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The Protective effect of Pomegranate and Red Beetroot Juices against Aluminum Toxicity in Brains of Male Rats

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ABSTRACT

The purpose of this work is to investigate the protective effect of pomegranate, red beetroot juices or their mixture on the brain toxicity induced by $AlCl_3$ in rats. Five groups (7 rats each) were used in the experiment; one of them continued feeding on basal diet only during the period of experiment (normal control). Another group was injected daily by $AlCl_3$ beginning from the 14th day and served as positive control while the remaining three groups were treated by pomegranate, red beetroot and their mixture juices from the first day till the end of the experiment and from the 14th day, they were injected by $AlCl_3$ until the end of the experiment. Phenolic compounds, flavonoids, vitamin C and DPPH scavenging activity of pomegranate and red beetroot juices were estimated. The results revealed that the positive group exhibited low levels of growth parameters, hemoglobin, iron and high level of aluminum as compared to normal control, while the treated groups revealed significant improvements in these parameters. Concerning antioxidant parameters, the three treated groups showed significant increases in superoxide dismutase, glutathione and total protein and significant decreases in malondialdehyde and acetylcholine esterase in their serum and brains as compared to positive control. However, the β-amyloid level decreased while RNA and DNA contents increased significantly in brains of the treated rats in comparing with the positive control. The mixture juice exhibited the best protective effect among the three juices against aluminum toxicity, so it is important for those who suffering from aluminum toxicity.

Keywords: Pomegranate, beetroot, aluminum toxicity, antioxidants, brain.

INTRODUCTION

This work was designed in a trial to minimize the effect of aluminum toxicity in brains of the experimental rats. Aluminum is used in keeping foods either during processing or after cooking and is also added to drinking water for purification purposes (Newairy *et al*., 2009). It is widely used in canning and packing foods such as beverages, juices and milk drinks. It is also added to some medications such as antacids and toothpaste (Abbasali *et al*., 2005), as well as its production as a result of environmental pollution from industry especially aluminum factories which expose the workers and the people to high levels more than the permissible levels. The brain is one of the organs most vulnerable to oxidative stress, due to several reasons, including high oxygen consumption, low mild division rate, and high fat content. Stevanović *et al*. (2009) have shown that aluminum toxicity caused brain cell damage as a result of producing free radicals. Lin et al. (2015) reported that oral administration of aluminum chloride with a dose of 100 mg/ kg b.w. daily for 42 days caused brain toxicity, or via consuming aluminum chloride in a dose of 17 mg /kg/ day orally for four consecutive weeks (Mahdy *et al*., 2014 and Ghoneim *et al*., 2015). Neurotoxicity resulting from the ingestion of aluminum can be attributed to several mechanisms, involving oxidative brain injury and damage to neurons as well as inflammatory reaction of neurons and reduce the biosynthesis of neurotransmitters (Zatta *et al*., 2003). Aluminum also led to many changes in biochemical indicators, reduced the activities of the antioxidant enzymes and upraise lipid peroxidation in rabbit brain and rat plasma (Yousef, 2004 and Newairy *et al*., 2009).

As it is well known, fresh fruits and vegetables are considered very important in improving human health due to their high content of useful phytochemicals and other micronutrients (Opara and Al-Ani, 2010). The importance of these compounds is attributed to their antioxidant activity which can scavenge the free radicals that cause oxidative stress which can lead to cellular damage and many degenerative disorders (Boyer and Leo, 2004).

Pomegranate (*Punica granatum* L) juice is a rich source of antioxidants because it contains phenolic components that possess antioxidant activity. Previous studies have shown that consuming pomegranate juice reduced oxidative stress by increasing the level of internal antioxidants, protecting cells from damage caused by free radicals by increasing resistance to oxidative stress (Li *et al*., 2006 and Bachoual *et al*., 2011 and Mahmoud and Abdel Moneim, 2013).

