Sarcouncil journal of Medical sciences

ISSN(Online): 2945-3526

Volume- 03| Issue- 06| 2024

Research Article

Received: 28-04-2024 | **Accepted:** 26-05-2024 | **Published:** 13-06-2024

Assessment Study of the Effect of Using Ascorbic Acid with and Without Contrast Agent on the Blood Samples Exposed with Different Doses of X-Ray in Fluoroscopy by Cyclic Voltammetry

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Abstract: The effect study of blood samples that were exposed at different doses of X-ray using the Fluoroscopy technique by electrochemical analysis with the oxidation-reduction properties using the cyclic voltammetric technique to find the oxidative stress of the radiation of X-ray on the blood composition without contrast medium and with contrast, and also, the study focused on the impact of the ascorbic acid as an anti-oxidative reagent on blood without contrast and with contrast medium (iodine solution). In the patient and method that was chosen, 40 patients were divided into 20 females and 20 males, with the range of age from 20 to 40 years. Ten of the patients as control samples without any exposed to the X-ray; the other 30 patients were divided into three groups ten patients exposed with different doses of X-ray (300-600 reem) using fluoroscopy type Shimadzua, type HP/Flowro without contrast medium, ten patients was injected (by IV) with contrast medium (Iodine solution) and exposed with the same different doses of x-ray by fluoroscopy technique to study the effect the radiation on the blood with contrast medium, and the other ten patents were injected with contrast medium and with ascorbic acid to study the impact of ascorbic acid as anti-oxidative reagent on the exposed blood with contrast agent samples. The results illustrated that the blood samples which exposed to various doses of radiation were caused to enhance the oxidation current peak regarding the blood as proportionality with increasing of the radiation dose compared with the control blood sample (not exposed to radiation) and the other blood samples that injected with contrast medium (Iodine solution) were enhanced the oxidation peak more than the control samples and also the samples exposed without contrast medium. The other study was used ascorbic acid, which acts as anti-oxidative stress to the blood with and without a contrast medium. It can be concluded that the exposed of patients in X-ray caused a high oxidative stress of the blood composition with and without contrast medium, which show more effected with contrast medium, and the ascorbic acid acted as an anti-oxidative reagent that decreased the oxidation current peak when injected the patients with and without contrast medium.

Keywords: Fluoroscopy, Iodine solution, Ascorbic acid, Cyclic voltammetry, Blood samples.

INTRODUCTION

Fluoroscopy can be defined as one of the medical imaging methods for creating real-time images regarding the internal structures of the body using X-rays. Known commonly as a contrast agent, contrast medium is frequently utilized in fluoroscopy for improving the visibility regarding specific fluids or parts, like the blood vessels or gastrointestinal tract (Hill, N. E., & Giampetro, D. M. 2023; Rednam, M., & Tiwari, V. 2021; Imai, K. *et al.*, 2022; Macready, D. M. *et al.*, 2007; Forbrig, R. *et al.*, 2020). Vitamin C, or ascorbic acid, is a popular antioxidant as well as a dietary supplement. There is a dearth of scholarly material on the precise topic of its impact on the contrast medium in fluoroscopy (Crawley, M. T. *et al.*, 2004; Aroua, A. *et al.*, 2007; Radhi, M. M. *et al.*, 2015; Radhi, M. M. *et al.*, 2017). When combining the pulsed and low-dose modes, RADs were considerably lower than when utilizing the traditional method alone.

Consequently, radiation exposure to patients and clinicians can be minimized through appropriate utilization of fluoroscopy and its C-arm positions (Cho, J. H. *et al.*, 2011). For creating a straightforward and practical technique for detecting vitamin C in real samples, the electrochemical characteristics of vitamin C (ascorbic acid) have been investigated using cyclic as well as differential pulse voltammetry in model systems. The findings showed that the ascorbic acid oxidation product, dehydroascorbic acid, is adsorbed on the surface regarding the glassy carbon electrode and that vitamin C oxidation is a diffusion-controlled and semi-reversible process. By utilizing cyclic voltammetry, vitamin C was effectively measured in real samples (supplements and fruit juices) in the concentration range of 0.034 mol dm-3 to 0.340 mol dm-3. Furthermore, the DPPH test was utilized to ascertain the antioxidant activity regarding vitamin C in real samples. A strong linear connection has been observed between the outcomes of cyclic voltammetry and the results assessed through the DPPH assay of the samples (Blažević, J. *et al.*, 2020). Medical drugs known as radiographic contrast medium (RCM) are utilized in X-ray-based imaging procedures to enhance the image of internal organs and structures. It can cause side effects that range from itching to contrast-induced nephropathy (CIN), a potentially fatal condition. CIN is specified as acute renal failure, which cannot be attributable to any other cause and occurs 24-72 hours after exposure to RCM.

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DOI-https://doi.org/10.5281/zenodo.11641366

Patients who already have diabetes and renal impairment are typically the ones who experience it. Reactive oxygen species (ROS) formation, direct damage to tubule cells, and reduced cerebrospinal blood flow resulting in hypoxia are the causes driving CIN. Finding patients who are at risk of getting CIN is crucial. We have examined risk factors and preventative measures, and we have included a lengthy list of references so that readers can assess each in-depth. When a patient is at risk for CIN and is having radiography, the initial recommendation is to measure their serum creatinine and calculate their glomerular filtration rate prior to the procedure and once a day for five days following it to monitor their renal function (Andreucci, M. *et al.*, 2014).

To investigate the impact of oral radioprotective agent on the degree of γ-H2AX foci formation generated by X-rays, using a mixture of antioxidants and glutathione-raising agents. 25 healthy participants' blood lymphocytes have been used in in vitro tests, both without and with antioxidants added, either prior to and following irradiation (10 mGy). Blood samples have been taken before, and 15, 30, and 60 mins (n = 17) and 2, 3, and 5 hrs (n = 11) following oral antioxidant tablets have been taken and irradiated for the in vivo/in vitro testing. (10 mGy). Through counting the γ-H2AX foci, DNA double-strand breaks (DSBs) have been measured in isolated cells for 5 min (in vitro and in vivo/in vitro) and 15 mins (in vitro) following irradiation. Further in vitro tests were conducted utilizing 53BP1, an additional independent marker for DSBs, for validating the data. The control samples were non-irradiated ones. Antioxidants significantly decreased average levels of γ-H2AX foci by 23% (P<.0001) in in vitro tests when they were preincubated for 15 minutes while adding antioxidants just following irradiation had no effect on X-ray-induced foci. (p = 0.6905). Antioxidant oral pretreatment produced a considerable reduction in in vitro and in vivo studies (Kuefner, M. A. *et al.*, 2012). In the case when combined with contrast administration, fluoroscopy is a useful imaging modality which could minimize radiation exposure to patients while providing real-time functional as well as anatomic assessment of the patient. Costs associated with medical treatment could be reduced by using this approach to direct therapeutic interventions, which could be carried out in outpatient settings. In order to improve interprofessional communication between healthcare providers and lower infection rates, such activity reviews the advantages regarding fluoroscopic-guided contrast administration, the mechanism of action of contrast utilized in fluoroscopy, and the management of patients undergoing fluoroscopic contrast injections.

Morbidity and mortality rates among patients having these treatments (Hill, N. E., & Giampetro, D. M. 2023). Comparing exposed blood in females and males, blood incubated with iodinated contrast medium alone, and blood exposed to radiation all showed no change in hematological parameters or red blood cell morphology in the case when put to comparison with control blood in all fluorescent X-ray machines. Following a 5-minute incubation period with 5 mg/ml iodinated contrast fluid, female and male blood samples were exposed to 10, 30, and 60 minutes of X-ray fluorescence (70 kV, 0.8 mA) radiation. Clinical service center assessed the hematological parameters regarding blood samples and used optical microscopy to look at the morphology of red blood cells. Student's t-tests were used to compare the mean values of hematological indices between blood that had been exposed to radiation alone, blood that had been incubated with iodinated radioactive contrast medium alone, and blood that had been subjected to each matching control blood. Those findings suggest that when human blood is exposed to fluorescent X-rays using iodinated radiography contrast material in a laboratory setting, no negative consequences arise (Supawat, B. *et al.*, 2021).

