0.16	31	10.080±0.17	50	9.835±2.11
13	10.574±0.24	32	10.330±0.18	51	10.086±1.15
14	9.820±0.22	33	9.580±0.17	52	10.336±2.14
15	10.072±0.16	34	9.831±2.15	53	10.333±2.18
16	10.325±0.17	35	10.081±0.15	54	10.071±0.16
17	10.575±0.18	36	10.331±4.10	55	10.073±1.16
18	9.825±0.25	37	10.582±0.14	56	10.322±2.18
19	10.025±0.16	38	9.832±0.31		

Table IV. Radon Concentration Hand Pumped – Borehole Water (Week 1 to 8)

Day	Radon (Bq/m ³) Mean	Day	Radon (Bq/m ³) Mean	Day	Radon (Bq/m ³) Mean
1	43.570±2.13	20	45.326±1.17	39	43.082±1.19
2	44.500±1.15	21	45.576±1.16	40	45.332±2.13
3	45.900±1.80	22	45.826±2.11	41	46.573±2.14
4	45.650±2.13	23	45.075±2.12	42	44.823±2.18
5	44.854 ±0.82	24	44.078±1.15	43	43.033±2.16
6	43.850 ±1.82	25	44.328±1.16	44	44.334±2.15
7	44.941±2.12	26	47.538±1.14	45	44.574±2.14
8	44.211±2.32	27	41.820±1.15	46	44.634±3.14
9	43.850 ±2.42	28	46.329±0.15	47	43.085±2.24
10	44.422±1.25	29	45.580±2.18	48	45.335±1.12
11	42.431±1.42	30	44.830±1.15	49	45.575±1.21
12	43.321±1.16	31	45.080±1.17	50	44.825±2.16
13	43.574±2.16	32	45.330±2.11	51	44.086±0.11
14	43.820±2.21	33	43.580±1.15	52	45.326±1.16
15	44.072±1.26	34	44.831±2.15	53	44.332±2.24
16	44.325±2.17	35	43.081±1.18	54	45.071±1.26
17	44.575±1.18	36	44.331±1.12	55	45.073±2.15
18	44.825±2.16	37	45.582±1.14	56	44.322±1.18
19	45.025±2.14	38	44.832±1.16		

Table V. Radon Concentration Hand Pumped – Borehole Water (Week 9 to 16)

Day	Radon (Bq/m ³) Mean	Day	Radon (Bq/m ³) Mean	Day	Radon (Bq/m ³) Mean
1	40.570±2.13	20	45.326±2.17	39	44.082±2.14
2	41.500±1.15	21	45.576±1.18	40	43.332±1.16
3	40.900±1.80	22	45.826±1.21	41	43.583±2.18
4	43.650±2.13	23	45.075±0.16	42	43.833±1.16
5	44.85±1.42	24	46.078±0.15	43	42.083±2.14
6	43.85±2.32	25	47.328±1.16	44	42.334±1.19
7	44.95±1.22	26	47.578±1.16	45	43.584±1.15
8	44.25±3.82	27	43.820±2.16	46	44.834±2.18
9	43.85±2.82	28	44.329±2.15	47	43.085±2.16
10	45.47±0.85	29	44.570±1.15	48	44.335±1.14
11	44.44±1.42	30	44.830±1.37	49	43.585±0.12
12	43.323±2.16	31	45.080±2.53	50	44.835±0.14
13	43.574±1.18	32	44.330±2.15	51	44.086±1.15
14	45.820±2.16	33	44.580±2.31	52	44.336±2.17
15	45.072±0.76	34	45.831±2.45	53	44.333±0.78
16	44.325±1.17	35	44.081±1.23	54	43.071±0.76
17	44.575±2.18	36	43.331±2.22	55	44.073±1.65
18	44.825±1.16	37	44.582±1.34	56	46.312±0.16
19	45.025±2.16	38	44.832±1.24		

Table VI. Radon Concentration Hand Pumped – Borehole Water (Week 17 to 24)

