

## MODIFICATION OF HUMAN BLOOD IRRADIATION TECHNIQUE WITH THE RADON GAS: *INVITRO* STUDY

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### ABSTRACT

*The aim of this study is to design radon irradiation technique in the field of hematology for an invitro study. In addition, deposit of alpha particles into the human blood surface and its effects on the thrombocytopenia estimated using nuclear track detectors (NTDs). In this technique, amount of radon gas ( $2210 \pm 5.1 \text{ Bq/m}^3$ ) collected in a tight PVC container with the appropriate engineering dimension using two sources of radium ( $5 \mu\text{Ci}$ ). Blood samples (10 male and 10 female) and CR-39NTDs (40 pieces) are exposed to radon gas at various exposure time. Complete blood test and the computer scanning for each piece of CR-39NTDs before and after exposure has done. The results show that the present technique has a good efficiency ( $\approx 96\%$ ) to the invitro exposure of human blood. When the radon gas moved on the surface of blood sample, alpha tracks registered into CR-39NTDs. Thus, this technique improved that the comparative method to evaluate alpha particle density into exposure blood samples is an effective way; this depended on the geometry of design and the sensitivity of CR-39NTDs to track registration. Radon detector version 7 (RAD7) used to make a certain suitability of CR-39NTDs. Amount of radon concentration losses during the exposure process, in the present work it was variable from 0.41% to 1.4%. Radon concentration effected on the thrombocytopenia; this depended on time of exposure and alpha energy loss into the blood and CR-39 through the atomic displacements. At the time of exposure of 10 minutes, rate of absorption dose was  $2.255 \pm 0.11 \mu\text{Sv}$  (39%), and the platelet (PLT) cont reduced rapidly (high effected on reduce PLT, this makes thrombocytopenia.*

### ABSTRAK

*Tujuan kajian ini adalah untuk merancang teknik iradiasi radon dalam bidang hematologi untuk kajian invitro. Selain itu, deposit zarah alfa ke permukaan darah manusia dan pengaruhnya terhadap thrombocytopenia estimasi menggunakan pengesan lagu nuklear (NTDs). Dengan teknik ini, jumlah gas radon ( $2.210 \pm 5.1 \text{ Bq/m}^3$ ) dihimpunkan dalam bekas PVC rapat dengan dimensi teknik yang tepat dengan menggunakan dua sumber radium ( $5 \mu\text{Ci}$ ). Darah sampel (10 lelaki dan 10 wanita) dan CR-39NTDs (40 buah) yang terkena gas radon pada berbagai masa bukaan. ujian darah lengkap dan pengimbasan komputer untuk setiap potongan CR-39NTDs sebelum dan selepas kenalan telah dilakukan. Keputusan kajian*

*menunjukkan bahawa teknik ini mempunyai kecekapan yang baik ( $\approx 96\%$ ) ke paparan invitro darah manusia. Bila gas radon bergerak pada permukaan sampel darah, alfa trek berdaftar ke CR-39NTDs. Dengan demikian, teknik ini diperbaiki bahawa kaedah perbandingan untuk menilai kepadatan zarah alpha menjadi sampel darah pendedahan adalah cara yang berkesan, hal ini bergantung pada geometri dari desain dan sensitiviti CR-39NTDs untuk mengesan pendaftaran. Radon versi pengesan 7 (RAD7) digunakan untuk membuat kesesuaian tertentu CR-39NTDs. Jumlah kerugian kepekatan radon semasa proses paparan, dalam karya ini itu variable dari 0,41% menjadi 1,4%. kepekatan Radon berpengaruh pada thrombocytopenia, hal ini bergantung pada masa paparan dan kehilangan tenaga alfa ke dalam darah dan CR-39 melalui perpindahan atom. Pada saat pendedahan dari 10 minit, tahap penyerapan dosis  $2,255 \pm 0.11\mu\text{Sv}$  (39%), dan platelet (PLT) cont berkurang dengan cepat (berpengaruh tinggi pada mengurangkan PLT), ini membuat thrombocytopenia*