Thermal dosimeters (TLDs) are used to measure the radiation exposure levels that veterinary staff members get at work when performing fluoroscopy. Three participants with TLDs—two enrolled and one observer—were the subjects of a prospective study to determine their radiation dosimetry. TLDs were positioned both within and outside the arm shield; thyroid shield, apron, and panorama mask bulletproof similar protective items. There were five anatomical areas where TLDs were found: the eye, breast, thyroid, hand, and gonad. The hand radiation dosage was the greatest of all the lead shielding devices for all three participants, and observer C's thyroid radiation dose result was identical to the hand radiation dose. Each of the three subjects' eyes received non-negligible radiation exposures. Veterinary professionals should wear protective gear, particularly for their eyes, when they are

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exposed to radiation from fluoroscopy as well as radiography (An JeongSu, A. J. *et al.*, 2019). Initial institutional (local) LDRLs have been established through comparing the evaluation regarding the radiation dose level that the patient had received during general fluoroscopic examinations with international diagnostic reference levels (IDRLs). With the use of medical records of a top regional hospital's radiology department, a thorough study of general endoscopic exams was carried out over the course of about a year. Sixty percent of children were found to be below the sole KAP diagnostic reference levels that were provided. Proposed are local diagnostic reference levels (LDRLs) in relation to DRLs lost due to fluoroscopy, KAP, and Ka r time. The patient dosage values for each type of examination were found to vary widely, with the mean values exceeding international diagnostic reference limits. The bulk of the examinations in the study involved extended fluoroscopy times. This calls for suitable and enhanced radiation protection training and expertise among responsible personnel, particularly equipment operators (Wambani, J. S. *et al.*, 2014).

PATIENTS AND METHOD

Preparation of Patients:

Forty patients (20 male and 20 female) with average ages from 20 – 40 years were prepared to examine the fluoroscopy with X-ray in different doses. The forty patients have been divided into four groups as follows:

Group I: The test was performed on ten patients without exposed X-rays (control group).

Group II: ten patients were exposed in different doses of X-ray from 300-600 reem.

Group III: ten patients were given a contrast medium (50 ml of iodine solution) and exposed with various doses of X-ray from 300-600 reem.

Group IV: ten patients were given contrast medium (50 ml of iodine solution) and ascorbic acid (50 ml of 0.1 Molar) and exposed with various doses of x-ray from 300-600 reem.

The four groups of patients (except group I) were examined in Fluoroscopy type Shimadzua, HP/Flowro (Japan), as shown in Fig. 1.

All groups of patients were received 1ml of blood from each patient to be exam in the electrochemical analysis by cyclic voltammetry.

Fig. 1: Fluoroscopy (Shimadzua, type HP/Flowro)

Cyclic Voltammetry (CV):

One popular electrochemical method for examining the redox processes in blood samples is cyclic voltammetry or CV.

With regard to the electrochemical analysis lab, a potentiostat/glvanostat of the NuVant Systems Inc. (USA) type was utilized.

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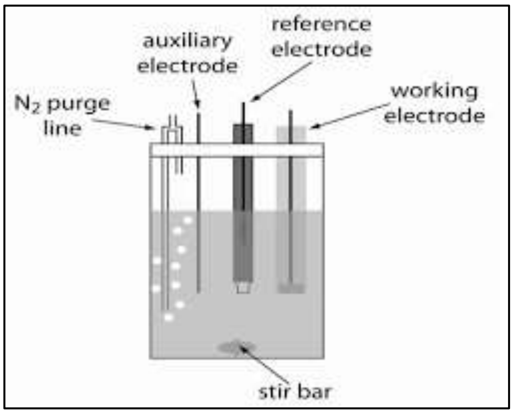
Three electrodes and a 15-milliliter Pyrex cell were utilized:

- The working electrode was a glass carbon electrode (GCE).
- The reference electrode was silver electrode Ag / AgCl (3M KCL).
- The counter electrode was the platinum wire (1 mm diameter).

To determine the characteristics of materials investigated in the blood medium, all three electrodes have been dipped in the solution under investigation and attached to the potentiometer, which was then connected to a personal computer. Scheme 1 and Figure 2 illustrate how the clean GCE was utilized in all studies following a 10- to 15-minute sonic cleaning procedure.



Scheme 1: Cyclic voltammetric cell

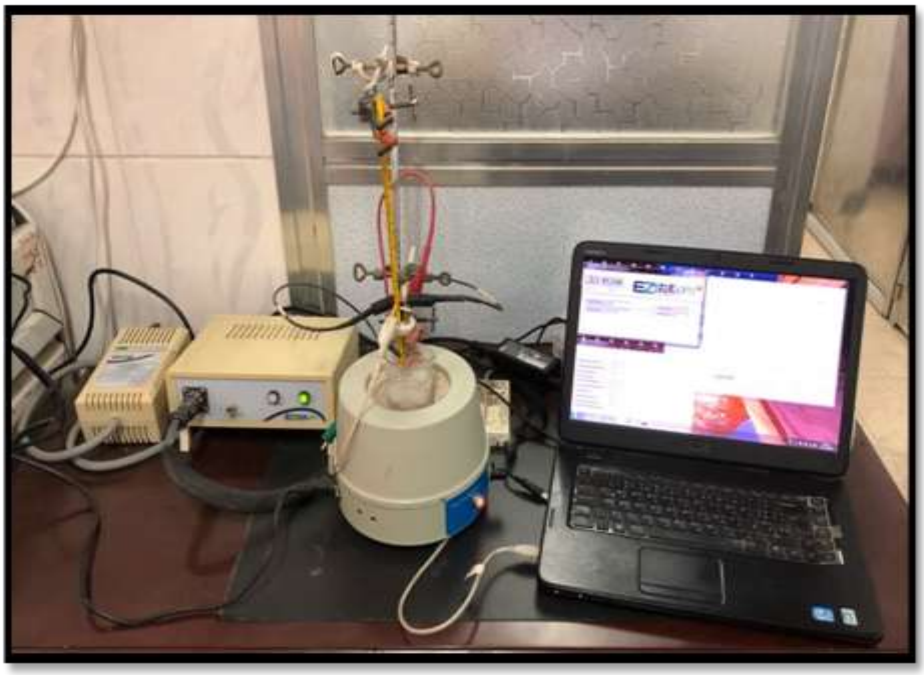


Fig. 2: Cyclic Voltammetry System

RESULTS AND DISCUSSION

By studying the blood samples that were exposed in different doses of radiation in the fluoroscopy technique in terms of oxidation-reduction reaction by cyclic voltammetry with and without contrast medium (iodine solution), and in the other study using ascorbic acid (AA) as an anti-oxidative reagent to show the effect of this reagent on the oxidative effect of the x-ray on the blood components with contrast medium (Radhi, M. M. *et al.*, 2010).

Electrochemical study by cyclic voltammetry (Effect contrast medium)

The results showed that the samples of blood which exposed to different doses of x-ray had enhanced in oxidation current peak according to increasing the radiation dose from 300 to 600 reem. It was found that contrast medium effected on the oxidation current peak of blood samples

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(with contrast medium) compared with the blood sample exposed with x-ray without contrast medium in different doses of 300, 360, 500, 525, 550, 570, 600, 630, 665, and 680 reem, as shown in Figs. 3, 4, 5, 6, 7, 8, 9, 10, 11, and 12, respectively, all Figures show the exposed patients with different doses of X-ray have effected as oxidative stress with contrast medium compared with exposed patients without contrast medium (Nuszkiewicz, J. *et al.*, 2020).

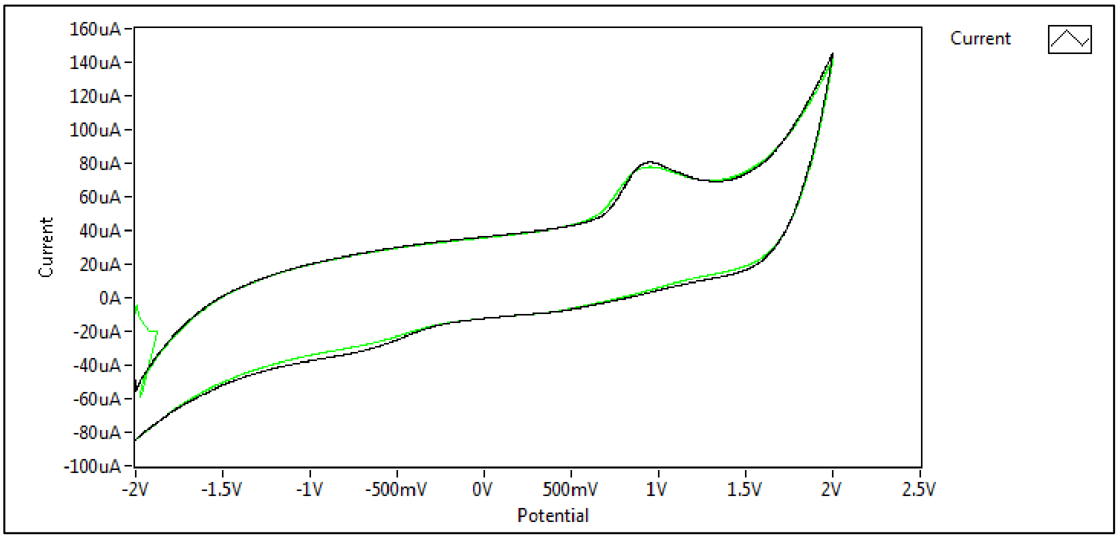

Fig. 3: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 300 Reem (black line with contrast and green line without contrast)