Beetroot (*Beta vulgaris L*) is planted biennially from the Chenopodiaceae family and contains many varieties in bulb colors ranging from yellow to red. Deep red beetroot is

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the most popular for human consumption, whether cooked or raw as salad or juice. Red beet (*Beta vulgaris* L.) is grown worldwide for its roots that are utilized in food and as a source of natural dye. Beetroot is a cure for a range of diseases associated with oxidative stress and inflammation because it contains many active substances, especially betalain pigments, which has an antioxidant and anti-inflammatory activity, whether internally or externally. It has been also noted that beetroot supplements may provide health benefits; this may be due to its ability to improve cognitive function, vascular function, cancer, inflammation, insulin resistance and pain relief especially in patients (Clifford *et al*., 2015). Beetroot improves blood flow through the body, inclusively the brain, muscles and heart because of its content of nitrates. Nitric oxide helps open vessels and allows more oxygen flow; it also reduces blood pressure and diminishes cardiovascular disease (Kenjale and Ham,2011).

Therefore, this study was carried out to investigate the protective effect of pomegranate juice and red beetroot juice on the toxicity of aluminum in brain of experimental rats in attempt to diminish the toxicity of aluminum produced among the workers of aluminum manufactures and as a result of using aluminum in canning and preserving foods and drinks as well as in Alzheimer patients.

MATERIALS AND METHODS

Materials

Raw materials: Pomegranate (*Punica granatum* L) and red beetroot (*Beta vulgaris* L) were obtained from local market in Mansoura City.

Chemicals: All the used kits and chemicals of analytical grade were purchased from Al-Gomhorya Company for Trading Medicines and Medical equipment, Mansoura, Egypt.

Diet: A standard diet of fine ingredients was prepared according to Reeves *et al*. (1993).

Animals: Thirty-five adult male albino rats (weighing 130 to 140 g), were purchased from Helwan farm of experimental animals, Cairo, Egypt. (Guidelines for ethical conduct in the care and use of animals in research were obtained from the concerned department in Mansoura University).

Methods

Pomegranate juice preparation: Fruits were washed manually, peeled and their separated seeds were blended in a blender to obtain their extract which subjected twice to filtrations through a piece of clothes to obtain the fresh juice which has been prepared daily.

Preparation of red beetroot Juice: The beetroots were washed with tap water, cut into small pieces. The juice of the freshly chopped beetroots was prepared by household juice extractor and then filtered through a stainless-steel strainer by sterile gauze, to obtain the fresh juice which has been prepared daily.

Experimental design: Thirty-five male rats were used in this experiment; they were housed in metallic cages under healthy environmental conditions for acclimatization. Water and diet were provided *ad-libitum*. They were divided into 5 groups (7 rats each), one of them remained on the basal diet only which served as normal control. The rats of another group were injected interperitoneally by AlCl3 in a dose of 20 mg/kg b.w. (dissolved in normal saline) to induce aluminum toxicity beginning from the 14th day until the end of the experiment; this is the positive group. The remaining three groups were treated separately by the juices of pomegranate, red beetroots and their mixture (1:1 v/v) in a dose of 15 ml/kg b.w. by means of stomach tube from the first day to the end of the experiment (6 weeks). On the 14th day of the experiment, the three treated groups were injected interperitoneally by AlCl3 in a dose of 20 mg/kg b.w. daily until the end of the experiment as described by Berlyne *et al*. (1972). At the end of the experiment, all the rats were sacrificed, and their blood was drawn from the portal vein and their plasma were separated as well as their brains were isolated, washed in normal saline, dried and kept in refrigerator for further analysis.

Gross chemical composition of pomegranate and red beetroot juices:

Estimation of moisture, total protein, total ash, dietary fiber and ascorbic acid were carried out as mentioned in the procedures of AOAC (2012). Total carbohydrates were estimated by difference.

Determination of flavonoids and phenolic content of pomegranate and red beetroot juices:

Total flavonoid content was determined according to the method used by Zilic *et al*. (2011) which expressed as mg of catechin equivalent (CE) per g of sample .Total phenolic content was determined according to the Folin-Ciocalteu procedure and expressed as mg of gallic acid equivalent (GAE) per g of sample (Singleton *et al*.,1999).