**Keywords:** Human blood; CR-39 NTDs; Radon-222; Alpha particles; Invitro study

## INTRODUCTION

Radon ( $^{222}\text{Rn}$ ) is a natural radioactive gas, emitting from radium ( $^{226}\text{Ra}$ ) decay. Three radioactive isotopes  $^{218}\text{Po}$ ,  $^{214}\text{Po}$  and  $^{210}\text{Po}$  are generating from radon decay, the doses from radon, however, are contributed to by the radon decay products (daughters) rather than by the gas itself. Thus, these isotopes will make risks on the human internal and external organs such as Lung, trachea (inhalation) and the skin (deposits). The exposure due to deform in DNA, blood components and improve the cancers (Pecaut *et.al.* , 2002; Hamza *et.al.* , 2009). – (whole paragraph - please do some checking on grammar)

When nuclear radiations (Alpha, Beta & Gamma) enter the body, some of their energy will lost in collisions with the body's atoms. Ejected electron loses energy by causing additional ionization; deflected alpha goes on to cause additional ionization. The major characteristic of the organ's atomic interactions is the stripping away of electrons from atoms in the body. This removal of electrons called ionization, therefore, alpha, beta, and gamma radiation often called ionizing radiation .The ionizations and their effects on cells cause the biological effects of radiation, and the body cells depend on individual atoms working together(Pohl-Rüling *et.al.*, 1990).

Alpha particle energy relatively heavy, positively charged particles and fully absorbed within the first 20 micrometers of an exposed tissue mass. Therefore, all the radiation energy will be absorbed in a very small volume of tissue immediately surrounding each particle. Alpha particles have such limited penetrating ability that the maximum range for the highest energy alpha particle in tissue is less than 100 micrometers. Beyond a radius of about 20 micrometers, so the deposition of energy is very small (Somosy , 2000). (Grammar)

When the human body exposes to radiation doses will cause injury to tissue systems from free radical damage. Exposure can be acute or chronic; this depends on total dose, dose rate, distribution of dose and the susceptibility of the patient to the radiation. Tissue systems with greater rates of cell division, such as the hematopoietic and gastrointestinal systems, In addition, platelet levels at the time of diagnosis could be a useful prognostic factor in lung cancer. Thus, designing and modification in radon irradiation technique in the field of hematology for an



**Figure1:** Two blood tubes. Tube A: after standing, tube B: contains freshly drawn blood

invitro study considered an interest scientific research (Geoffrey, 1998; Francisco et al, 2009; Pecaut *et al*, 2002).

As known, blood accounts for 8% of the human body weight with an average density of approximately  $1.060 \text{ kg/m}^3$  (3-4). The average adult has a blood volume of roughly five liters, composed of plasma and several kinds of cells; these formed emends of the blood are erythrocytes (red blood cells), leukocytes (white blood cells), and thrombocytes (platelets). By volume, the red blood cells constitute about 45% of whole blood, the plasma about 54.3%, and white cells about 0.7%. Therefore, important parameters should be taken into account during design or modify any radon exposure technique. Radon's parameters; half time, concentration, detecting method, controlling, range of alpha particles, restricted energy loss and the detector efficiency. Blood parameters; mixing blood components (see Fig.1. – not functional to this part of text), laboratory's temperature/ humidity, blood age and blood air exposure (to avoid Bactria and virus), because the exposure to ionizing radiation due to increases the number of chromosome aberrations in human blood lymphocytes (Stephan et. al., 2005; Elaine et. al.2001). (grammar)

Decrease in the number of platelets in the blood due to a thrombocytopenia, which can result in poor blood clotting. Thrombocytopenia usually defined as less than 150,000 platelets per cubic millimeter of blood (Krailadsiri et. al. 2001). Therefore, for study the effects of alpha particles (that are emitting from radon's progeny) on thrombocytopenia, new technique and idea presents in this research to irradiate human blood samples, using a nuclear track detector type CR-39; invitro study. (Whole paragraph – please check grammar)