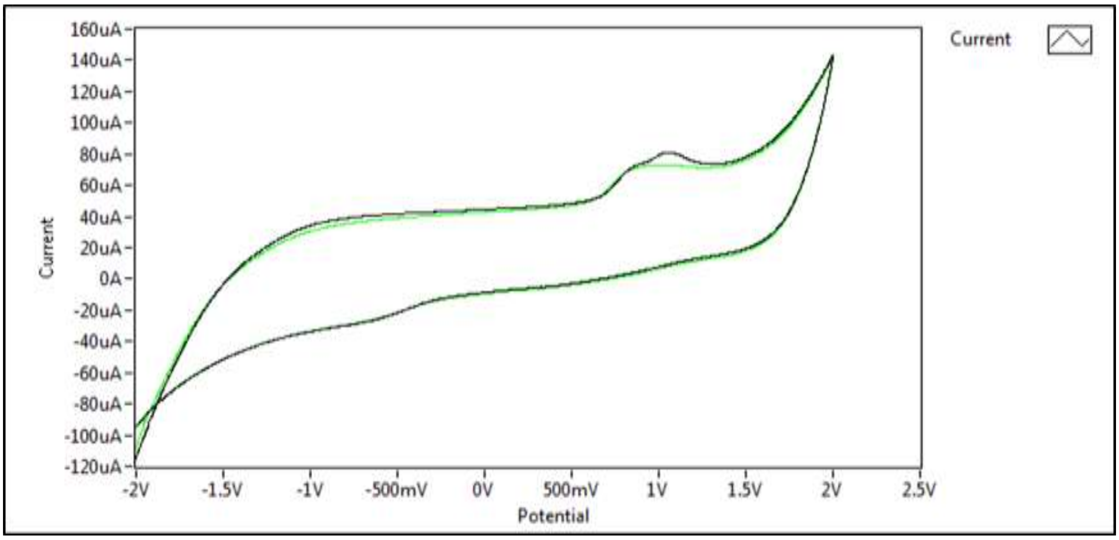

Fig. 4: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 360 Reem (black line with contrast and green line without contrast)

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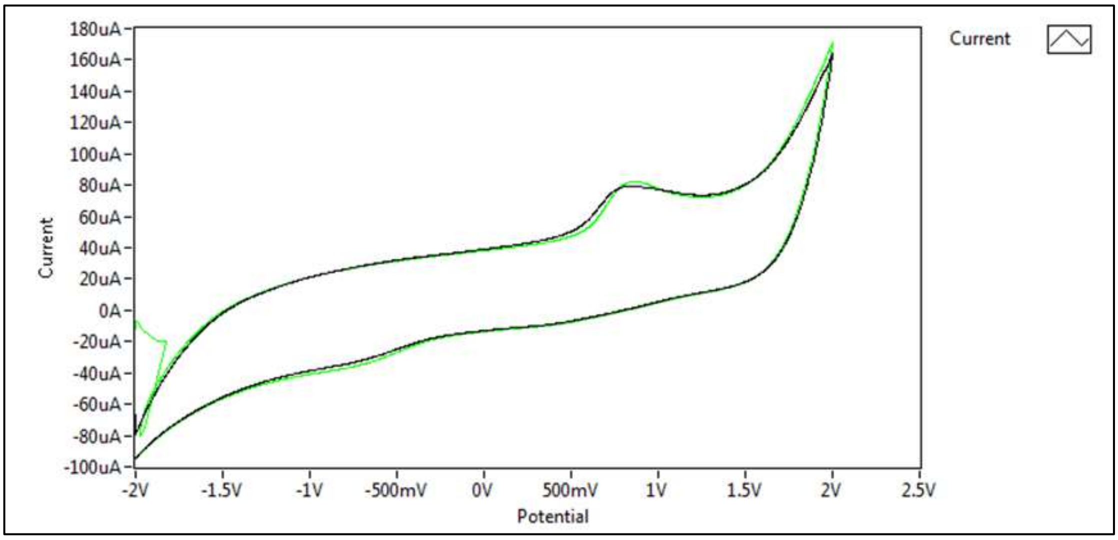

Fig. 5: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 500 Reem (green line with contrast and black line without contrast)

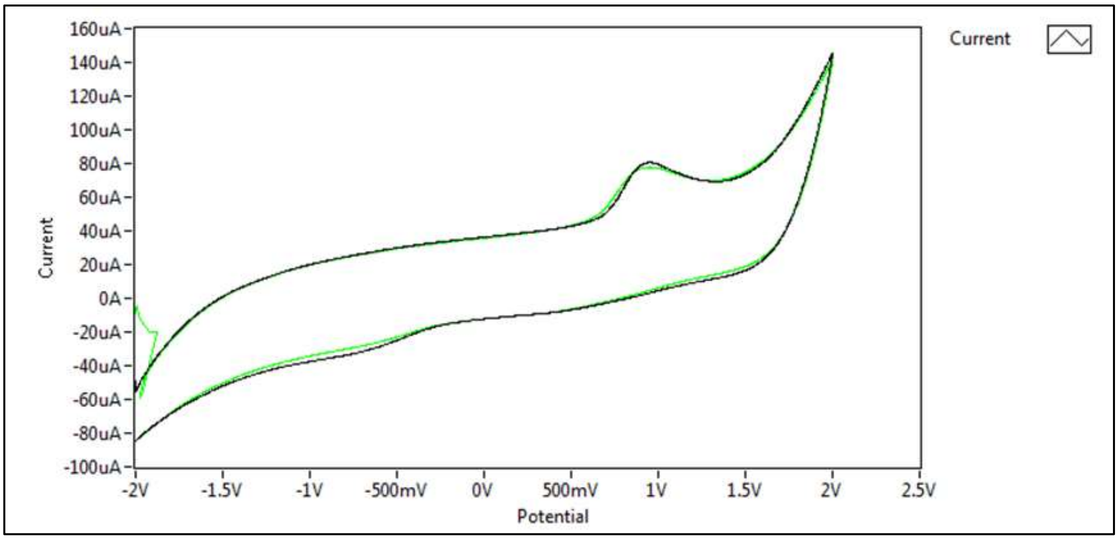

Fig. 6: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 525 Reem (black line with contrast and green line without contrast)

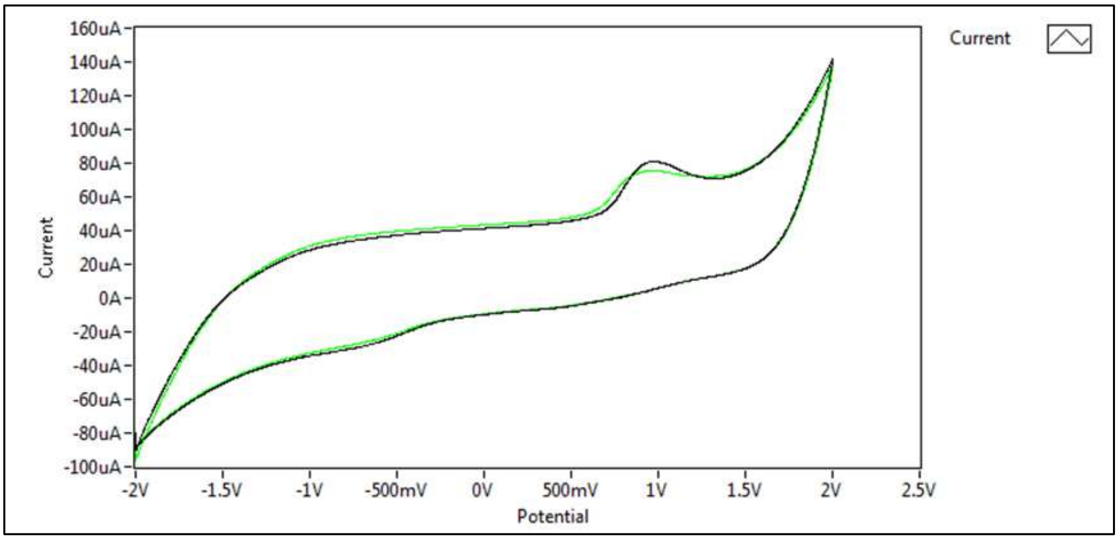

Fig. 7: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 550 Reem (black line with contrast and green line without contrast)