Antioxidant activity:

Determination of radical DPPH scavenging activity was determined using the stable 1, 1-Diphenyl-2-picryl-hydrazyl (DPPH) according to Hwang and Do Thi (2014).

Biological assays:

At the end of the experiment, evaluation of the tested diets was carried out by determining total feed intake (FI) and body weight gain % (BWG%) according to Chapman *et al*. (1959).

Determination of antioxidant biomarkers of both serum and brain tissue:

Glutathione (GSH) was determined according to the method described by Beutler *et al*. (1963) while Superoxide dismutase (SOD) was estimated according to the method of Nishikimi *et al*. (1972). Malondialdehyde (MDA) was assessed as described by Ohkawa *et al*. (1979). ***Acetycholinesterase (AchE) activity*:** It was determined according to the method of Knedel and Boottger (1967) .

Determination of blood hemoglobin:

Blood hemoglobin was estimated according to the method of Drabkin (1949).

Determination of aluminum and iron in serum:

Aluminum (Al) and iron (Fe) were assayed as described by Toda *et al*. (1980) and Perrotta (1984), respectively.

Determination of β- amyloid in brain tissue:

Determination of β- amyloid (Beta-Mark Beta-Amyloid x-40 Chemiluminescent ELISA kit) was carried out in rats' brain as described by Portelius *et al*. (2006).

Determination of total protein, DNA and RNA in brain tissues:

Determination of DNA, RNA and total protein in brain tissues were estimated according to the method described by Shibko *et al* (1967). Total protein was extracted

from brain homogenate and determined calorimetrically using BioMerieux kit.

Statistical analysis: The results were statically analyzed by using computer program (SPSS); one-way analysis of variance (ANOVA) was carried out, the difference was considered significant at P-value < 0.05.

RESULTS AND DISCUSSION

Proximate chemical composition of pomegranate and red beetroot juices:

Data recorded in Table 1 indicated that proximate chemical composition of pomegranate and red beetroot juices were as follows; moisture (85.55 and 88.1 g/ 100ml), protein (0.14 and 1.20 g/ 100ml), fats (0.04 and 0.25 g/ 100ml), ash (0.33 and 1.31 g/ 100ml), dietary fiber (2.79 and 2.01) and total carbohydrates (11.15 and 7.13 g/100 ml), respectively. These results are similar to those of Kale *et al.* (2018) who reported that red beetroot juice contains 87.4, 1.35, 0.3, 1.9, 1.4 and 7.59 % for moisture, protein, fat, dietary fiber, ash and total carbohydrates respectively. Chemical composition of pomegranate juice was comparable with the values reported by Khedr *et al.* (2015) who stated the values of 0.207 g/100 ml for crude protein, 0.0448 g/100 ml for total fat , 0.379 g/100 ml for ash, and 3.38 g/1090 ml for total carbohydrates .

Table 1. Proximate chemical composition of pomegranate and red beetroot juices.

Components / Juices	Moisture	Protein	Fat	Ash	Dietary fiber	Carbohydrate
Pomegranate	85.55	0.14	0.04	0.33	2.79	11.15
Red Beetroot	88.1	1.20	0.25	1.31	2.01	7.13

Total phenolics, flavonoids, vitamin C contents and antioxidant activity of pomegranate and red beetroot juices.

The data recorded in Table 2 stated that the total phenolic compounds of the pomegranate and red beetroot juices were 750 and 985 mg/100ml as GAE while the total flavonoids contents were 12.1 and 44mg /100ml as CE and their content of vitamin C were 6.09 and 6.42 mg /100ml, respectively. These moderate values of antioxidants of the two juices make them of great benefits in fighting much toxicity especially those caused by chemicals and heavy metals. The benefits of these antioxidants reflected in the free radical scavenging as determined by DPPH scavenging activity which reached 47.38% for pomegranate juice and 62.19% for red beetroot juice. These results agreed with the findings of Arora *et al.* (2019) who reported that the antioxidant activity of beetroot juice which represented in the scavenging activity of DPPH radical was 62.20% and ascorbic acid content (6.44 mg/100 g). The results of this study were in same line with the previous findings of Venkatachalam *et al.* (2014) who found that the total phenolic compounds of red beetroot were 57.64 mg/100 g. Esatbeyoglu *et al.* (2014) stated that the antioxidant activity of red beetroot is attributed to its content of phenolic compound, flavonoids and its red pigment betanin which has a radical-scavenging efficacy.