## RESEARCH METHODOLOGIES

### Research Materials

**CR-39 Nuclear Track Detectors (NTDs)** : CR-39NTD is a  $\text{C}_{12}\text{H}_{18}\text{O}_7$  polymer with the density of  $1.31 \text{ g/cm}^3$  thickness is  $700\mu\text{m}$  cut into  $(1 \times 1.5) \text{ cm}$  engraved code (16 pieces are used in this research) . It produced from INTERCAST EUROPE SRL (43100 PARMA-ITALY). The

efficiency of CR-39NTDs changes with the time, therefore, the calibration process is interesting request before use it (Ismail, 2009). In present work, CR-39NTDs calibrated and the efficiency of it was 80.1% (Ismail & Jaafar, 2009).

### ***RAD7***

Radon Detector version7 (RAD7) is a highly versatile instrument for short-term measurement, that can form the basis of a comprehensive radon measurement system. It is used to the calibration and known a radon concentration in each stage of irradiation; so, it considered a part of blood irradiation technique.

### ***Radiation Sources***

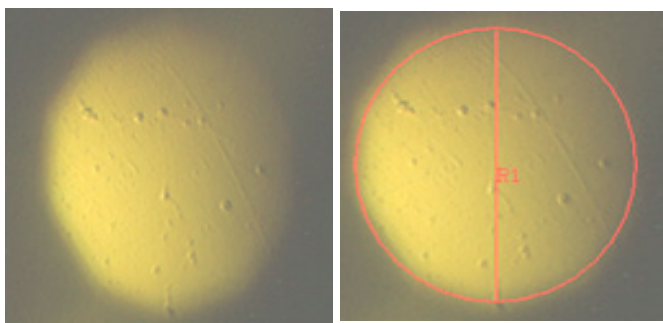
Two radium ( $^{226}\text{Ra}$ ) source with the activity  $5\mu\text{Ci}$  use to combination of radon gas into the PVC cylinder chamber (length = 21cm; diameter= 7cm), it is used as a controlling radon gas (radon concentration is changeable) to blood exposure.

### ***Radiation Dosimeter***

The geometry form of radiation dosimeter type VICTOREEN, model 491-30, USA makes it consider a suitable dosimeter for the present technique; it used to know the average radon dose inside the irradiation chamber (PVC chamber). And for the calibration, digital dosimeter type RAM DA3-2000, model 2-0033-10, RCTEM INDUSTRIES C€ used, as shown in Fig.2.

### ***Water bath and chemical solutions***

Etching systems consist on etch track detectors at 6Normality of NaOH at  $70^\circ\text{C}$ , distiller water and water bath "GOTECH TESTING MACHINES INC" model GT-7039-M, 220 volt, 50Hz.



**Figure 3:** Alpha track register by CR-39NTDs (Area  $0.101912\text{ mm}^2$ )

### ***Microscope Image and Scanning***

The etched tracks observed using an optical microscope fitted with an objective lens of 400-time magnifications. At this magnification, counting field covers an area ( $=0.10912 \text{ mm}^2$ ). The microscope image viewed with a high-quality monochrome charge coupled device (CCD) TV camera, which connected to a PC-based image analyzer, as shown in Fig.3. Measurements carried out under magnification of 400X for a total scanned area of the detector equal to  $1.5 \text{ cm}^2$ .

### ***Blood Sample***

Blood samples (2.5 ml) collected by venipuncture into heparinized syringes for 20 male and 20 female (20- 52 years old) using established blood-borne pathogen/biohazard safety protocols. This study conducted with the cooperation of the wellness center of Universiti Sains Malaysia (USM). Blood samples to the exposure of radon gas have to be prepared inside a plastic dish (diameter = 1.42cm: depth =0.5cm).

