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Research Article

Analysis of Antioxidant Effects of Quercetin, Rutin and Phagnalon Rupestre on Rats Intoxicated by Aluminium

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Abstract



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Aluminium (Al) is one of the most abundant chemical elements in nature and metal in the earth's crust. Accumulation of Aluminium ions (Al^{3+}) in target tissues results into formation of oxygen radicals causing oxidative damage through inducing cytotoxicity. The aim of this study is to analyse the antioxidant effect of selected polyphenols (quercetin, rutin and a medicinal plant *phagnalon rupestre*) on rats intoxicated by aluminium with specific focus on the heart. This experiment was carried out on 6 groups of wistar albino rats, group 1; positive control, group 2; Al male intoxicated group treated with quercetin, group 3; Al male intoxicated group treated with rutin, group 4; Al female intoxicated, group 5; Al intoxicated treated with *phagnalon rupestre*, group 6; female control. Several Biochemical assays were carried out such as protein test, Thiobarbituric acid reactive substance (TBARS) assay, catalase assay (CAT), glutathione-S-transferase (GST) assay, superoxide dismutase (SOD) assay, Glutathione reduced and glutathione peroxidase (GSH & GPX) assay, lipids (cholesterol & triglycerides) assays. Aluminium intoxicated group showed decrease in the content of protein compared to the control and treated groups. Aluminium intoxicated group showed an increase in the activity of thiobarbituric reactive substances (TBARS) but a significant decrease in the activity of catalase (CAT), glutathione S transferase assays (GST) compared to the control group. Biomarkers of oxidative stress significantly reduced in heart Al-induced oxidative stress by administration of Quercetin. Therefore, Quercetin is an effective antioxidant against oxidative stress caused by free radicals produced because of aluminium exposure.

Keywords: antioxidant ;aluminium ; quercetin ; rutin ; *phagnalon rupestre*

INTRODUCTION

Aluminium (Al) is one of the most widely distributed metal in the environment ¹ This metal was first discovered in 1808 by English chemist Davy² and its physical and chemical properties highlighted by chemist Friedrich Wöhler in 1827³. In nature, aluminium exists in trivalent state (Al^{3+}) as silicates hydroxides and oxides however it can combine with other elements such as chlorine, fluorine, Sulphur and form complexes with organic matters⁴.

Different forms of Aluminium are extensively used in various products and processes associated with human activities for example in crude oil refining and cracking of petroleum; manufacturing of cooking utensils and foils, parchment paper, printing ink, glass, ceramics, pottery, incandescent filaments, fireworks, explosives, photographic flashlight, electric insulators, cement, paints and varnishes, fumigants and pesticides, lubricants, detergents, cosmetics, pharmaceuticals (drugs), vaccines⁵, as well as in water treatment and purification, treating sewage and fur, tanning leather, waterproofing clothes and concretes, industrial filtration, hemodialysis, measuring radiation exposure, in products as flame retardant and fireproofing, anticorrosion agent⁴, food additives to prevent caking as well as components of baking powders and colorants⁶.

Humans are exposed to aluminium particles through several ways such as air (aerosols), drinking water, food, pharmaceuticals and agrochemicals⁷. These exposures influence the Al intake, absorption and elimination which determines the level of tissue accumulation and development of toxicosis⁸. Al intake is mainly through two routes; inhalation (via cigarette smoke, dust from soil and rocks) and ingestion (via food, drugs, water)⁷. Inside the human body, aluminium has been found in the skin, lower gastrointestinal tract, lymph nodes, adrenals, parathyroid gland and in most soft tissues⁹. Absorbed aluminium has several diverse toxic effects arising from its pro-oxidant activities inducing oxidative stress, free radical attack and oxidation of cellular proteins and lipids¹⁰. Al interactions may induce inhibitory or stimulatory effects to extracellular surface and intracellular ligands, inhibition or activation of metabolic or other enzymes¹¹. Exposure of hepatocytes to Al impedes their ATP production, inhibits glycolysis, impairs the function of tricarboxylic acid (Krebs') cycle and promotes lipid and protein oxidation¹². Aluminium exposure induces a disruption in the iron homeostasis resulting to iron overload linked to oxidative stress and pathogenesis of neuro-degenerative disorders¹³. Aluminium ions stimulate apoptosis of several cells such as erythrocytes, osteoblasts through inhibiting apoptotic Bcl-2 protein expression and increasing the expression of pro-apoptotic Bax, Bak and Bim proteins¹⁴. The oxidative stress induced due to aluminium exposure affects the activities of superoxide dismutase (SOD),

catalase (CAT), glutathione peroxidase (GPx) and glutathione (GSH), increases levels of malondialdehyde (MDA) and thiobarbituric acid reactive substances (TBARS) in several tissues¹⁵.