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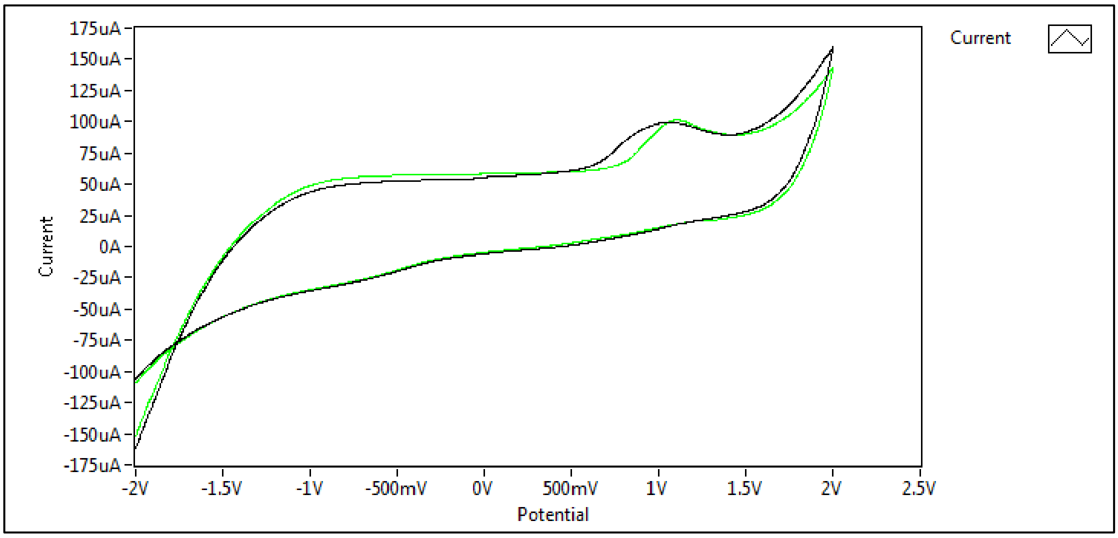

Fig. 8: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 570 Reem (green line with contrast and black line without contrast)

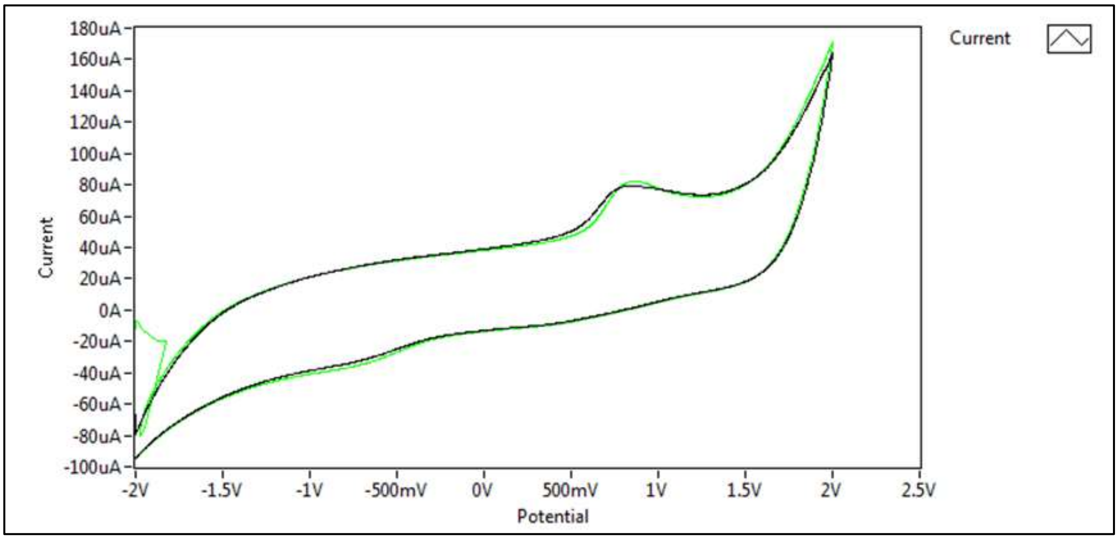

Fig. 9: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 600 Reem (green line with contrast and black line without contrast)

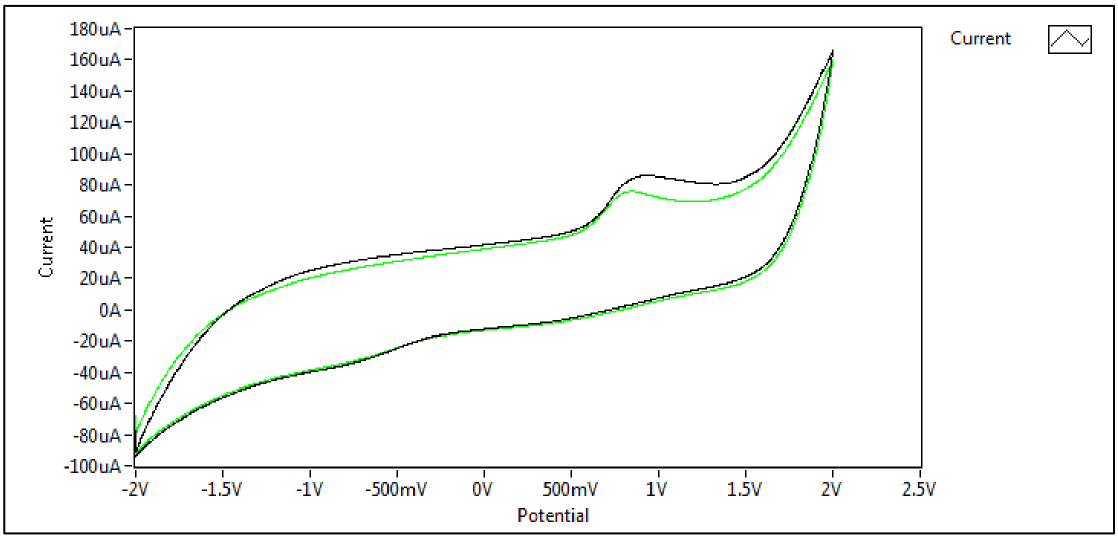

Fig. 10: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 630 Reem (black line with contrast and green line without contrast)

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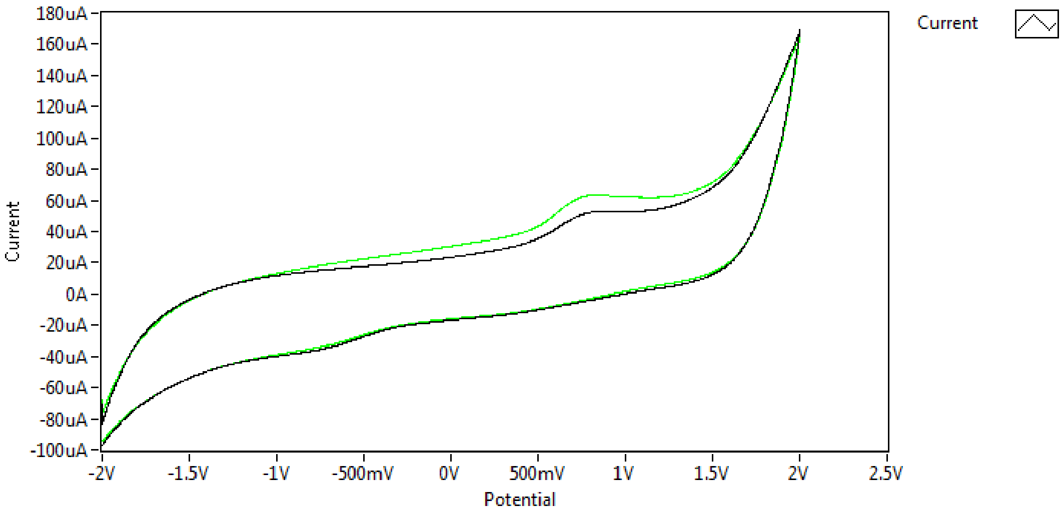

Fig. 11: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 665 Reem (green line with contrast and black line without contrast)