### ***Blood Analysis Machine***

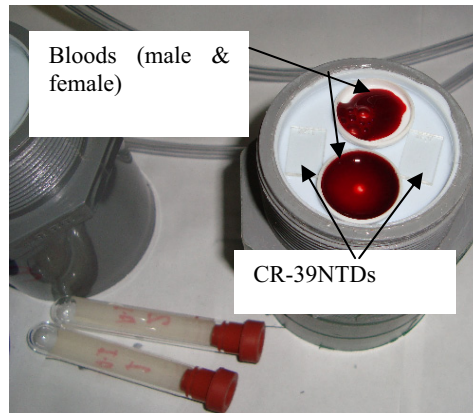
Sysmex pocH-100i 3-Part Hematology Analyzer used to analytic human blood samples (See Fig.4). The instrument designed for a laboratory running 20 or fewer samples a day. The operation of the instrument is simple. It takes about 5 minutes to start up and 3 minutes to shut down, with an analysis time of approximately 148 seconds per sample. More details are available in the web of it. (Check English Grammar)



**Figure 2** : Radiation dosimeters (a ): RAM DA3-2000 and (b): VICTOREEN.



**Figure 4:** Blood analysis ; [Sysmex pocH-100i 3-Part Hematology Analyzer.](#)



**Figure 5:** Systematic of human blood exposure to radon gas.

## Experiment procedure

### *Procedures of Irradiation Technique*

Specifically, the concept and idea of present design (radon exposure technique to an invitro study of the human blood) come from explain radiation effects on blood components via evaluating alpha particle density, range and restricted energy loss of alpha particles into the blood samples. For investigating that purpose, alpha tracks into the surface of CR-39NTDs used as a correlation and relative method. Therefore, thickness ( $\approx 0.5\text{cm}$ ), volume ( $\approx 0.5\text{ml}$ ) and place of blood samples justified with the thickness and place of CR-39NTDs, as shown in Fig.

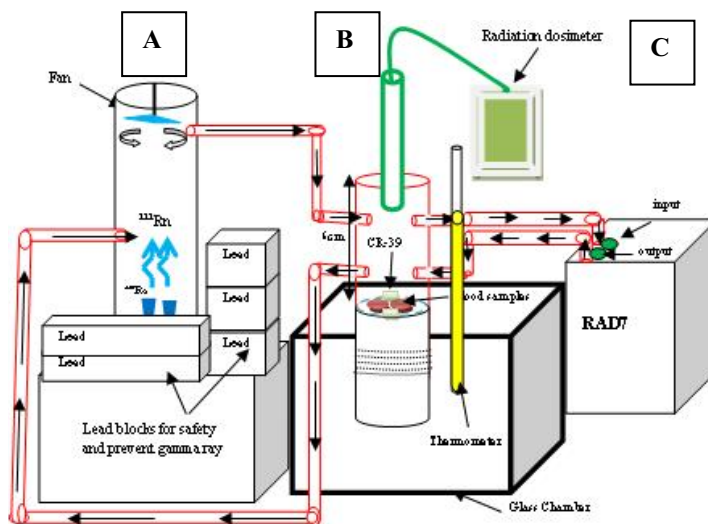
5. Thus, by knowing the number of monuments belonging to the surface of the detector can find out the amount of alpha particle incidents on a blood.

Generally, present technique (see Fig.6) consists of three parts; first part (A), is the PVC radon chamber radon, to collect radon gas inside a control chamber (i.e. radon concentration to be under control) two sources of radium were put inside the mentioned chamber. After 28 days of exposure, radon concentration is saturated, and then it used to blood exposure. Second part (B), this part included systematic of human blood and CR-39NTDs exposure to radon gas, radiation dosimeters (type VICTOREEN), Thermometer and the radon chamber (high =7cm, diameter 6 cm) (Ismail & Jaafar, 2009). Third part (C), include RAD7 that used to measure radon concentration during the period of exposure. Specifically, Rad7 use to demonstrate estimate that the CR39NTDs are in high efficiency. Continuity in the measurement of exposure and declined further comment are two ways; RAD7 and CR-39NTDs considered important points in this technique.