Aluminium overload can be diagnosed and quantified in several biological materials such as blood, bone urine and feces using a variety of analytical methods such as accelerator mass spectroscopy, neutron activation analysis, laser microprobe mass spectrometry and others¹⁶. After diagnosis, several approaches can be employed to treat Al toxicosis such as prevention of intake, reduction of Al absorption, increasing Al excretion, maintaining functional kidneys, reducing load by chelation with chelating agents (deferoxamine, malic acid, citric acid, oxalic acid)¹⁷ and amelioration of the toxic effect with antioxidants (selenium, melatonin, vitamin c, plant extracts¹⁸, quercetin and Ginsenoside Rb1¹⁹).

This study was aimed to explore the in vitro analysis of the redox status by determination of (CAT, GST and TBARS) and analysis of the level of protein in both male and female rats intoxicated by Aluminium with specific focus on the heart. This study further examined the effect of quercetin, rutin and medicinal plant extract on some biomarkers of oxidative stress induced by aluminium exposure in albino Wistar rats.

MATERIAL AND METHOD

Animals and Experimental Design

In this experiment Male and Female Wistar albino rats of varying weight (300-900 ± 10 g and 200 ± 10 g) respectively aged 4–5 months were obtained from the Animal Care Unit of Oran1 University and were kept in cages. The experimental conditions were environmentally controlled in terms of temperature (23 ± 2 °C), humidity (50 ± 5%), and light (12 h of light and dark cycle). The animals were fed with pellet diet with elevated aluminium concentration and water for 30 days.

The rats were grouped into 5 groups of (6) rats.

Group 1: Aluminium male intoxicated group (fed on a conventional rat diet with distilled water containing elevated levels of AlCl₃ 200mg/kg).

Group 2: Aluminium male intoxicated group treated with quercetin (fed on a conventional rat diet with distilled water containing elevated levels of AlCl₃ 200mg/kg and I.p quercetin 100mg/kg).

Group 3: Al male intoxicated group treated with rutin (fed on a conventional rat diet with distilled water containing elevated levels of AlCl₃ 200mg/kg and I.p rutin 100mg/kg).

Group 4: Aluminium female intoxicated group (fed on a conventional rat diet with distilled water containing elevated levels of AlCl₃ 200mg/kg).

Group 5: Aluminium female intoxicated group fed on a conventional rat diet with distilled water containing elevated levels of AlCl₃ 200mg/kg and I.p of plant extract phagnalon rupestre 50mg/kg.

Group 6: Female control group fed on conventional rat food and distilled water.

This experiment was carried out for 30 days. After 30 days the rats were euthanized (sacrificed). After decapitation, whole liver, spleen, kidney, brain, heart and ovary tissues were rapidly resected. The tissues were stored at -80 °C in a deep freezer.

Tissue preparation

The heart tissues were extracted from rats and stored in a deep freezer for preservation. Homogenized by using a Teflon-glass homogenizer with 1.15% KCl to obtain 1:10 (w/v) homogenate

that was later centrifuged and supernatant used for biochemical assays.

Preparation of aluminium chloride solution (AlCl₃)

60mg of AlCl₃ were weighed and dissolved in 1000ml of distilled water to make a solution of 60mg/L supplemented in drinking water daily.

Preparation of quercetin and rutin

15mg of powdered quercetin (95% pure from cell biolabs, Inc) and rutin (95% pure from cell biolabs, inc) were weighed respectively and poured into beakers containing 100ml of distilled water. The solutions were homogenized by stirring and introduced to animals by intraperitoneal (IP) injection at a dose of 100 mg/ kg body weight daily.

Preparation of the plant extract

The genus *Phagnalon* belongs to the Asteraceae family and has a wide range of distribution, expanding from Macaronesia in the West to the Himalayas in the East, from S. France and N. Italy to Ethiopia and Arabian Peninsula. Various species of *Phagnalon* have been used in the popular medicine of several countries as medicinal herbs and food.

The flowering aerial parts of the plant were collected from Misserghin province in Oran Algeria. Identification and authentication were done in the Laboratory of Experimental Biotoxicology, Bio depollution and Phytoremediation of University of Oran1, Ahmed Ben Bella and were deposited at the Herbarium of the Laboratory. The plant extract was got by maceration of leafy stems with methanol, ethanol and water solvents.

BIOCHEMICAL ASSAYS FOR OXIDATIVE STRESS

Protein assay

In this experiment, the protein quantity was determined using Lowry assay protein (Lowry et al 1951).

100µl of homogenized heart tissue was added to 500µl of solution C (Lowry's reagent), vortexed, incubated for 10 mins, then 50µl of solution D (Folin-Ciocalteu reagent). The solution was vortexed again and incubated in the dark for 30 mins. The wavelength reading was taken at 650nm against a blank solution of standard solution of Bovine serum albumin (BSA). The quantity of protein in the sample was determined through extrapolation against a standard curve of BSA (0.2mg/ml).