Table 2. Total phenolics, flavonoids, vitamin C contents and antioxidant activity of pomegranate and red beetroot juices.

Values / Juices	Total Phenolic (GAE mg/100ml)	Total Flavonoids (CE mg/100ml)	Vitamin C mg/100mL	Antioxidant activity (%)
Pomegranate	780	12.1	6.09	47.38
Red Beetroot	985	44	6.42	62.19

Effect of pomegranate, red beetroot juices or their mixture on growth parameters, hemoglobin (Hb) and serum Iron (Fe) and Aluminum (Al) of the rats affected by aluminum toxicity.

Data in Table 3 describes the influence of pomegranate, beetroot juices or their mixture on both growth parameters and levels of blood hemoglobin and serum iron and aluminum. The positive control which received $AlCl_3$ only showed significant decreases in all the mentioned parameters except serum aluminum which increased significantly in comparing with normal control. The decreases percentages reached 58% in body weight, 26% in feed intake, 31.5% in serum hemoglobin and 61% in serum Fe while the increase in serum Al content was very high.

On the other hand, the results revealed that the treated groups showed significant improvements in all the determined parameters recorded in table three. Regarding body weight and feed intake, the groups of pomegranate and beetroot have nearly the same effect while their mixture group exhibited the highest increase which was like that of the normal rats (negative control). The blood level of hemoglobin has been improved in the treated groups; the increase caused by beet root was more than that of pomegranate while their mixture showed the best increase in this respect which was as that of the normal control. Manthou *et al.* (2017) reported that the phytochemicals include in Pomegranate juice may have protected hemoglobin from oxidize agents

Concerning the serum levels of iron and Aluminum, the recorded results in Table 3, revealed that the serum level of iron increased significantly in all the treated groups as compared to positive control which means that both pomegranate and beetroot have an amelioration effect on serum iron which reflects on their blood content of hemoglobin which lead us to hypothesize that the decrease occurred in serum hemoglobin of the positive control is attributed to decrease in iron content as a result of aluminum toxicity. The findings also showed that the high toxicity caused by aluminum demonstrated to the positive control has been reduced significantly in the treated groups either by pomegranate, beetroot or their mixture juices in comparing with the positive control. Although the reduction in Al content in the treated groups was remarkable but it was still lower than that of the negative control. However, the highest

reduction percentage in aluminum concentration (79%) was registered in the mixture group followed by beetroot group (66%) and pomegranate (52.5%) which means that the mixture of both pomegranate and red beetroot juices is more effective than each of them separately which makes it the best among the three treated groups.

Table 3. Effect of pomegranate, red beetroot juices or their mixture on growth parameters, hemoglobin (Hb) and serum Iron (Fe) and Aluminum (Al) of the rats affected by aluminum toxicity.

Parameters groups	BWG (%)	FI (g/d)	Hb (g/dl)	Fe (µmol/L)	AL (µmol/L)
Negative control	88.86±5.38[a]	18.85±0.57[a]	13.75±0.44[a]	62.03±2.87[a]	0.187±0.004[e]
Positive control	37.03±2.31[c]	13.97±0.88[d]	9.42±0.46[d]	24.06±0.14[d]	1.834±0.027[a]
Pomegranate juice	79.96±5.54[b]	17.11±0.60[bc]	11.17±0.29[c]	48.14±0.51[c]	0.868±0.011[b]
Red beetroot juice	82.33±3.84[b]	16.77±0.59[c]	12.91±0.81[b]	52.90±0.31[b]	0.618±0.004[c]
Mixture juice	84.28±4.19[ab]	17.97±0.79[ab]	13.28±0.51[ab]	61.83±0.88[a]	0.385±0.005[d]

Means ± SD.
Values with the same letters in each column are not significant at P < 0.05 .