Blood samples (male and female) have put inside plastic dishes, then fixed temporary beside two pieces of CR-39 to exposure radon gas. Thus, radon exposure for different time (6, 10, 15, 20 minutes) done for the select human blood samples (20 male & 20 female). To mixing blood components and to avoid a plasma deposit above red blood, both; blood samples and CR-39 NTDs moved slowly (left to right, right, also appositively) without any effect on radon concentration, because the design supplied with the scroll move. Age of blood samples has taken into account, blood tests before and after irradiation was less than one hour.

In addition, before using radon exposure technique, and for control (to avoid the effects of Virus and infection on the results) , complete blood test for 20 human blood samples processed before and after selected time ( 6 , 10, 15 , 20 minutes ) .

Exposure dose was 10 to 41  $\mu\text{Sv/hr}$  and for exposures to the radiation dose of the radon gas, whole blood samples transported at a temperature of  $(27 \pm 2.2 \text{ } ^\circ\text{C}^0)$  inside the technique. SRIM 2010 program used to calculate the range of alpha particle and restrict energy loss into the blood and CR-39NTDs.



**Figure 6** Schema diagram of human blood exposed invitro to radon gas (irradiation technique).

### *Etching and scanning process of CR-39 NTDs*

The etching's systems consist on etch track detectors at six Normality of NaOH at 70C°, distiller water and water bath “GOTECH TESTING MACHINES INC” model GT-7039-M, 220 volt, 50Hz. For scanning process, the etched tracks observed using an optical microscope fitted with a magnification of 50 X to 1000X. The microscope image viewed with a high-quality monochrome charge coupled device (CCD) TV camera, which connected to a PC-based image analyzer.

## RESULTS AND DISCUSSION

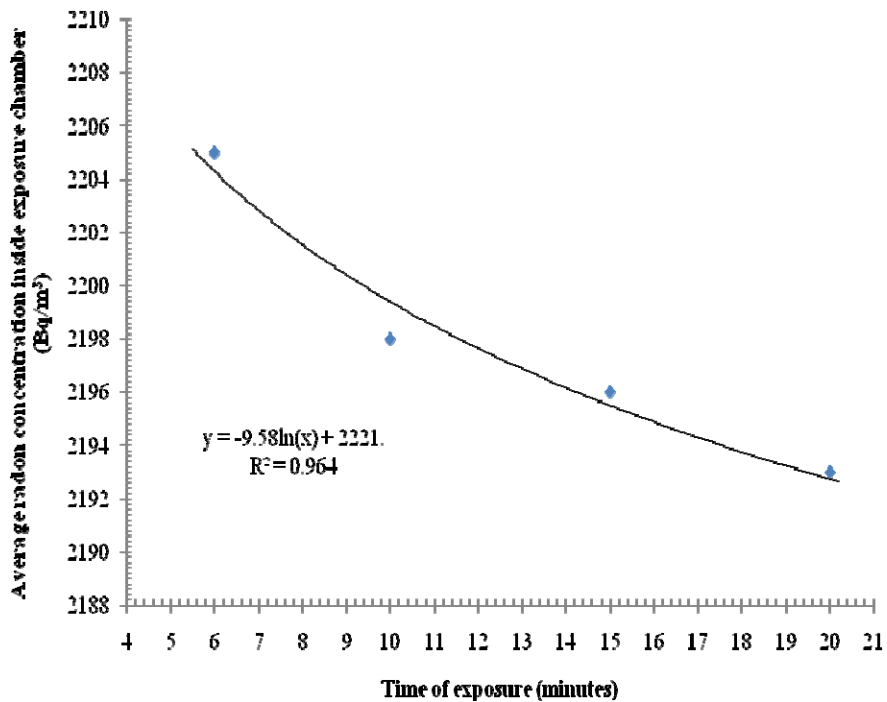
The variation of the range of alpha particle was depended on the energy of it and the target density (blood or CR-39NTD), this depended on the restricted energy loss, as shown in Table 1. High LET radiation of alpha particles, which emitted from  $^{226}\text{Ra}$ , is more efficient in inducing biological damage because all the energy deposited within a short distance, causing dense ionization in the trajectory.