Thiobarbituric acid reactive substances (TBARS assay)

Malondialdehyde (MDA) concentration of tissue homogenates expressed as the thiobarbituric acid reactive substances (TBARS) was assayed spectrophotometrically according to the method of Placer et al. (1966). The MDA concentrations were expressed as nmol/g protein. In this experiment lipid peroxidation was determined by measuring malondialdehyde (MDA) in the sample homogenate. This was done by adding 100µl of the homogenate (heart tissue) in a glass tube, added 100µl of sodium dodecyl sulfate (SDS 8.1%), then 1.5ml of the acetic acid solution (20%; pH =3.5) and 1.5 ml of TBA (0.8%). The solution was then made up to a total volume of 4ml with distilled water, vortex, incubated at 95°C for 60mins, then cooled in an ice bath for 5 mins and 500µl of distilled water and 2ml of solvent (n-butanol), centrifuged at 4000rpm (rotations per minute) for 10 mins. The wavelength of organic phase is read (greater than 523nm). The TBARS is calculated from.

OD: Optical density read at 532nm

εMDA: Molecular extinction coefficient

Catalase assay

In this experiment, the method is based on the fact that dichromate in acetic acid reduces to chromic acetate when heated in the presence of hydrogen peroxide H₂O₂ with the formation of perchloric acid as an unstable intermediate.

The chromic acetate thus produced is measured calorimetrically at 610 nm. Since dichromate

has no absorbance in this region, the presence of the compound in the assay mixture does not

Interfere with the colorimetric determination of chromic acetate. The catalase preparation is

allowed to split H₂O₂ for different periods of time. The reaction is stopped at specific time

intervals by the addition of dichromate / acetic mixture and the remaining H₂O₂ is determined by measuring chromic acetate calorimetrically after heating the reaction (Sinha,1972).

In this assay, 0.1 ml of homogenate was mixed with 1ml of phosphate buffer (0.001M, pH=7.4) and 0.4 ml of H₂O₂ solution, vortexed then incubated at 37°C for 10 min. The mixture is added to 2 ml of phosphate dichromate solution (K₂Cr₂O₇) at 5% / acetic acid (1v/3v). The reaction was monitored by a spectrophotometer reading at a wavelength of 620 nm and it was expressed in (μmol/mg protein).

Glutathione-s-transferase (GST) assay

In this experiment, assay was used, Solution C was prepared using 20.26mg 1-chloro, 2,4-dinitrobenzene (CDNB) and 153.56mg of glutathione (GSH) diluted into 1ml of ethanol and 100ml of PBS. Then 100μl of solution C was poured into the Elisa plate, added 600μl of the sample. The absorbance of the sample (1.2ml of CDNB + GSH mixture +200μl of supernatant) was measured at absorbance at 340nm each min for five mins against a blank of (1.2ml of CNDB+GSH mixture 200μl of distilled water. The specific activity of GST was calculated from.

$$GST = \frac{\Delta DO \cdot V_t}{e \cdot V_s} \text{ mg of protein}$$

Δ DO: slope of the regression line obtained after hydrolysis of the substrate.

e: Molar extinction coefficient of CDNB = 9.6 mM⁻¹ cm⁻¹.

V_t: Total volume in the tank = 1.4 ml [0.2 ml supernatant + 1.2 ml CDNB / GSH mixture

V_s: Volume of the supernatant in the tank = 0.2 ml.

mg of protein: Quantity of protein expressed in mg

Superoxide dismutase (sod) assay

The SOD activity was performed based on the method by Sun et al. (1988). In this assay, 950μl (Tris-HCl +EDTA) was added to 20μl of homogenate sample and added 50μl of pyragallol. The absorbance was read every after a minute for 5 minutes at 420nm.

GLUTATHIONE REDUCED AND GLUTATHIONE PEROXIDASE (GSH & GPx)

Concentration of GSH of tissue homogenates was measured by an assay using the dithionitrobenzoic acid recycling method by Sedlak and Lindsay (1968).

The GSH-Px activity was determined according to the method of Lawrance and Burk (1976) which records the decrease of NADPH at 340 nm.

GPx Assay

To 100ul of Tris-HCl add 50ul of Sodium Azide then 100ul of supernatant for each specimen then 100ul of GSH then 50ul of H₂O₂. Mix and incubate at body temperature for 10minutes then add 200ul of TCA. Centrifugation at 2400tr/5min. Get the new supernatant and add 200ul of Ellman's Reagent to it. Read the Optical density of each specimen.

LIPIDS (CHOLESTEROL & TRIGLYCERIDES)

In this assay, we used only hearts from male rats divided into 3 categories; positive control (Al intoxicated), Al intoxicated treated with quercetin, Al intoxicated treated with rutin. 0.5g of each specimen was added to 10ml of mixture of chloroform and ethanol (2:1). The mixture was incubated overnight, filtered into flacon tubes. The flacon with and without the lipids were weighed and 3ml of n-xexane to the mixture. The optical density of each specimen was determined using BIOLABO cholesterol and Triglyceride assay kit by cell biolabs.inc. This assay was done by adding 10μl of each specimen to 1ml of Assay reagent (ready for use comes with kit). The blank was prepared by adding 1ml of the kit Assay reagent to 10μl of a standard solution (ready for use comes with kit) of concentration (200mg/dl) from the kit. The concentration of cholesterol of both cholesterol and triglyceride was calculated using the formula below.

concentration =

$$\frac{\text{absorbance of specimen}}{\text{absorbance of standard solution}} \times \text{standard solution (200 mg/dl)}$$

RESULTS

Total proteins.

