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BIOCHEMICAL EFFECT OF CERTAIN ANTIOXIDANTS ON OXIDATIVE STRESS AND MITOCHONDRIAL DYSFUNCTION IN BRAIN OF EXPERIMENTLLY INDUCED HEPATIC FAILURE IN RATS

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A B S T R A C T

The present research aimed to evaluate the hepato/neuroprotective effects of Rutin and Resveratrol as natural antioxidants on brain and liver tissues of experimental rats exposed to acute liver failure induced by i.p. administration of Thioacetamide (TAA), Through evaluation of plasma and brain Ammonia, serum Alanine Aminotransferase (ALT), Aspartate Aminotransferase (AST), Alkaline Phosphatase (ALP) and Gamma Glutamyl-Transferase (γ-GT), Albumin, Total Protein, Total Bilirubin, Urea and Uric acid. Levels of reduced glutathione (GSH) and activities of Superoxide Dismutase (SOD), Catalase (CAT), Glutathione Peroxidase (GPx), were determined in the liver and brain tissues. Extent of oxidative stress was also assessed by hepatic and brain lipid peroxides (MDA), in addition to brain nitric oxide (NO) and Monoamine oxidase (MAO). Thioacetamide induced a significant increase in 1) ALT, AST, ALP, γ-GT, Albumin; Total Protein, Total Bilirubin, Urea and Uric acid Levels in serum, 2) plasma and brain ammonia level 3) brain NO level, 4) liver and brain MDA. Also marked depletion in liver and brain GSH, CAT, SOD, GPx and brain MAO were observed after TAA intoxication. Rutin and Resveratrol Pretreatment was able to mitigate hepatic and brain damage induced by TAA and showed pronounced curative effect against lipid peroxidation and deviated serum enzymatic variables as well as maintained glutathione status and antioxidant enzymes toward control levels. Pretreatment of rutin and resveratrol was highly effective and protective against TAA induced hepatic encephalopathy. The results of the present study suggest that rutin and resveratrol have potential to exert curative effects against liver injury.

KEY WORDS: Oxidative stress, Liver Failure, Rutin, Resveratrol, Thioacetamide

1. INTRODUCTION

Hepatic encephalopathy (HE) is a major neuropsychiatric complication of both acute and chronic liver failure. Symptoms of HE include attention deficits, alterations of sleep patterns and muscular incoordination progressing to stupor and coma. HE in acute liver failure may include seizures. Despite several decades of intensive scientific research, the precise causes of HE are still unknown. Attention has been focused on two major areas, namely the role of blood-borne neurotoxins (particularly ammonia) and the key role of the astrocyte [11]. The main symptoms of HE are ranging from minimal intellectual dysfunction to coma. Baskarana *et al.* [7] found that the pathological lesions caused by hepatotoxins may resemble those of any known type of liver diseases. As proposed by Luster *et al.* [40], hepatotoxins initially damage the centrilobular regions of liver where there are high levels of cytochrome P450 mixed function oxidases that mediate their conversion to toxic intermediates,

followed by reactive oxygen species (ROS) production, lipid peroxidation and release of pro-inflammatory cytokines. Thioacetamide is a highly specific hepatotoxic material causing liver injury and dysfunction, containing thiono-sulfur compound and is well known to induce hepatic damage by generation of ROS [63]. Shortly after administration, the thiono-sulfur group of TAA undergoes an extensive metabolism by the mixed function oxidase system in the body to produce acetamide, that does not have liver necrotizing properties, and TAA-S-oxide by a microsomal monooxygenase requiring NADPH and cytochrome P450 [7]. In a further step, TAA-S-oxide is transformed to TAA-S-S-dioxide, which is a highly reactive, unstable compound that is thought to covalently binding to liver macromolecules and responsible for initiation of hepatic damage and centrilobular necrosis[16], hyperammonemia [59], and generation of ROS that leads to hepatocellular death via oxidative stress [55], which is implicated, in part, in the pathogenesis of FHF by enhancing free radical-mediated damage to proteins, lipids and DNA [21].

Rutin is one of the most common native flavonoids occurring mainly in glycosidic forms, it is the flavonoid most abundantly consumed in foods, and is abundantly present in onions, apples, tea and red wine [41]. It is used by the animal feed, cosmetic, and chemical industries as a natural pigment, stabilizer, food preservative, and UV absorbent [46]. Also it exhibits multiple pharmacological activities including antibacterial, antiprotozoal, anti-tumor, anti-inflammatory, anti-diarrheal, anti-ulcer, anti-mutagenic, vasodilator and immunomodulator properties [13]. In addition, hypo-lipidaemic, cytoprotective [33], antispasmodic and anti-carcinogenic activities have also been reported. It has also been found to prevent gastric mucosal ulceration in animal models including restraint stress [28].