**Table 1:** Range and energy loss of alpha particles into the blood samples and CR39NTDS using SRIM-2010 program. (Check the Columns' entry)

Alpha particle energy(MeV)	Range of alpha particles in CR-39NTDs ( $\mu\text{m}$ )	Range of alpha particles in blood ( $\mu\text{m}$ )	Energy loss alpha particle in blood (MeV / ( $\text{mg}/\text{cm}^2$ )) $\times 10^{-3}$	Energy loss alpha particle in CR-39NTDs (MeV / ( $\text{mg}/\text{cm}^2$ )) $\times 10^{-3}$
1	4.3	5.18	2.82	2.54
2	8.57	10.13	1.56	1.40
3	14.11	16.66	1.10	0.996

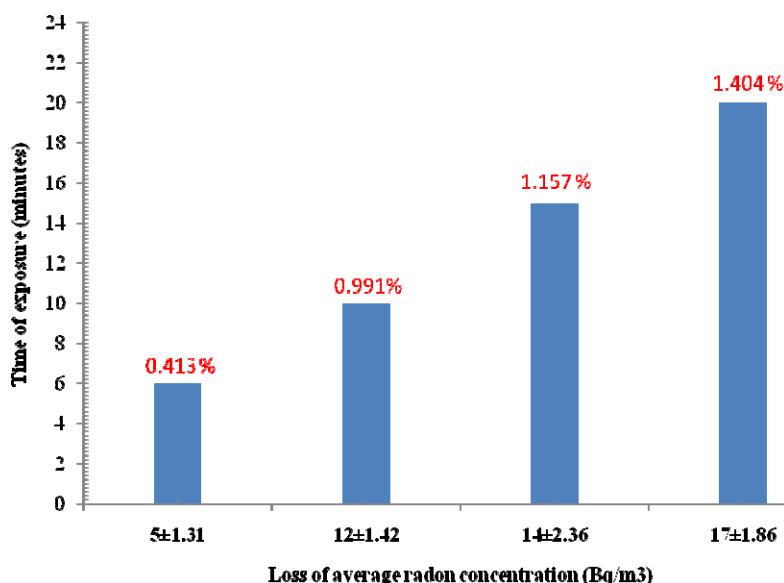
Average radon concentration ( $2198 \pm 5.1 \text{Bq}/\text{m}^3$ ) in PVC chamber ranged between 2194 to 2206  $\text{Bq}/\text{m}^3$ , as shown in Fig. 7 9 (but the figure shows decrease in conc, not average or range ...). This means that there is relatively a loss in radon concentration in each stage of exposure, because there is absorbed dose by CR-39NTDs and the blood sample, also the process of the mixing of blood samples affected on keep of radon concentration.



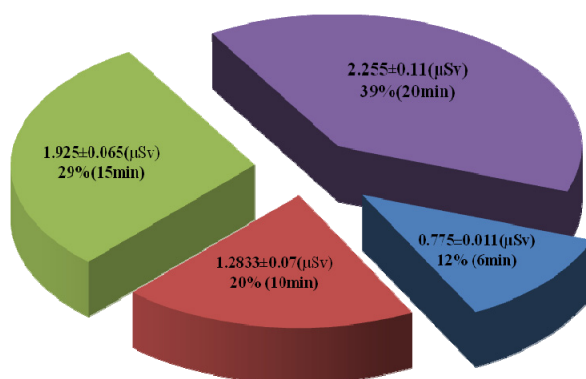


**Figure 7:** Decreases of radon concentration with the stages of exposure during exposure. (What physical basis or justification in using  $\ln$  to the data points?)

Average loss ratio of radon concentration variable from 0.413% to 1.404 %, this depends on time of exposure, as shown in Fig.8. This means that the efficiency of the present design that related to keeping radon concentration for blood exposure very good (= 96%). On the other hand, average absorbed dose by selected samples (CR-39NTDS +0. 5ml Blood sample) was enough for the selected time of exposure. Thus in Fig. 9, rate of radiation absorbed dose into the human blood samples is high at 20 minutes, so the effects on the thrombocytopenia high as shown in Fig.10. In fact, anyone can be exposure to that periods and that concentration of radon gas during workplace, so the present design considered an important system to knowledge the effect of radon gas on the blood component, especial platelet cont.