The protein content results demonstrate a significant difference of (P<0 .05) between the intoxicated (Al) group compared to the control group. It shows decrease of protein in both male and female intoxicated group. The group intoxicated with Aluminium and treated with Quercetin showed a slight increase compared to the control group, group treated with rutin, and group treated with plant. Protein concentration in mg/ml

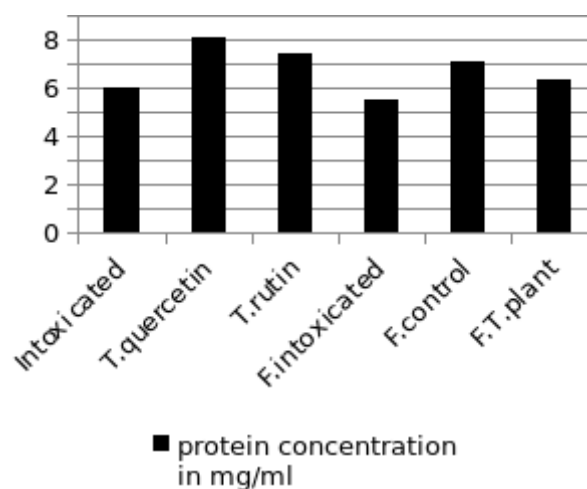


Figure 1: This graph shows the variation of protein activity in the specimen samples.

Catalase

Catalase activity reflected a significant decrease in the groups intoxicated by aluminium compared to the control group, group treated with quercetin, group treated with plant and group treated with plant has showed in graph below

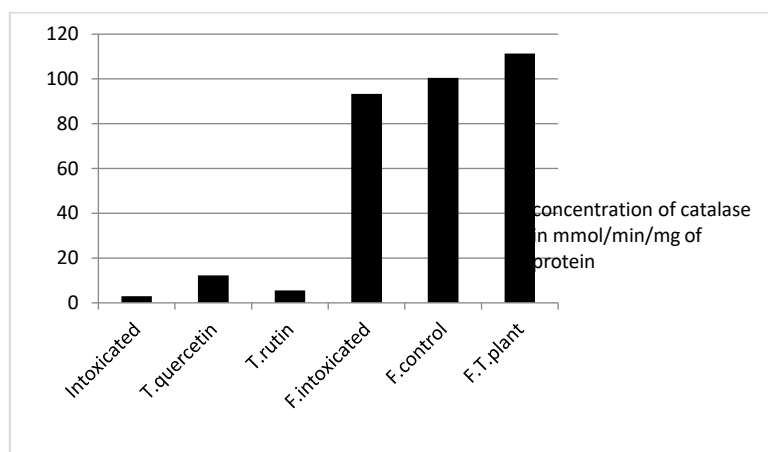


Figure 2. The graph shows the variation of catalase activity in specimen samples.

Values are represented as means $\pm$ SD for each group

TBARS

TBARS activity revealed a significant increase in both male and female Al intoxicated samples compared to the control group; group treated with quercetin has the lowest TBARS activity followed by group treated by rutin and lastly group treated with the medicinal plant.

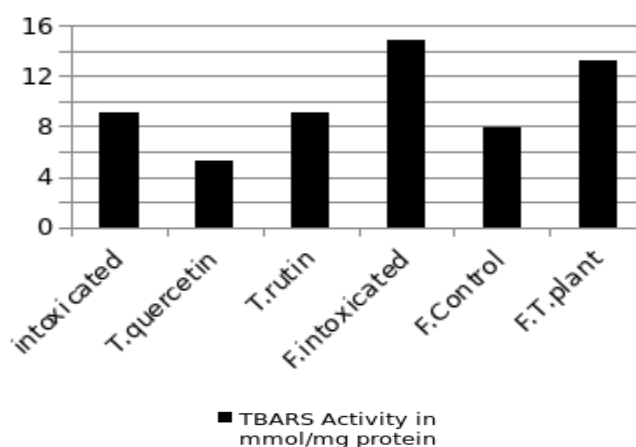


Figure 3: The graph showing variation of TBARS activity in the specimen samples. Values are represented as means $\pm$ SD for each group

GST

GST activity showed a significant decrease in all intoxicated samples compared to the control group, quercetin group, rutin group and group treated with plant.