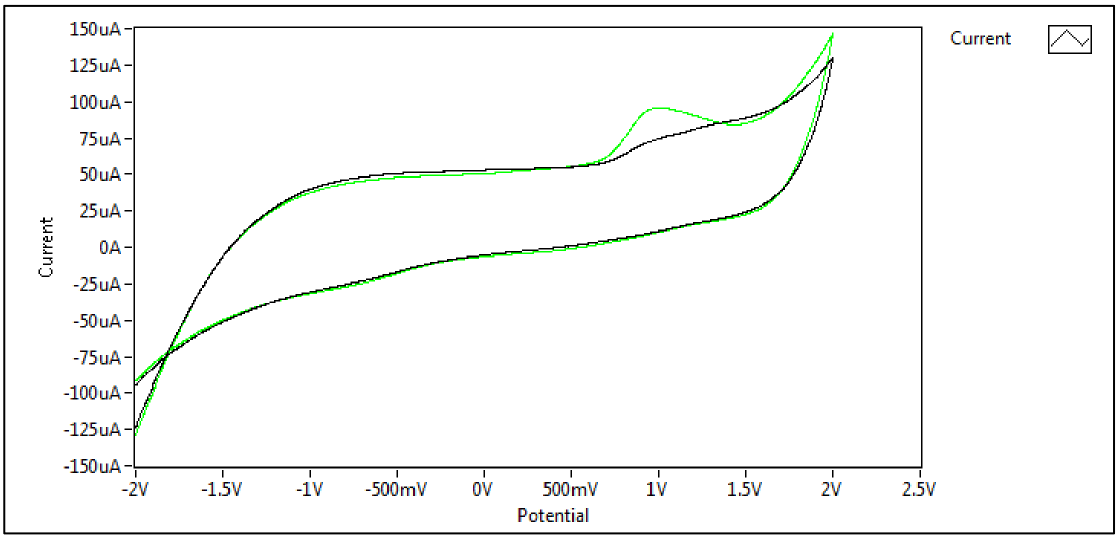

Fig. 12: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 680 Reem (green line with contrast and black line without contrast)

Effect the radiation on the blood with contrast medium

Exposure to radiation and the use of contrast medium in medical imaging can have effects on the blood. Let us discuss each of these factors separately:

Radiation and blood: When a person is exposed to radiation during fluoroscopic X-ray medical imaging procedures, it can affect the blood in several ways:

DNA damage: Radiation can cause direct damage to DNA in cells, including blood cells. This damage could result in genetic mutations and possible prolonged effects on the blood cells.

Radiation sickness: exposure to high radiation dosages could lead to a condition that is referred to as acute radiation syndrome or radiation sickness, which could cause damage to bone marrow, resulting in a considerable blood cell production decrease, in addition to other serious health effects.

Reduction in the production of blood cells: Exposure to radiation could impact bone marrow, in which the production of blood cells takes place. Reduced platelet, white blood cell, and red blood cell production could be a result of that, which could result in increasing risks of infections, anemia, and impairing blood clotting.

Contrast medium and blood: Contrast medium can be described as a substance that is utilized in some imaging procedures for the purpose of improving the view of the organs, blood vessels, or tissues. Contrast could be given intravenously (intravenously), orally, or through other

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approaches depending on utilized imaging approaches. While contrast medium mainly impacts organs that are being imaged, it can have an indirect impact on blood in the following ways:

Allergic reactions: Some individuals might have an allergic reaction to the contrast material, which could range from mild symptoms, like rash, to severe reactions, like anaphylaxis. Circulation, blood pressure, and other elements of cardiovascular health may be impacted by such allergic reactions.

Kidney function: specific contrast medium types, especially iodine-based contrast agents, can be putting stress on the kidneys. Which could result in a temporary dysfunction of the kidneys or, seldom, more serious complications like contrast-induced nephropathy. Poor function of the kidneys could have an indirect effect on blood composition and health in general.

There is an importance in noting that risks related to radiation exposure and utilization of the contrast media often outweigh the advantages and benefits of obtaining precise medical diagnoses. However, medical professionals follow strict protocols and take into account each patient's individual factors to minimize any potential risks and ensure patient safety during imaging procedures that involve radiation and contrast medium (Pasternak, J. J., & Williamson, E. E. 2012; Shalom, N. E. *et al.*, 2020; Lungu, E., & Moser, T. P. 2015; Supawat, B. *et al.*, 2021; Sharma, R.S. and Kumar, A. 2020; Böhm, I. *et al.*, 2017)..

Fig. 13 illustrated the relationship between the difference between the oxidation peak current with and without contrast medium (Iodine solution) as ΔIpa against to the different exposer doses of fluoroscopy; the results of the study was produce a linear line of the relationship with the equation:

$$Y = 0.0398\ X - 12.688 \ldots \text{ with accepted sensitivity } R^2 = 0.8621$$

It was found a clear effect of contrast medium on the oxidative stress that causes by the exposer radiation of x-ray, which has increased the oxidation through increasing the x-ray doses.

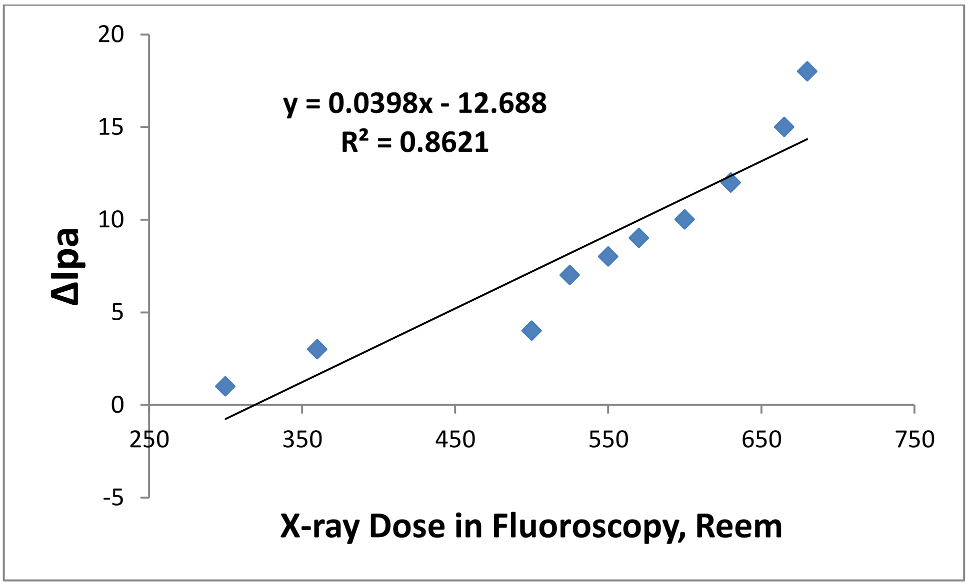

Fig. 13: Relationship between the oxidative difference (ΔIpa) of blood samples with and without contrast agent (Iodine solution) against to the different doses of x-ray in fluoroscopy (300-685 reem).

Effect the ascorbic acid on the oxidative stress of radiation in fluoroscopy.

Ascorbic acid, also referred to as vitamin C, is a powerful antioxidant that can help relieve oxidative stress caused by radiation exposure, including fluoroscopy. When the body is exposed to radiation, it can generate ROS, which could result in oxidative stress cell and tissue damage. As an antioxidant, ascorbic acid has the ability to neutralize and eliminate ROS, thereby leading to the reduction of oxidative stress as well as its adverse effects. Here's how the ascorbic acid can impact radiation-induced oxidative stress:

Scavenge ROS: Ascorbic acid is capable of scavenging and neutralizing the ROS, which includes free radicals that are highly reactive molecules produced throughout the exposure to radiation. Through the neutralization of the ROS, ascorbic acid is helpful in the prevention of oxidative damage to tissues and cells, including

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the ones that are under the exposure to radiation throughout the fluoroscopy.

Replenishing other anti-oxidants: Ascorbic acid is also effective in replenishing other antioxidant materials in the organism, including vitamin E, which represents another valuable anti-oxidant that has the ability to protect the cells from oxidative damage. Through replenishing vitamin E as well as other antioxidants, the ascorbic acid leads to the enhancement of the overall antioxidant defense system, preparing the body more effectively in combatting radiation-induced oxidative stress.

Protection of DNA: Radiation-induced oxidative stress could lead to causing damage to the molecules of DNA, resulting in genetic mutations and possible long-term effects to health. Anti-oxidant characteristics of the ascorbic acid could be helpful in protecting the DNA from the oxidative damages that result from the ROS, decreasing DNA mutation risks that result from exposure to radiation.