The obtained results were like that those of El-Metwally and El-Serwy (2014) who stated that aluminum toxicity caused significant decreases in body weight gain %, feed intake and hemoglobin of experimental rats. Red beetroot juice appears to be more effective as an antioxidant, anti-ischemic and anti-anemic. Consumption of red beetroot products may also be helpful in improving intestinal peristalsis and fat metabolism (Babarykin *et al*., 2019). The Improvement in BWG, FI and Fe as a result of consumption of red beetroot juice is attributed to its content of carotenoids, minerals, flavonoids ,vitamin A and B complex vitamins as niacin, pantothenic acid, pyridoxine and folate (Mohammed *et al*., 2011).

Effect of pomegranate, red beetroot juices or their mixture on oxidative status in serum of the rats affected by aluminum toxicity.

The results in Table 4 showed that the serum superoxide dismutase (SOD) concentration in the positive control group has reduced significantly as compared to the normal control. The reduction resulted from aluminum toxicity reached 43%. The treated groups revealed significant increases in SOD values where the juices of pomegranate, red beetroot and their mixture led to significant increases in serum SOD with values of 170.6, 175.0 and 184.0 U/ml, respectively as compared to positive control (107.6 U/ml). The mixture group revealed the best amelioration where the value of its serum SOD is nearly like that of the normal control (188.6 U/ml).

Regarding serum glutathione (GSH) levels, it was noticed that the aluminum toxicity in the positive control caused significant decrease in serum glutathione (7.95 nmol/ml) in comparing with negative control (14.76 nmol/ml). On the other hand, significant increases in serum glutathione were noticed in the three treated groups. All the treated groups revealed the same effect in reducing the toxicity of aluminum, but the mixture juice revealed the best results in this respect where the improvement caused by the mixture reached the normal value of the negative control.

Table 4 . Effect of pomegranate, red beetroot juices or their mixture on oxidative status in serum of the rats affected by aluminum toxicity .

Parameters Groups	SOD (U/ml)	GSH (nmol/ml)	MDA (mmol/L)	T.P (g/dl)	AchE (u/L)
Negative control	188.60±4.04[a]	14.76±0.14[a]	4.98±0.04[e]	7.01±0.06[a]	109.20±1.30[e]
Positive control	107.60±3.44[c]	7.95±0.18[c]	11.97±0.03[a]	4.90±0.11[e]	382.40±5.50[a]
Pomegranate juice	170.60±4.39[b]	12.46±0.25[b]	5.70±0.05[b]	6.49±0.04[d]	224.60±5.03[b]
Red beetroot juice	175.00±4.12[b]	12.61±0.32[b]	5.39±0.03[c]	6.68±0.03[c]	204.60±5.03[c]
Mixture juice	184.00±4.18[a]	14.50±0.54[a]	5.22±0.12[d]	6.88±0.04[b]	125.80±7.69[d]

Means ± SD.
Values with the same letters in each column are not significant at P < 0.05

Glutathione plays a protective role in the metabolism of many toxic agents, so it is usually used as an antioxidant marker. It acts as a free radical blocking agent and maintains cytochrome P-450 by blocking lipid peroxide (Yuan *et al*., 2008). The improvement in GSH level as a result of consumption pomegranate, red beetroot or their mixture juices may be attributed to their protection mechanism against $AlCl_3$ toxicity as a result of GSH restoration.

The finding in the same table stated that the aluminum toxicity in the positive control group led to significant increase in the serum MDA concentration which reached a percentage of 140% in comparing with negative control. The treated groups showed significant decreases in serum MDA levels as compared to positive control. The maximum reduction percentage reached 56.4% in the mixture group.