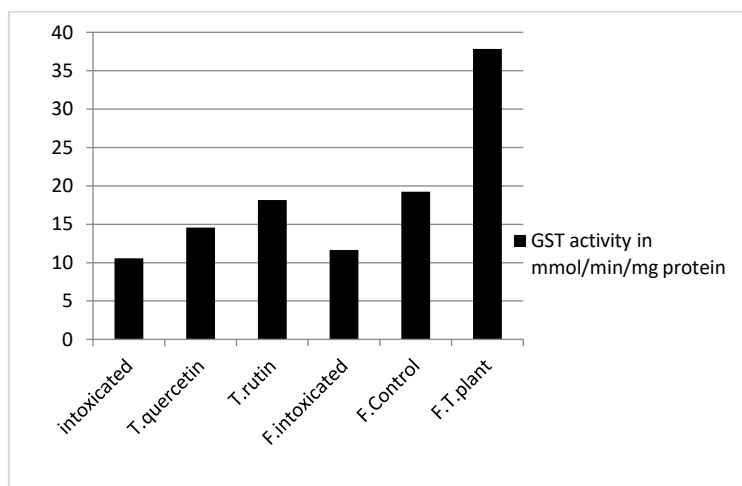


Figure 4: The graph shows the variation of GST activity in the specimen samples.

Values are represented as mean $\pm$ SD for each group.

SOD

SOD % inhibition was most significant in both male and female AI intoxicated groups and more pronounced in females compared to the control group, quercetin group, rutin group and group treated with plant.

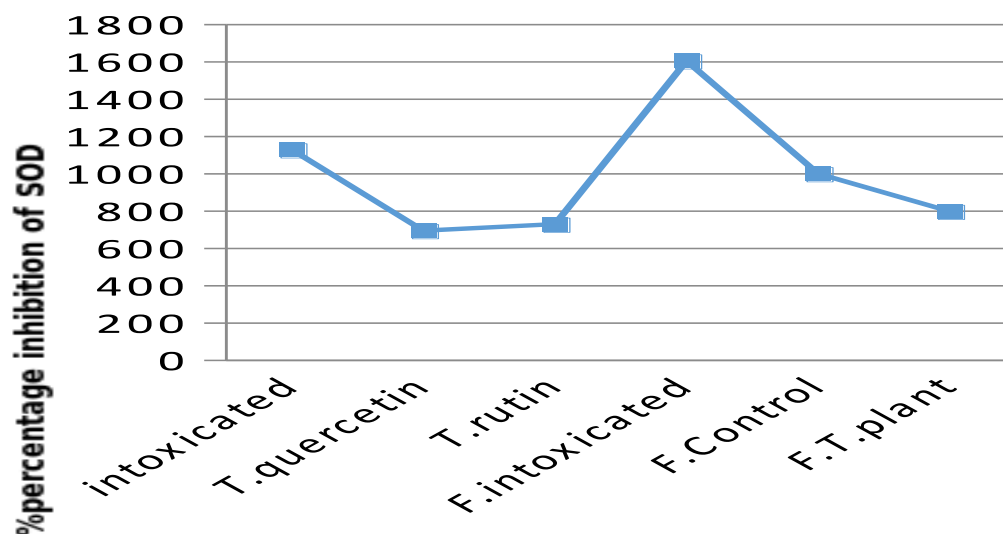


Figure 5: The graph showing the variation of SOD % inhibition in the specimen sample.

values are represented as mean $\pm$ SD for each group.

GSH & GPx

From the results GSH increased drastically with initial acute intoxication but decreased after continuous chronic intoxication while GPx showed an overall increase in activity with increase in intoxication.

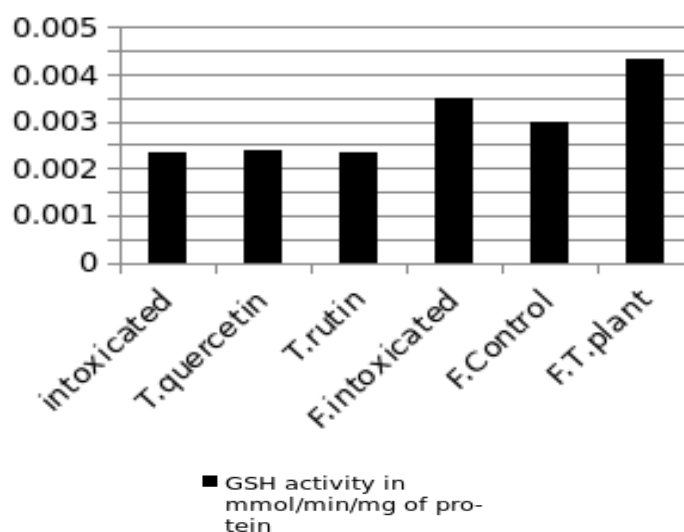


Figure 6: A Graph shows the variation of GSH activity in the specimen sample.