Day	Radon (Bq/m ³) Mean	Day	Radon (Bq/m ³) Mean	Day	Radon (Bq/m ³) Mean
1	43.570±2.13	20	45.326±2.17	39	44.082±2.23
2	43.500±1.15	21	45.574±2.26	40	44.332±2.17
3	43.900±1.80	22	45.816±2.34	41	44.583±2.14
4	44.650±2.13	23	45.075±1.15	42	44.843±2.24
5	44.850±1.82	24	46.078±1.18	43	44.083±1.34
6	43.851±2.12	25	47.318±2.21	44	44.334±1.83
7	44.940±1.32	26	47.548±2.26	45	45.574±1.68
8	44.252±3.82	27	43.820±1.17	46	45.734±2.64
9	43.850±2.22	28	44.329±1.15	47	43.085±0.14
10	45.461±1.35	29	43.580±1.13	48	44.335±2.15
11	44.442±1.42	30	44.830±2.16	49	45.385±2.13
12	43.323±1.16	31	44.080±1.15	50	45.435±0.17
13	44.574±2.15	32	45.330±2.18	51	44.036±2.19
14	43.820±1.19	33	45.580±2.17	52	44.356±1.18
15	45.072±1.18	34	45.831±2.15	53	44.323±2.64
16	44.325±1.17	35	44.081±2.11	54	44.181±1.16
17	44.525±2.18	36	44.331±1.16	55	44.173±1.18
18	44.825±2.16	37	45.582±1.14	56	45.122±2.19
19	45.025±1.16	38	44.832±1.13		

Table VII. Haematological Parameters (Week 8)

S/N	Group	PCV (%)	HGB	TWBC	ABS Platelet Count
1	CM	46	12.6±3.4	4500	263000
2	CF	44	10.5±2.3	3500	258000
3	EM	37	12.5±0.8	4400	324000
4	EF	41	12.0±0.5	2500	251000

Table VIII. Haematological Parameters (Week 16)

S/N	Group	PCV (%)	HGB	TWBC	ABS Platelet Count
1	CM	44	13.0±1.20	3600	327000
2	CF	42	11.8±0.42	2700	283000
3	EM	41	12.1±0.06	3550	305000
4	EF	37	12.4±4.74	5050	340000

Table IX. Haematological Parameters (Week 24)

S/N	Group	PCV (%)	HGB	TWBC	ABS Platelet Count
1	CM	46	14.2±2.40	2850	211500
2	CF	42	11.0±1.50	2150	255000
3	EM	41	12.1±0.07	3550	305000
4	EF	38	13.3±1.63	6400	419000

In the eighth (8), sixteenth (16) and twenty-fourth (24) weeks of ingestion, a low PCV value was observed for the exposed rats compared to the control ones, for males and females respectively, which is an indication that the blood of the latter has been infected [22]. A low PCV implies that the patient has a low number of red blood cells and is suffering from anaemia. Also, a reduction in PCV value due to the high concentration of radon ingested is a pointer to the future growth of cancerous cells [23].

Whereas, for Haemoglobin, lower values were observed in exposed males; a condition that can lead to different kinds of anaemia and cancer, which normally results in fatigue and weakness [24]. Higher values were observed for exposed females across the twenty-four (24) weeks of radon ingestion.

A high value of haemoglobin is always associated with fatigue, dizziness, weight loss, swelling and headaches; if prolonged can cause pulmonary fibrosis, polycythemia vera, liver cancer, kidney cancer, or emphysema [25].

As observed from Table VII for Packed Cell Volume (PCV) of controlled and exposed rats, the statistical analysis using t-test with $\alpha = 0.05$, there is no significant difference between the two groups for males and females, respectively. However, there is a reduction in the values of PCV. Male PCV was reduced by a value of 9% while female PCV was reduced by a value of 3%.

There was a decrease in the value of White Blood Corpuscle for male and female exposed groups, as observed from Table VII - IX, a condition that may lead to leukaemia [26]. This was widely observed in weeks 8, 16 and 24, respectively.

Statistical analysis also shows using a two-tailed t-test at $\alpha = 0.05$ confirm a p-value of 0.004 at week 8 and 0.002 at week 24 for both males and females, respectively.

The ABS platelet count increased in value for both males and females; however, the p-values for males and females, which are 0.44 and 0.23, respectively, increased with the number of ABS platelet counts in the blood of exposed rats.

An increase in blood platelets can result in a condition known as thrombocytosis [27]. If left untreated, this condition can lead to a stroke.

IV. CONCLUSION

This study focused on a region where water quality awareness is relatively low, and many individuals are indifferent to the type of water they consume or use daily. The results of this study indicate the danger embedded in drinking radon-laden water from hand-pumped boreholes. The ingestion of this water above the recommended contaminant level from groundwater obtained from hand-pumped boreholes has a great effect on the blood properties of Wistar rats. The presence of radon in the blood contaminated the blood parameters and the effect depends on the period of exposure and the concentration consumed. To guarantee the quality and safety of groundwater, a thorough geological survey of the bedrock should be conducted before drilling boreholes, and safe drinking water should be made available for public consumption.

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