Concerning serum total protein, the findings showed that its concentration has been reduced significantly in the serum of positive group rats (4.9 g/dl) as compared to normal control (7.01g/dl). The three treated groups exhibited significant increase values of their serum total protein levels which reached 6.49, 6.68 and 6.88 g/dl, respectively in comparing with positive control. On the other hand, high level of AchE enzyme was noticed in the positive control group which treated by $AlCl_3$ only as compared to negative control, the increase percentage reached 250%as a result of Aluminum toxicity. Remarkable decrease in AchE occurred in the three treated groups in significant values as compared to positive control. The maximum reduction has been observed in the

mixture group with a percentage of 67%. It is obvious that the phytochemical content of both pomegranate and red beetroot such as carotene, Vit. E, Vit. C and thiamine improved the levels of SOD and GSH. The enzymatic scavenging of ROS by the used juices should be achieved by the combination of their content of the phytochemical they contain. Georgiev *et al*. (2010) stated that the higher content of the plant phytochemicals which are good antioxidants act as a very good free radical scavenger and hence prevent the diseases occurred as a result of oxidative damage.

Effect of pomegranate, red beetroot juices or their mixture on brain function parameters of the rats affected by aluminum toxicity.

The data in Table (5) describes the main parameters related to brain functions. Superoxide dismutase (SOD) and glutathione peroxidase (GSH) are the main antioxidants either in blood or in the brain. The results revealed that the rats of the positive group which had been affected by aluminum toxicity exhibited low significant levels of brain SOD (64.9 U/g) and GSH (11.14mg/g) as compared to the negative control group as being (245.6 U/g and 24.81mg/g), respectively. The treated groups either by pomegranate, red beetroot or their mixture showed significant increases in levels of SOD and GSH in comparing with the positive control group. The best improvement was noticed in the rats treated by the mixture of the two juices where the increases percentages reached 260% in brain SOD level and 107% for GSH level. This means that Aluminum toxicity leads to severe decreases in brain antioxidants which make the cells of brain are susceptible to oxidation and stroke. It has been reported that there is a correlation between oxidative stress status and most neurodegenerative disorders caused by high levels of aluminum (Verstraeten *et al*. ,2008). However, the used juices in this work showed remarkable improvements in the brain content of the antioxidant levels which preserve the brain cells in a healthy state. Regarding the brain malondialdehyde (MDA), Acetylcholine esterase (AchE) and β-amyloid concentrations, the findings in table (5) show that their levels increased significantly in the positive control group in comparing with the negative control group. The increase percentages were 65.25% for MDA, 65.4% for AchE and 880% for β -amyloid. On the other hand, the treated groups revealed significant decreases in the mentioned three parameters as compared to the positive control group. The rats of the mixture group exhibited the best results especially in MDA level where its concentration reached nearly to that of the normal (negative) group while there is no significance between them. Regarding AchE, the treated groups of beetroot and mixture juices have the same effect in reducing brain AchE level which was better than that of pomegranate group despite their levels remain higher than that of the normal control group. Although the decrease in brain β -amyloid concentrations occurred in the treated groups was very high but it was still significantly low than that of negative control. It is reported that the increase in β -amyloid (βA) is a potential marker of Alzheimer's disease (Glenner and Wong ,1984) and aluminum is one of the risk factors in β -amyloid elevation (Zhang *et al*., 2015) .

Table 5. Effect of pomegranate, red beetroot juices or their mixture on brain function parameters of the rats affected by aluminum toxicity.