Resveratrol (3,4,5-trihydroxy-trans-stilbene), a natural polyphenol found mainly in grapes and red wine, has been reported to have a wide range of biological properties and potent antioxidant activities [48], however, evidence has demonstrated that this compound also possesses anti-inflammatory [35], anti-aggregate and neuroprotective properties [54]. Based on these findings, resveratrol has become attractive as a therapeutic agent in the treatment of a variety of pathologies including neurodegeneration, cancer, cardiovascular disease and diabetes mellitus [35]. Resveratrol has an intrinsic antioxidant capacity that could be related to its chemopreventive effects. In vitro, the induction of detoxification enzymes has been shown after low doses of resveratrol [39]. In vivo, resveratrol has been shown to increase plasma antioxidant capacity and to decrease lipid peroxidation [64], which is strongly associated with the risk of coronary heart disease and myocardial infarction [8].

2. MATERIAL AND METHODS

2.1. Chemicals and antioxidants

Thioacetamide (purity~99%) (Loba Chemi. Co, Delhi. India) was purchased from El-Gomhouria Co. for Trading Chemicals, Medicines and Medical Appliances, Egypt. Resveratrol (purity~ 99%) (Sigma Chemical Co., St. Louis, Mo, USA) and purchased from Schnelldorf, Germany through the Egyptian International Center for Import Cairo, Egypt. Rutin (purity~99%) (Egyptian Pharmaceutical International Company (E.P.I.C.O)). All other chemicals were of analytical grade and were obtained from standard commercial suppliers.

2.2. Experimental animals

A total number of 52 Male albino rats, 6-8 weeks old and average body weight 150-180 gm were used in the experimental investigation of this study, and obtained from the Laboratory Animals Research

Center, Fac. Vet. Med., Benha University, housed in separate wire mesh cages, exposed to good ventilation, humidity and to a 12-hr light - dark cycle, and provided with a constant supply of standard pellet diet and fresh, clean drinking water ad libitum.

2.3. Preparation and administration of dosage

Thioacetamide was dissolved in 0.9% NaCl solution, and administered to rats at a dose of (300 mg/kg b.wt) through i.p route, for two consecutive days with 24 hrs interval for induction of acute liver failure. Resveratrol was dissolved in 5% Ethanol, and administered to rats at a dose of (15 mg/kg b.wt) daily through i.p route. Rutin was dissolved in propylene glycol, and administered to rats at a dose of (200 mg/kg b.wt) daily p.o.

2.4. Experimental design

Rats were randomly divided into four main groups (n=7/group) , placed in individual cages and classified as following: (Group 1) served as control normal group (15 rats); (Group 2) served as induced hepatic failure group (15 rats) administered with TAA (300 mg/kg b.wt, i.p); (Group 3) served as rutin protected group (15 rats) administered with rutin (200 mg/kg b.wt, daily p.o) for 3 weeks followed by induction of AHF by TAA dose (300 mg/kg b.wt, i.p at the last 2 days of protection period; (Group 4) served as resveratrol protected group (7 rats) administered with resveratrol (15 mg/kg b.wt, i.p daily for 7 days followed by induction of AHF by TAA dose (300 mg/kg b.wt, i.p at the last 2 days of protection period.

At the end of the experimental period, rats were fasted overnight, blood samples were taken from retro-arbitral plexus. 1ml of the blood was collected on EDTA for ammonia analysis. The rest of blood samples were collected in dry, clean test tubes and allowed to clot for 30 min and serum was separated by centrifugation at 3000 rpm for 15 min at 4 ºc. The serum was separated by automatic pipette and received in dry sterile tubes, processed directly for ALT, AST, ALP, and GGT. Then kept in a deep freezer at -20 ºc until used for subsequent biochemical analysis .All serum samples were analyzed for the following parameters: Albumin, Total Protein, Total Bilirubin, Urea and Uric acid. Then liver and brain samples were collected for estimation of L-MDA, GPx, CAT, SOD, GSH, MAO and NO.

2.5. Statistical analysis

The results were expressed as mean (±S.E.) and statistical significance was evaluated by one way ANOVA using SPSS (version 10.0) program followed by the post hoc test, least significant difference (LSD). Values were considered statistically significant when $p < 0.05$.

3. RESULTS AND DISCUSSION

The liver plays a crucial role in the metabolic elimination of most drugs and other foreign compounds, thus making it an important target for toxicity. Hepatotoxic agents can react with the basic cellular components and consequently induce almost all types of liver lesions [58]. When liver injury occurs, intracellular components released from necrotic cells are able to activate immune cells and trigger the -ROS-mediated cell killing process, which leads to more necrosis and amplified inflammation [25]. TAA is a potent hepatotoxic agent [14] induces FHF that considered being a well-characterized model of acute liver failure-related hepatic encephalopathy [12]. TAA is metabolized by hepatic cytochrome P4502E1 to more toxic products: thioacetamide sulfoxide (TAASO) and thioacetamide-S, S-dioxide (TAASO2) to initiate hepatocellular necrosis with ROS [17]. During hepatic inadequacy, as occurs in AHF, large quantities of ammonia in the portal blood escapes the detoxification process and

enters systemic circulation. Thus, blood and tissue (brain) ammonia levels are elevated rapidly in AHF [51]. Maximum reduction in ammonia level was observed following treatment with rutin and resveratrol, which may be due to the significant anti-hyperammonemic activity of rutin and resveratrol that is related to modulation of oxidant antioxidant imbalance in AHF and free radical scavenging properties [19].