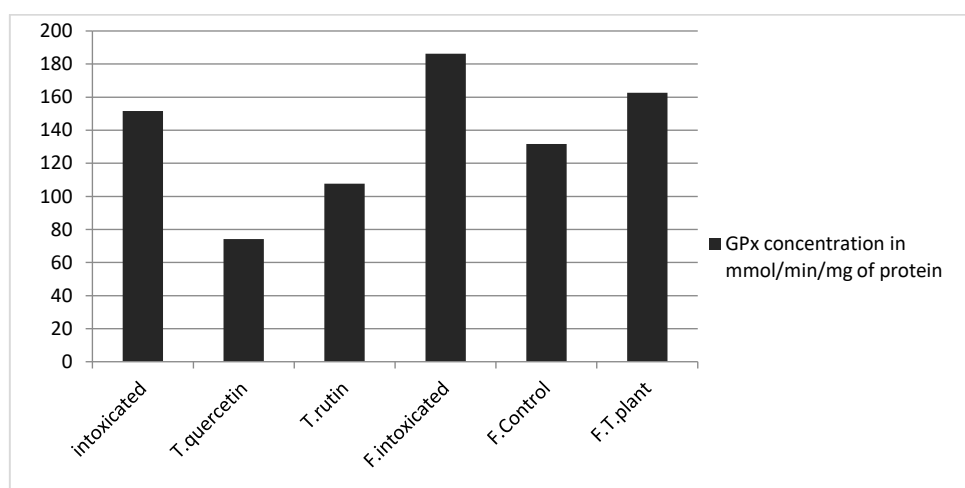


Figure 7: Graph of variation of GPx activity in the specimen samples.

Lipids (Cholesterol & Triglycerides)

From the results obtained it is evident that both Cholesterol and Triglycerides count decreased in Al intoxicated specimen samples and the sample that received treatment with quercetin and rutin showed the higher concentrations in lipids.

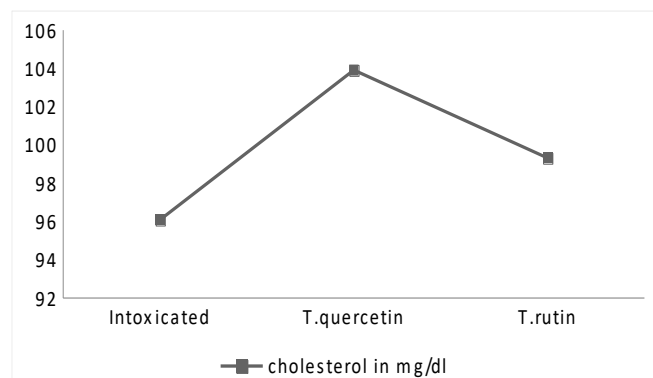


Figure 8: Graph show the variation of cholesterol concentrations in the specimen samples

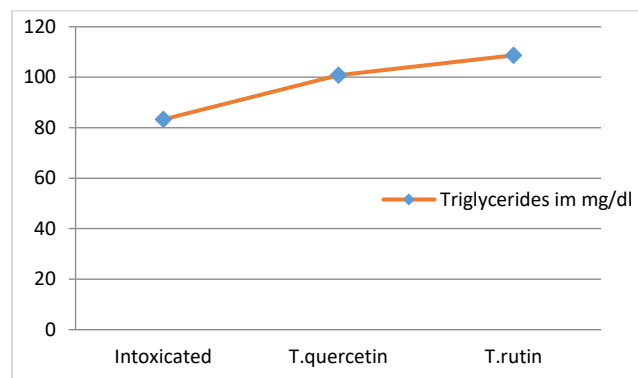


Figure 9: Graph of variation of triglyceride concentration in the specimen samples.

DISCUSSION

This study shows a general decrease in the number of proteins in both rats' male and female intoxicated with Aluminium as compared to the control group and groups treated with quercetin, rutin and the medicinal plant. This may be as a result of reactive oxygen species which oxidate the amino acid residue side chains, protein back bone forming protein fragments thus a reduction in the number of proteins as compared to the control group(20). This further can be explained by the ability of metal ions to affect cellular homeostasis under extreme conditions through interference of the folding process and stimulating aggregation of nascent or non-native protein. The significant increase of proteins in group treated with quercetin agrees with a study (21) that demonstrates the antioxidant effect of quercetin. In this study, there is a significant decrease in the concentration of catalase in both male and female aluminium intoxicated rats with the males being hit harder probably due to a higher metabolism compared to the control group, this may be as a result of increase of O_2^- that gradually inactivate catalase(22). The results show a decrease in GST concentrations in aluminium intoxicated groups, this may be due to the inhibition of GST activity by aluminium as seen in the similar study of (23). The high GST activity of group treated with plants may be due to the chelation or free radical scavenging effect of bioactive molecules in *phagnalon rupestre*. The TBARS results show an increase in the concentration of MDA in both aluminium intoxicated groups with the female group having the highest compared to the control group, this agrees to with results from similar studies of (15),(24),(25). The low concentration of MDA in quercetin treated groups compared to other groups shows its ameliorative effect compared to rutin and phagnalon rupestre. The intoxicated female and male groups have high percentages of SOD inhibition that further proves that aluminium has a pro-oxidant effect through enzyme inhibition while treatment with quercetin, rutin, *phagnalon rupestre* (medicinal plant) shows their ability to reverse the inhibition activities of aluminium. GSH is a co-substrate for GPx. The initial increase in GSH activity can be explained by the enzyme being the first line of defense against pro-oxidants and its ability to act as a coenzyme but with increase in intoxication the enzyme is inhibited. GPx increased in intoxicated samples probably due to its high affinity for H_2O_2 as compared to catalase(26).