Parameters Groups	SOD (U/g)	GSH (mg/g)	MDA (mmol/g)	T.P (mg/g)	AchE (U/mg)	β- amyloid (pg/ml)	RNA (mg/g)	DNA (mg/g)
Negative control	245.62 ±7.18^{a}	24.81 ±2.83^{a}	176.42 ±4.59^{d}	108.60 ±2.07^{a}	379.60 ±33.19^{d}	5.14 ±0.08^{e}	75.77 ±3.30^{a}	112.00 ±2.55^{a}
Positive control	64.90 ±4.78^{e}	11.14 ±0.81^{d}	291.54 ±3.38^{a}	73.60 ±8.02^{c}	593.80 ±30.07^{a}	50.04 ±0.32^{a}	27.19 ±2.36^{e}	42.62 ±3.33^{d}
Pomegranate juice	202.81 ±6.42^{d}	19.34 ±0.56^{c}	213.17 ±7.59^{b}	97.00 ±4.53^{b}	472.60 ±21.29^{b}	19.04 ±0.53^{b}	48.42 ±3.20^{d}	94.98 ±4.40^{c}
Red beetroot juice	217.23 ±3.34^{c}	21.74 ±1.18^{b}	198.84 ±6.06^{c}	100.00 ±1.87^{b}	428.60 ±23.16^{c}	11.71 ±0.56^{c}	55.84 ±3.96^{c}	94.62 ±3.79^{c}
Mixture juice	233.95 ±5.97^{b}	23.02 ±1.81^{ab}	183.70 ±5.25^{d}	102.80 ±3.70^{ab}	425.80 ±18.29^{c}	10.68 ±0.40^{d}	69.44 ±2.00^{b}	103.66 ±2.81^{b}

Means ± SD.

Values with the same letters in each column are not significant at P < 0.05

The results in Table 5 also showed that the brain total protein level have been reduced significantly in the positive control group which treated by $AlCl_3$ only in comparing with negative control. The decrease percentage reached 47.5%. From the other side, the treated groups revealed significant increases in brain protein as compared to positive control which reached its maximum in the group treated by the mixture of the two juices that was similar nearly to that of the negative control. Regarding RNA and DNA concentrations in the brain, the findings revealed that their values reduced significantly in the positive control in comparing with negative control group where the RNA value reduced from 75.77 to 27.19 mg/g while DNA reduced from 112.0mg/g in negative control to 42.62mg/g in positive control. However, the treatments by pomegranate or beetroot or their mixture juices showed significant increases in both RNA and DNA levels. The mixture-treated group exhibited the highest improvement in the DNA and RNA levels followed by the groups of red beetroot and pomegranate juices.

Shati *et al*. (2011) stated that the reduction in antioxidants level versus the oxidation ratio is one of the risk factors that leads to generation of oxidative stress. It has been noticed in previous study that the exposure to aluminum contamination caused increased reactive oxygen species (ROS) due to electron leakage and increased electron chain activity. The risk of ROS represents in attacking the cell membrane lipids and hence causes lipid peroxidation (Flora *et al*.,2003). It is well known that aluminum binds by the protein transferrin which carries

ferric ions thus reducing the binding of ferrous ions which may cause oxidation of cell membrane lipids and hence leads to membrane damage (Nehru and Anand, 2005).

It was found that the high concentrations of intracellular aluminum decreased the activities of SOD enzyme and GSH because of reducing the synthesis of the enzyme proteins as mentioned by Albendea *et al*. (2007).

The decreases in brain levels of protein, RNA and DNA in the positive group which treated by $AlCl_3$ only referred to binding of Aluminum to phosphate group of these compounds as well as its binding by the different metals conjugated with protein and thus affect the balance of other metals which reflects on brain functions (Said and Abd Rabo, 2017).

The elevation in brain acetylcholinesterase activity, occurred as a result of $AlCl_3$ administration to rats in comparing with normal control, was in the same line with that obtained by Kumar *et al*. (2009) and Zatta *et al*. (1994) who referred that to the interaction between Al ions and peripheral sites of acetylcholinesterase which changes its structure and enhanced its activity.

The antioxidant activity of pomegranate and red beetroot is attributed to their content of phenolics, flavonoids and anthocyanins which might be the main factors in improving the brain function and lowering aluminum toxicity. Hassanen *et al*. (2011) stated that pomegranate is rich in in the three non-methylated anthocyanidins (cyanidin, delphinidin, and pelargonidin) and their 3-glucosides and 3,5-glucosides. It was mentioned that anthocyanidins could inhibit lipid peroxidation of liposome or cell membranes (De Gaulejac *et al*., 1999).