**Figure 8:** Loss of radon concentration (not tally with your horizontal axis) during the periodic of exposure; changed accurate in each stage of exposure, relatively.



**Figure 9:** Rate and percentage occupation absorption dose into blood samples for various exposure times.

**Table2:** Human blood (0. 5ml) and CR-39NTDs exposed to the Radon ( $^{222}\text{Rn}$ ) Gas; average exposed dose rate and radon concentration ( $=7.7\pm0.041$ )  $\mu\text{Sv/hr}$  and ( $=2198\pm5.1$ )  $\text{Bq.m}^{-3}$ , respectively. Average laboratory temperature ( $=17.975\pm0.05$ )  $^{\circ}\text{C}$ .

Time of exposure (minutes)	Average radon concentration( RAD7) ( $\text{Bq/m}^3$ ) Inside exposure chamber	average density of alpha into the blood and CR-39NTDs ( $\text{track/cm}^2$ ) per time of exposure	Average dose rate inside exposure chamber ( $\mu\text{Sv/hr}$ )	Average absorbed dose by blood and CR-39NTDs ( $\mu\text{Sv}$ )	Reduced value of PLT count $\Delta\text{PLT}$ (after and before exposure) Male:29 years old	Reduced value of PLT count $\Delta\text{PLT}$ (after and before exposure) Female:30 years old
6	2205 $\pm$ 18.15	<b>262.088<math>\pm</math>18.42</b>	7.75 $\pm$ 0.01	0.775 $\pm$ 0.011	9	7
10	2198 $\pm$ 28.51	<b>284.166<math>\pm</math>22.06</b>	7.7 $\pm$ 0.07	1.2833 $\pm$ 0.07	28	18
15	2196 $\pm$ 17.87	<b>326.065<math>\pm</math>22.8</b>	7.7 $\pm$ 0.03	1.925 $\pm$ 0.065	16	15
20	2193 $\pm$ 23.12	<b>350.106<math>\pm</math>14.8</b>	7.65 $\pm$ 0.14	2.255 $\pm$ 0.11	30	21
<b>Ave. <math>\pm</math> SDV</b>	<b>2198<math>\pm</math>5.1</b>		<b>7.7<math>\pm</math>0.041</b>			

As well as, results of average exposure and absorbed radon concentration into CR-39NTDs and the human blood samples (male and Female) with the time of exposure are listed in Table 2. Composite of alpha particle into the surface of blood samples measured via measuring tracks of alpha particles into the surface of CR-39NTDs are observed markedly.

In addition, average absorbed dose via the exposure samples increased with increasing time of exposure, this made the number of platelet count decreasing relatively. The reason of this phenomenon due to high dose of alpha particle, this is making the greatest damage to the blood cells, that the alpha particles are heavy ions. Thus, the alpha particles will be losing most of their energy at a short distance in their trajectory for irradiation.

The effect of radon absorbed dose by the blood sample variable, because biological factors, which may influence the effect of alpha radiation on an individual include age and sex, as shown in Fig.10.