There is an importance in noting that even though ascorbic acid is capable of providing anti-oxidant protection, it shouldn't be taken under consideration as a standalone radiation-induced oxidative stress treatment. Using ascorbic acid as an anti-oxidant supplement must be discussed with a healthcare professional that is qualified to provide guidance on suitable dosing and possible interactions with the medications or other treatment features. In addition, other strategies to reduce radiation exposure and protect against oxidative stress, such as appropriate shielding, can also be important in the context of fluoroscopy or other radiation-dependent procedures (Jelodar, G. *et al.*, 2013; Yamamoto, T., & Kinoshita, M. 2017; Straub, J. M. *et al.*, 2015: Narra, V. R. *et al.*, 1993; Zhen, D. I. N. G. *et al.*, 2017).

Electrochemical study by cyclic voltammetry (effect AA)

The results showed that the samples of blood which exposed to different doses of x-ray in the fluoroscopy technique had enhanced in oxidation current peak according to increasing the radiation dose from 300 to 600 reem. It was found that contrast medium effected on the oxidation current peak of the blood samples (with contrast medium) and enhanced the oxidation process, while the addition of ascorbic acid solution to the patients act to reduce the oxidative stress compared with the blood sample exposed with x-ray with contrast medium in different doses of 300, 360, 500, 525, 550, 570, 600, 630, 665, and 680 reem, as shown in Figs. 14, 15, 16, 17, 18, 19, 20, 21, 22, and 23, respectively, all Figures show the exposed patients with different doses of X-ray have effected as oxidative stress with contrast medium compared with the same patients with AA solution.

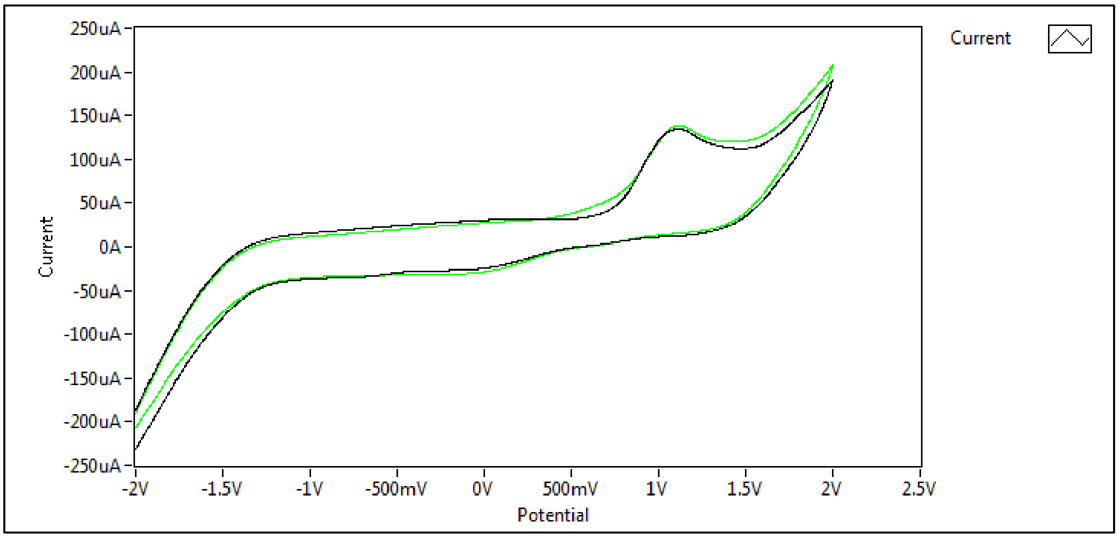

Fig. 14: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 300 Reem (green line with contrast and black line with contrast and AA)

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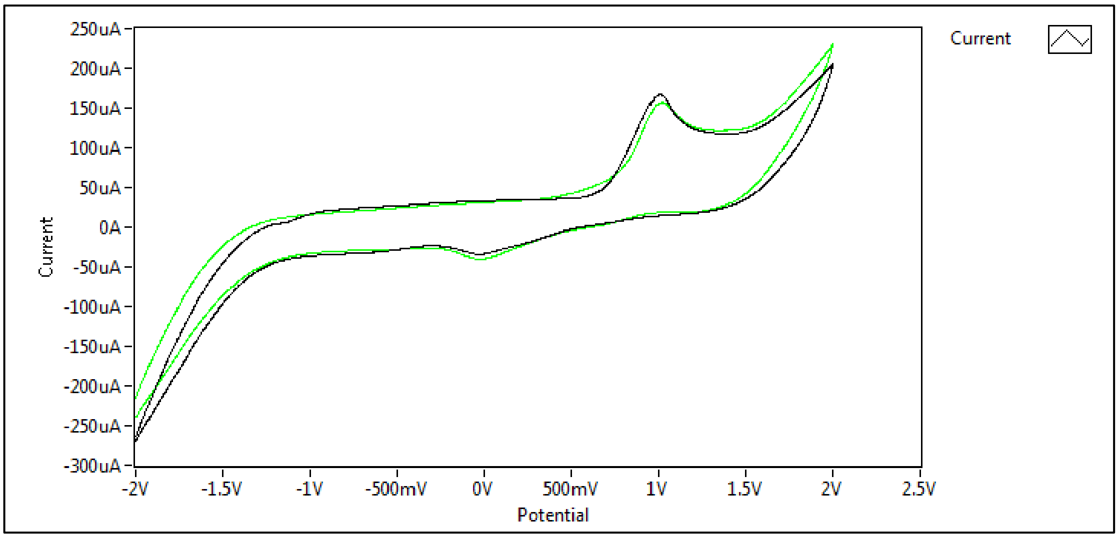

Fig. 15: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 360 Reem (black line with contrast and green line with contrast and AA)

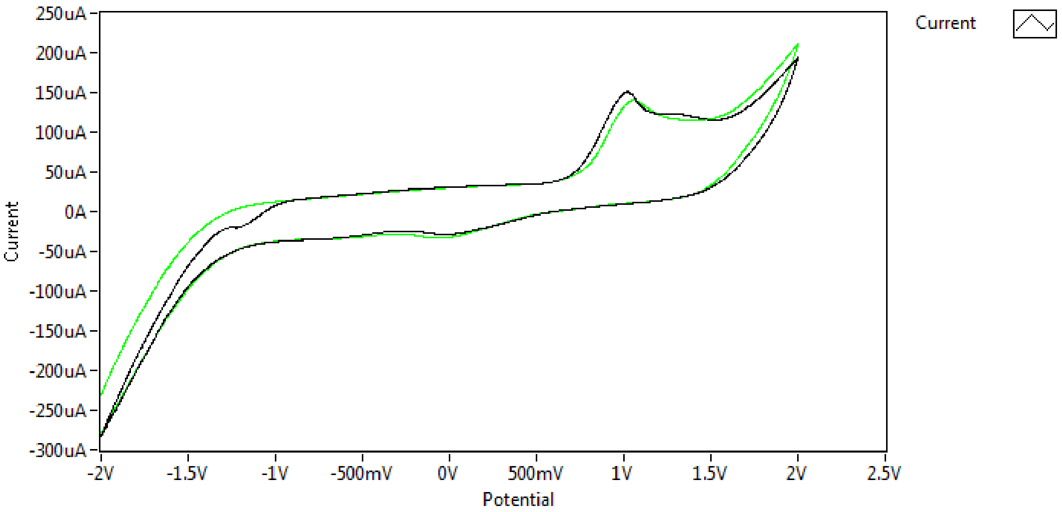

Fig. 16: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 500 Reem (black line with contrast and green line with contrast and AA)

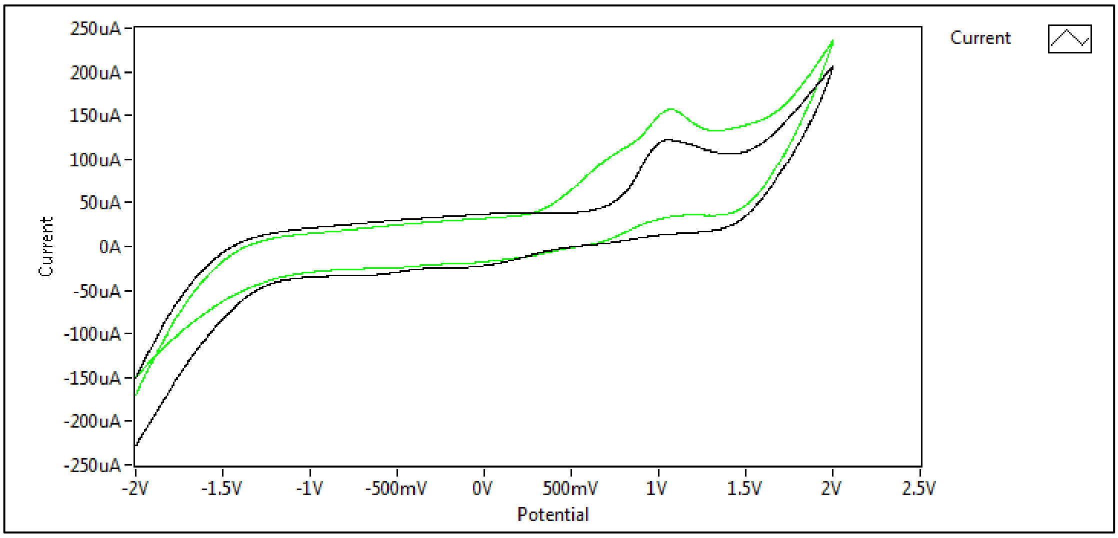

Fig. 17: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 525 Reem (green line with contrast and black line with contrast and AA)