CONCLUSION

It was concluded that pomegranate or red beetroots have protective effect against oxidative stress resulting from aluminum toxicity in rats because of their high content of phytochemicals such as phenolic compound, flavonoids, anthocyanins, vitamin C which make them act as strong antioxidants and hence prevent the oxidation of cell membranes especially those of brain cells. It was noticed that consumption of the examined juices improved the antioxidant parameters in both serum and brain cells as well as remarkable amelioration in brain function markers such as amyloid β- peptide, AChE enzyme, DNA and RNA content. So, it is recommended by consuming a cup (about 200 ml) daily of pomegranate juice or red beetroot juice or their mixture for those who are exposed to aluminum toxicity or the patients of Alzheimer's disease.

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التأثير الوقائي لعصائر الرمان والبنجر الاحمر ضد سمية الالمونيوم في المخ لدى ذكور الفئران

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تم إجراء هذه الدراسة بغرض دراسة التأثير الوقائي لعصير كل من الرمان والبنجر الأحمر أو خليطهما على سمية المخ الناجمة عن الحقن بكلوريد الالمونيوم لدى فئران التجارب. وقد استخدم في التجربة خمس وثلاثون فأرا من فئران التجارب تم تقسيمهم الى خمس مجموعات (سبعة فئران لكل مجموعة) ، تغذت إحداها على الوجبة القياسية فقط خلال فترة التجربة (المجموعة الضابطة السالبة). تم حقن مجموعة أخرى بواسطة $AlCl_3$ بدءًا من اليوم الرابع عشر حتى نهاية التجربة واعتبرت المجموعة الضابطة الموجبة) ، في حين تم معاملة المجموعات الثلاث المتبقية بواسطة عصائرالرمان والبنجر الأحمر وخليط منهما من اليوم الأول للتجربة وحتى نهايتها. وقد تم حقن المجموعات الثلاث المعالجة يوميًا بواسطة $AlCl_3$ ابتداءامن اليوم الرابع عشر للتجربة وحتى نهايتها. و قد تم تقدير المركبات الفينوليه، والفلافونويد ، وفيتامين C و النشاط الكابح للشقوق الحرة (DPPH) في كل من عصير الرمان والبنجر الأحمر. وقد أظهرت النتائج عن أن المجموعة الضابطة الموجبة أظهرت مستويات منخفضة من مؤشرات النمو والهيموجلوبين والحديد ومستوى عالٍ من الألمونيوم مقارنةً بالمجموعة الضابطة السالبة، في حين اسفرت المجموعات المعاملة بالعصائرعن تحسن كبير في هذه المؤشرات. وفيما يتعلق بمؤشرات مضادات الأكسدة فقد أوضحت النتائج أن المجموعات الثلاث التي تمت معالجتها اظهرت زيادة ملحوظة في إنزيم سوبراكسيد ديسموتاز والجلوتاثيون والبروتينات الكلية وانخفاضا كبيرا في المالوندايألدهيد وألأسيتيل كولين إستيريز في كل من سيرم و خلايا المخ لدى الفئران مقارنةً بالمجموعة الضابطة الموجبة. كشفت النتائج أيضا عن انخفاض معنوي في مستوى البيتا أميلويد بينما ارتفعت كمية كل من الحمض النووي الريبوزي والحمض النووي الديؤكسي ريبوزي بشكل ملحوظ في مخ الفئران المعاملة بالعصائر الثلاثة مقارنة بالمجموعة الضابطة الموجبة. وخلصت الدراسة إلى أن مخلوط عصيري الرمان والبنجر كان أفضل العصائر من حيث التأثير الوقائي ضد سمية الألومنيوم. ولذا توصي الدراسة بأهمية تناول عصائر الرمان والبنجر الأحمر أو خليطهما لأولئك الذين يتعرضون للألومونيوم أو للوقاية من مرض الزهايمر.

الكلمات الافتتاحية: الرمان – البنجر الاحمر- سمية الالمونيوم - المخ- مضادات الاكسدة .