Triglycerides levels decreased significantly in the specimen samples intoxicated with Al compared to the control group, group treated with plant, quercetin and rutin antioxidants. The reduction in lipids may be attributed to oxidative stress

resulting into lipid peroxidation, phosphine toxicity and reduced lipase activity due to Al toxicosis(27).

CONCLUSION

The redox mechanism involves two opposite molecular mechanisms, the production of oxidants and antioxidants. An equilibrium between these two mechanisms is a delicate task since there is continued production of free radicals from both endogenous and exogenous sources. Body organs such as the heart maintain this equilibrium in vivo by continuous production of cells to mop out the excess free radicals through defense mechanisms like apoptosis. This study shows that the maintenance of this balance is limited with time and increase in free radicals, organisms lose this battle against free radicals generated by aluminium intoxication. In this case antioxidants like vitamins, medicinal plants, polyphenols (quercetin, rutin) proved to provide a relief against oxidative stress caused by Aluminium toxicosis. This study shows that Quercetin is a better antioxidant than Rutin against Aluminium intoxication. The protective properties of Quercetin were even better than the medicinal plant (*Phagnalon rupestre*). The results also show that the plant had some protective abilities against the free radicals. The study also shows that female rats were more affected by the toxicosis than male rats. We recommend future studies to analyse the effect of aluminium on lipid membrane phospholipids and studies to analyse why female rats have higher aluminum effects than male rats.

Ethical consideration

This study was protocol was approved by the University of Oran 1's Scientific Committee.

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Author's contribution

Yiga Henry junior conceived, designed the study, and acquired the data. Yiga Henry junior and Mulondo Samuel analyzed, interpreted the data, drafted, and revised the article.

Conflict of interest

The authors declare that they have no conflicts of interest with the contents of this article.