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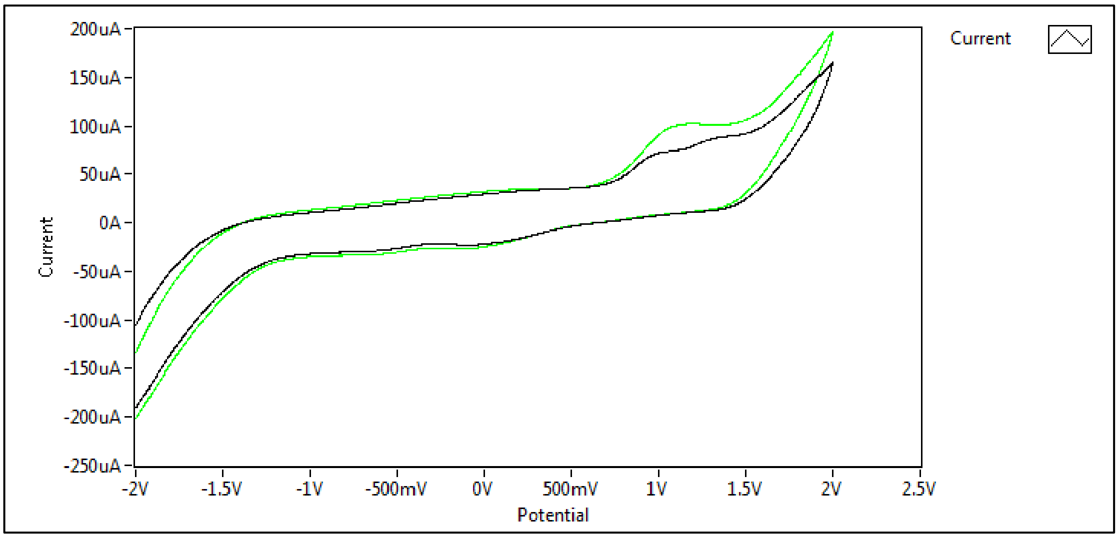

Fig. 18: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 550 Reem (green line with contrast and black line with contrast and AA)

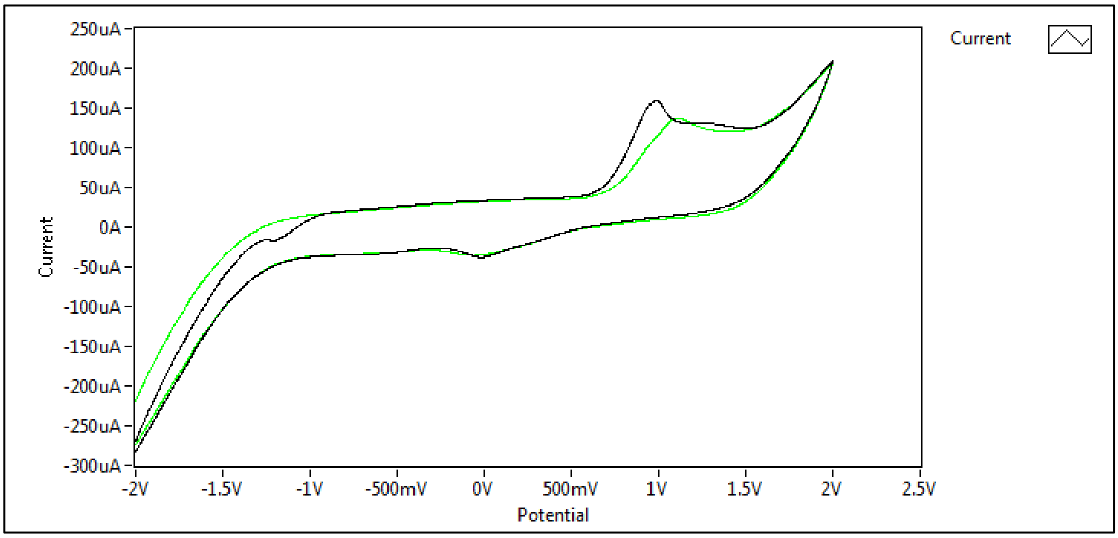

Fig. 19: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 570 Reem (black line with contrast and green line with contrast and AA)

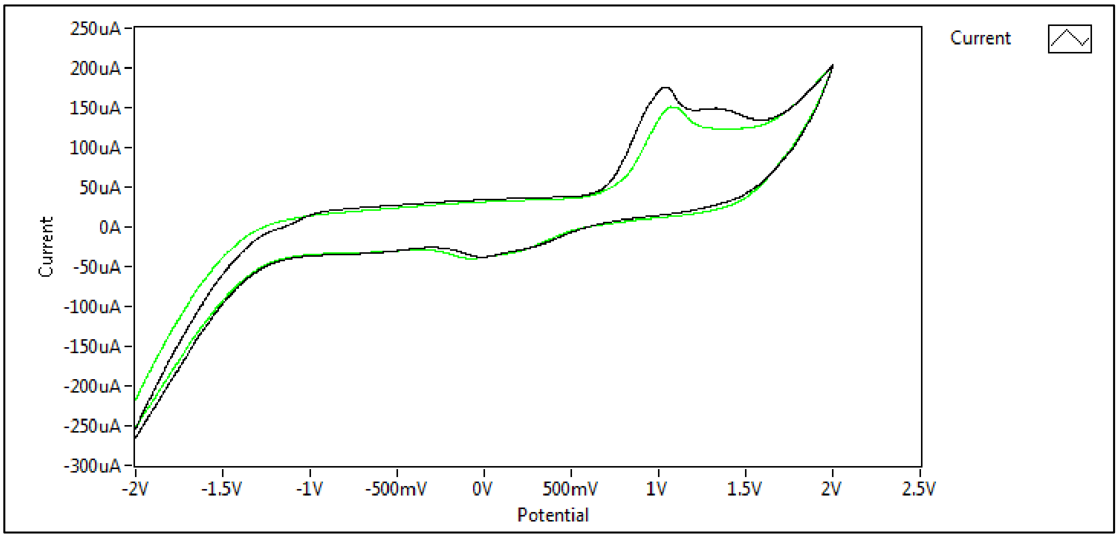

Fig. 20: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 600 Reem (black line with contrast and green line with contrast and AA)

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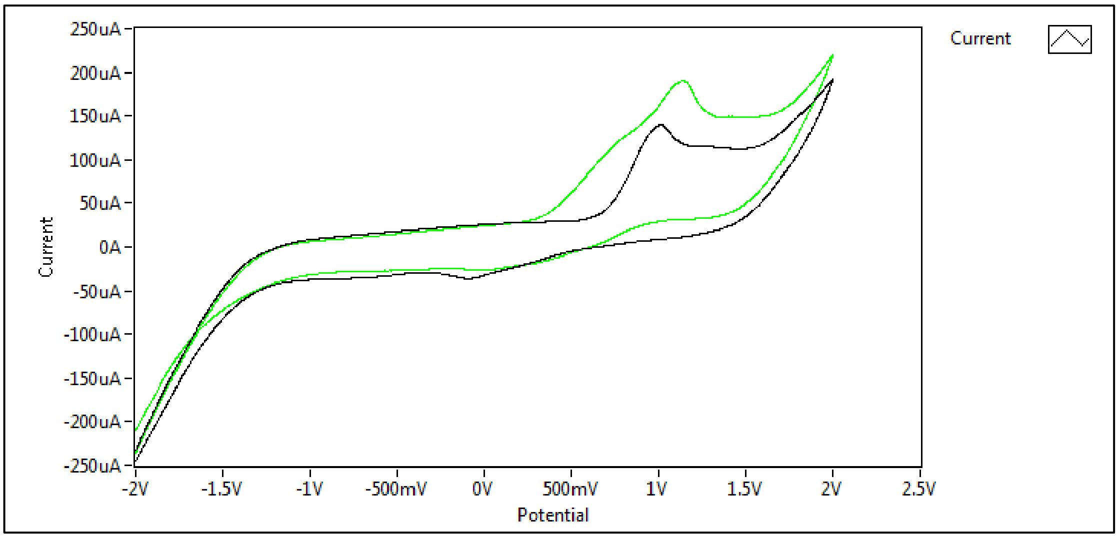