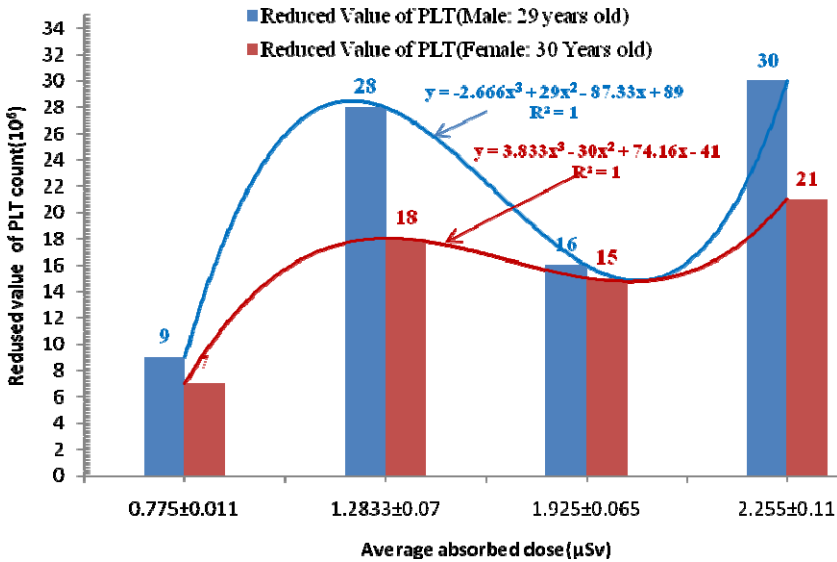


Figure 10: Relative change in reduced of platelet count (PLT) with average absorbed radon dose.

As well as, alpha particle density into the surface of CR-39NTDs and the human blood samples increased with the time of exposure as shown in Fig.11. Therefore, the mentioned design considered as an effect and suitable way to the *invitro* studies of hematology.

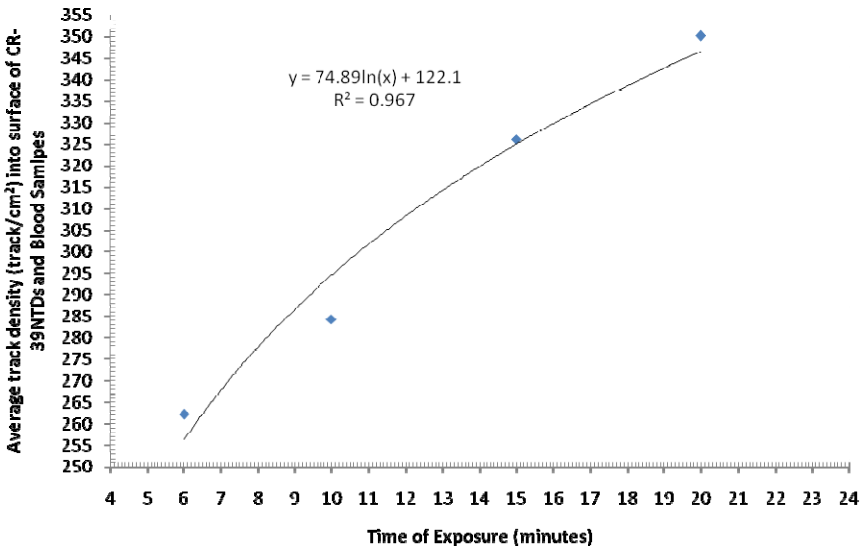


Figure 11: Variation of accumulation of the alpha particles into the surface of CR-39 NTDs and the human blood sample with the time of exposure.

## CONCLUSION

The distributional methods for alpha particles and radiation dose on the surface of CR-39NTDs and the completely human blood samples have been significantly improved through the design new system to exposure. In a new design, there was a little loss of radon concentration pursuant to several factors including the method of mixing components of blood sample and the quantity of absorbed dose. However, this did not affect on the efficiency ( $\approx 96\%$ ) of the present design.

The range of alpha particle in CR-39NTDs was less than the range of it in the human blood samples, depending on their density and restricted energy loss. Comparative study between CR-39NTDs and the human blood samples was a new technique for *invitro* studies of the blood ionization, especially to estimate ionization of alpha particle. Therefore, CR-39 NTDs considered as the most suitable nuclear detector to get alpha particle density deposited on the blood surface.

The present design is applicable in an invitro study of the blood ionization, decrease of the platelet count in each stage of exposure (different time of exposure), relatively, refers to an effect of radon absorption dose from 0.5ml of human blood on the blood components. In high time of exposure ( $\approx 20$  minutes), the change value of the platelet count before and after exposure go to high (reduce of PLT go to high).

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