REFERENCES

1. The Commonwealth Scientific and Industrial Research Organisation. *Curr Biol*. 1997 Mar;7(3):R126. [https://doi.org/10.1016/S0960-9822\(97\)70976-X](https://doi.org/10.1016/S0960-9822(97)70976-X)
2. Kvande H. Two hundred years of aluminum ... or is it aluminium? *JOM*. 2008 Aug;60(8):23-4. <https://doi.org/10.1007/s11837-008-0102-3>
3. Rabinovich D. The allure of aluminium. *Nat Chem*. 2013 Jan;5(1):76-76. <https://doi.org/10.1038/nchem.1535> PMID:23247182
4. Gourier-Fréry C, Fréry N. Aluminium. *EMC - Toxicol-Pathol*. 2004 Jul;1(3):79-95. <https://doi.org/10.1016/j.emctp.2004.04.002>
5. Tomljenovic L, A. Shaw C. Aluminum Vaccine Adjuvants: Are they Safe? *Curr Med Chem*. 2011 Jun 1;18(17):2630-7. <https://doi.org/10.2174/092986711795933740> PMID:21568886
6. Stahl T, Falk S, Rohrbeck A, Georgii S, Herzog C, Wiegand A, et al. Migration of aluminum from food contact materials to food-a health risk for consumers? Part I of III: exposure to aluminum, release of aluminum, tolerable weekly intake (TWI), toxicological effects of aluminum, study design, and methods. *Environ Sci Eur*. 2017 Dec;29(1):19. <https://doi.org/10.1186/s12302-017-0116-y> PMID:28458989 PMCID:PMC5388732
7. Igbokwe IO, Igwenagu E, Igbokwe NA. Aluminium toxicosis: a review of toxic actions and effects. 2019; <https://doi.org/10.2478/intox-2019-0007> PMID:32206026 PMCID:PMC7071840
8. Yokel RA, McNamara PJ. Aluminium Toxicokinetics: An Updated MiniReview: ALUMINIUM TOXICOKINETICS. *Pharmacol Toxicol*. 2008 Jul 7;88(4):159-67. <https://doi.org/10.1034/j.1600-0773.2001.d01-98.x> PMID:11322172
9. Rahimzadeh MR, Rahimzadeh MR, Kazemi S, Amiri RJ, Pirzadeh M, Moghadamnia AA. Aluminum Poisoning with Emphasis on Its Mechanism and Treatment of Intoxication. *Fedeli P, editor. Emerg Med Int*. 2022 Jan 11;2022:1-13. <https://doi.org/10.1155/2022/1480553> PMID:35070453 PMCID:PMC8767391
10. Exley C. Human exposure to aluminium. *Env Sci Process Impacts*. 2013;15(10):1807-16. <https://doi.org/10.1039/C3EM00374D> PMID:23982047
11. Kawahara M, Konoha K, Nagata T, Sadakane Y. Aluminum and Human Health : Its Intake, Bioavailability and Neurotoxicity.
12. Mailloux RJ, Hamel R, Appanna VD. Aluminum toxicity elicits a dysfunctional TCA cycle and succinate accumulation in hepatocytes. *J Biochem Mol Toxicol*. 2006 Aug;20(4):198-208. <https://doi.org/10.1002/jbt.20137> PMID:16906525
13. Milton NGN. Role of Hydrogen Peroxide in the Aetiology of Alzheimer's Disease: Implications for Treatment. *Drugs Aging*. 2004;21(2):81-100. <https://doi.org/10.2165/00002512-200421020-00002> PMID:14960126
14. Xu F, Ren L, Song M, Shao B, Han Y, Cao Z, et al. Fas- and Mitochondria-Mediated Signaling Pathway Involved in Osteoblast Apoptosis Induced by AlCl₃. *Biol Trace Elem Res*. 2018 Jul;184(1):173-85. <https://doi.org/10.1007/s12011-017-1176-y> PMID:29027106
15. Yu H, Zhang J, Ji Q, Yu K, Wang P, Song M, et al. Melatonin alleviates aluminium chloride-induced immunotoxicity by inhibiting oxidative stress and apoptosis associated with the activation of Nrf2 signaling pathway. *Ecotoxicol Environ Saf*. 2019 May;173:131-41. <https://doi.org/10.1016/j.ecoenv.2019.01.095> PMID:30771656
16. San Martín SP, Bauçà JM, Martínez-Morillo E. Determination of aluminum concentrations in biological specimens: application in the clinical laboratory. *Adv Lab Med Av En Med Lab*. 2022 Jun 30;3(2):153-9. <https://doi.org/10.1515/almed-2022-0056> PMID:37361866 PMCID:PMC10197291
17. Domingo JL, Gómez M, Llobet JM, Corbella J. Comparative Effects of Several Chelating Agents on the Toxicity, Distribution and Excretion of Aluminium. *Hum Toxicol*. 1988 May;7(3):259-62. <https://doi.org/10.1177/096032718800700305> PMID:3391623
18. Yan L, Riaz M, Liu J, Yu M, Cuncang J. The aluminum tolerance and detoxification mechanisms in plants; recent advances and prospects. *Crit Rev Environ Sci Technol*. 2022 May 3;52(9):1491-527. <https://doi.org/10.1080/10643389.2020.1859306>
19. Ye J, Wei D, Peng L, Chang T. Ginsenoside-Rb1 prevents steroid-induced avascular necrosis of the femoral head through the bone morphogenetic protein-2 and vascular endothelial growth factor pathway. *Mol Med Rep [Internet]*. 2019 Aug 6 [cited 2023 Jul 10]; Available from: <http://www.spandidos-publications.com/10.3892/mmr.2019.10553> <https://doi.org/10.3892/mmr.2019.10553>
20. Davies MJ. Protein oxidation and peroxidation. *Biochem J*. 2016 Apr 1;473(7):805-25. <https://doi.org/10.1042/BJ20151227> PMCID:PMC4819570
21. Marunaka Y, Marunaka R, Sun H, Yamamoto T, Kanamura N, Inui T, et al. Actions of Quercetin, a Polyphenol, on Blood Pressure. *Molecules*. 2017 Jan 29;22(2):209. <https://doi.org/10.3390/molecules22020209> PMID:28146071 PMCID:PMC6155806
22. Bauer G. Increasing the endogenous NO level causes catalase inactivation and reactivation of intercellular apoptosis signaling specifically in tumor cells. *Redox Biol*. 2015 Dec;6:353-71. <https://doi.org/10.1016/j.redox.2015.07.017> PMID:26342455 PMCID:PMC4564397
23. Röth E, Marczin N, Balatonyi B, Ghosh S, Kovács V, Alotti N, et al. Effect of a glutathione S-transferase inhibitor on oxidative stress and ischemia-reperfusion-induced apoptotic signalling of cultured cardiomyocytes. 2011;16(3).
24. Zhang Q, Cao Z, Sun X, Zuang C, Huang W, Li Y. Aluminum Trichloride Induces Hypertension and Disturbs the Function of Erythrocyte Membrane in Male Rats. *Biol Trace Elem Res*. 2016 May;171(1):116-23. <https://doi.org/10.1007/s12011-015-0504-3> PMID:26354416
25. Tsikas D. Assessment of lipid peroxidation by measuring malondialdehyde (MDA) and relatives in biological samples: Analytical and biological challenges. *Anal Biochem*. 2017 May;524:13-30. <https://doi.org/10.1016/j.ab.2016.10.021> PMID:27789233
26. Baud O, Greene AE, Li J, Wang H, Volpe JJ, Rosenberg PA. Glutathione Peroxidase-Catalase Cooperativity Is Required for Resistance to Hydrogen Peroxide by Mature Rat Oligodendrocytes. *J Neurosci*. 2004 Feb 18;24(7):1531-40. <https://doi.org/10.1523/JNEUROSCI.3989-03.2004> PMID:14973232 PMCID:PMC6730475
27. Ferretti G, Marchionni C, Bacchetti T, Galeazzi T, Dousset N. Effect of Aluminium on Lipid Peroxidation of Human High Density Lipoproteins. *Free Radic Res*. 2003 Jan;37(5):515-21. <https://doi.org/10.1080/1071576031000083125> PMID:12797472