Fig. 21: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 630 Reem (green line with contrast and black line with contrast and AA)

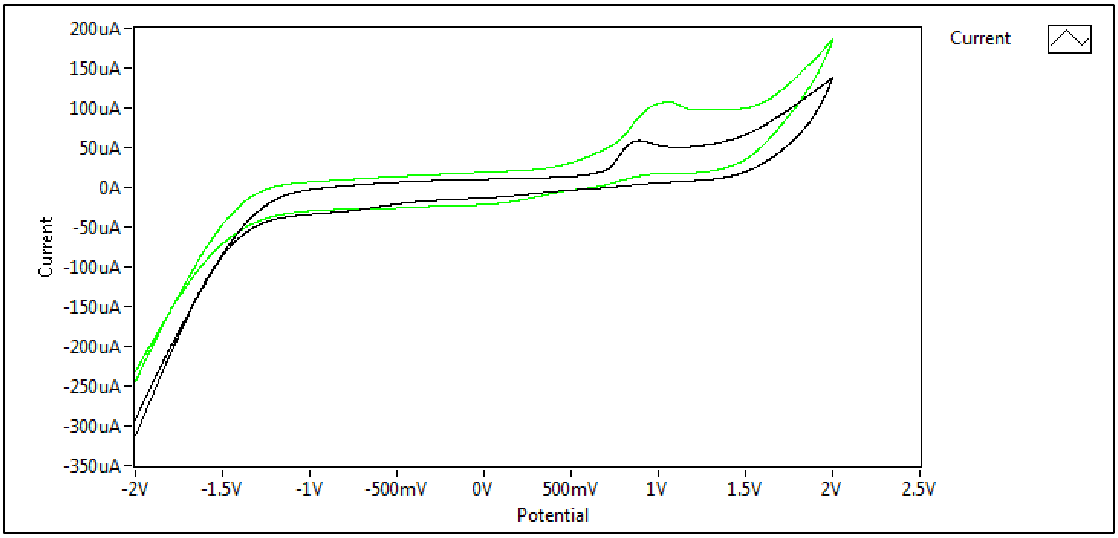

Fig. 22: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 665 Reem (green line with contrast and black line with contrast and AA)

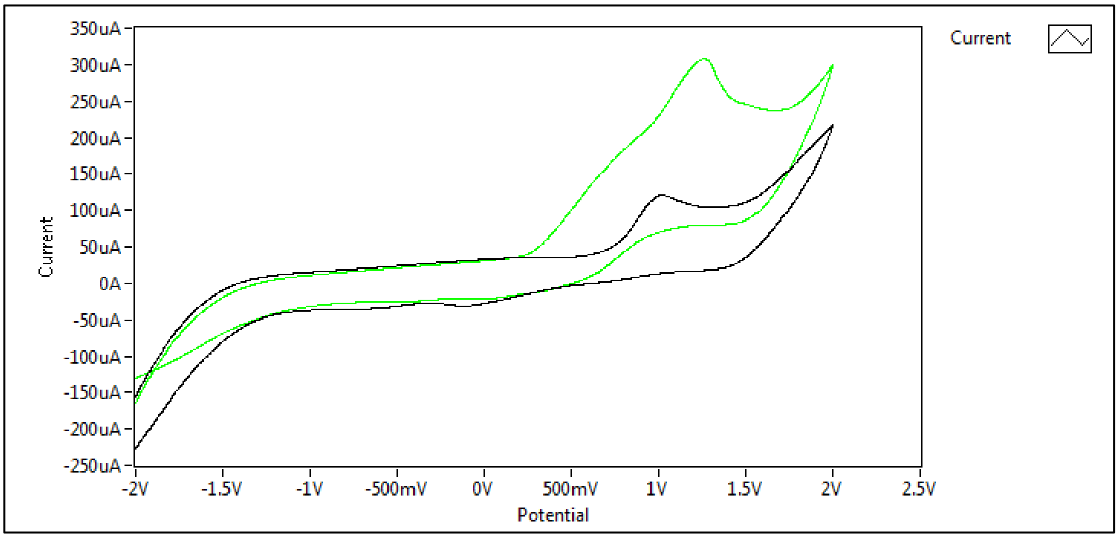

Fig. 23: Cyclic voltammogram of blood sample exposed with X-ray in fluoroscopy dosed 680 Reem (green line with contrast and black line with contrast and AA)

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Fig. 24 Illustrated the relationship between the difference between the oxidation peak current with contrast medium and with contrast medium (Iodine solution) and AA solution as ΔIpa against to the different exposer doses of fluoroscopy; the results of the study was produce a linear line of the relationship with the equation:

$$Y = 0.2299\ X – 83.004 \ldots \text{ with accepted sensitivity } R^2 = 0.727$$

It was found that the contrast medium has an effected in oxidative stress while the AA solution acts as an inhibition of the oxidative effect and is causes by the exposer radiation of X-ray, which has decreased the oxidation through increasing the X-ray doses.

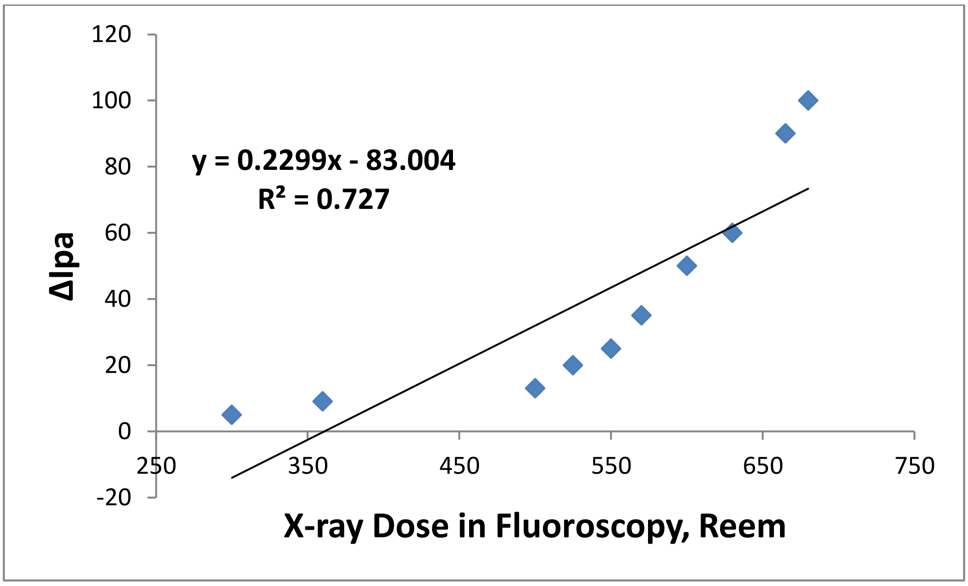

Fig. 24: Relationship between the oxidative difference (ΔIpa) of blood samples with contrast agent (Iodine solution) and with both contrast and AA against to the different doses of x-ray in fluoroscopy (300-685).

CONCLUSION

Through a study in which blood was examined using electrochemical analysis by cyclic voltammetry technique to determine the effect of the patients exposed to X-rays in fluoroscopy with contrast medium (iodine solution) then study the effect of using ascorbic acid with contrast medium, conclude:

1. Using the ionized radiation of fluoroscopy causes oxidative stress to the blood compositions
2. Using the ionized radiation of fluoroscopy for patients injected with contrast medium (iodine solution) causes enhancement of the oxidation process.
3. Using AA solution by injected the patients with a contrast medium and exposed with different doses of X-ray in fluoroscopy has been inhibition the effect of the oxidation process in the blood samples, so using AA acts as an anti-oxidative effect against to ionization radiation.

The advice is for reduce oxidative stress in using the contrast medium in fluoroscopy scanning must be added AA solution to avoid the oxidation reaction with the blood components.

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Source of support: Nil; **Conflict of interest:** Nil.

Cite this article as:

Joudha, L.N., Mustafa, D.A., AbdulJabbar, H.M. and Radhi, M.M. "Assessment Study of the Effect of Using Ascorbic Acid with and Without Contrast Agent on the Blood Samples Exposed with Different Doses of X-Ray in Fluoroscopy by Cyclic Voltammetry." *Sarcouncil journal of Medical sciences* 3.6 (2024): pp 17-32.

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