Liver cell destruction results in the leaking out of tissue contents into the blood stream. Serum AST, ALT, ALP and γ-GT are the most sensitive markers employed in the diagnosis of liver diseases [41]. When the liver cell plasma membrane is damaged, numerous enzymes normally located in the cytosol are released into the blood stream [50], and their estimation in serum is a useful quantitative marker to indicate hepatocellular damage [24]. Animals exposed to TAA showed necrotic changes resulting in the release of hepatic enzymes (AST, ALT, ALP, GGT and bilirubin) that mark liver injury [7]. Jain and Singhai [26] interpreted the elevated levels of AST and ALT as a result of the hepatocytes damage or alterations in the membrane permeability indicating the severity of hepatocellular damage induced by TAA, which is in accordance with previous reports of [57]. In contrast, an increase in ALP activity and bilirubin level reflects the pathological alteration in biliary flow. Increase in serum total bilirubin concentration after TAA administration might be attributed to the failure of normal uptake, conjugation and excretion by the damaged hepatic parenchyma [20]. Pretreatment with rutin significantly decreased the levels of AST, ALT, ALP and γ-GT, suggesting that it offer protection by preserving the structural integrity of the hepatocellular membrane against hepatotoxins [41]. Also resveratrol was able to protect against the increase in the activity of these enzymes in AHF rats, demonstrating the protective effect of this polyphenol against brain and liver damage induced by TAA. These results are in agreement with those found in studies using resveratrol [32, 56] and can be attributed to the capability of resveratrol to conserve the membrane integrity of cellular organelles [56]. Diminishment in γ-GT and ALP after resveratrol treatment is also indicative of its membrane stabilizing activity [20]. Also, Kasdallah-Grissa *et al.* [31] showed that resveratrol diminished the hepatic tissue injury associated with reduction in bilirubin level indicating a protective role of resveratrol against TAA toxicity in the liver.

The obtained data in table (1) revealed a significant increase in ALT, AST, ALP, GGT, Ammonia, Urea, Uric acid, and Total Bilirubin in TAA induced AHF group, accompanied with significant decrease in Albumin and Total protein levels, when compared with control normal group. Pretreatment with rutin and resveratrol in TAA-induced AHF in rats resulted in significant decreases in ALT, AST, ALP, GGT, Ammonia, Urea, Uric acid and Total Bilirubin, accompanied with significant increases in Albumin and Total protein levels, in comparison with TAA treated group.

The obtained data demonstrated in table (1) revealed that, administration of TAA to normal rats exhibited a significant decrease in serum total protein and albumin concentration, observed 24 hrs after induction of AHF when compared with control normal group. This decrease could be ascribed to increased rate of lipid peroxidation, decreased amino acids uptake, greatly decreased concentration of variety of essential amino acids, and increased conversion rate of glycogenic amino acids to CO_2 and H_2O and reduction in protein synthesis secondary to a decreased amount and availability of mRNA [1]. Also might be due to increased catabolism of proteins and defect in protein biosynthesis that might be due to the consequences of disruption and dissociation of polyribosomes from rough

endoplasmic reticulum [18]. Pretreatment of rats with rutin and resveratrol exhibited a significant elevation in serum total protein and albumin concentration, in comparison with TAA induced AHF group, these results may be related to enhanced protein biosynthesis. This suggestion was supported by the findings of Bhadauria et al. [9].

Blood BUN, uric acid and creatinine levels can be useful indicators of renal function. Renal damage can be accompanied by an increase in serum BUN, uric acid and creatinine indicating reduced urea, uric acid and creatinine clearance. The rate of urea formation depends on the rate of protein catabolism. Increases in BUN levels may reflect an accelerated rate of protein catabolism rather than decreased urinary excretion of urea [23]. Hyperuricemia was mainly attributed to impaired renal clearance of uric acid rather than over production [38]. Also the highly significant increase in serum urea concentrations of ALF rats may be due to depletion of serum protein, increase in the rate of circulating amino acids, and deamination takes place that consequently leads to the formation of large amount of ammonia which is eventually converted to urea [22]. Maximum reduction in urea levels was observed following treatment with rutin, which may be due to the significant anti-hyperammonemic activity of rutin. This is probably indicative of the antioxidant efficacy of the used polyphenolic flavonoid [41]. Of great importance, resveratrol prevented the increase in the urea levels in AHF rats. These findings suggest that resveratrol possesses the potential to attenuate renal injury caused by hyperammonemia state. This can be associated directly with the antioxidant capacity of this polyphenol, which protects the kidneys against oxidative damage [32].

MDA is the main product of lipid peroxidation and its concentration usually reflects the total level of lipid peroxidation [60]. Ansil et al. [4] observed that TAA treatment caused a significant increase in hepatic MDA level, when compared with normal control group. TAA has been found to stimulate lipid peroxidation by generation of ROS [10]. TAA also decreased the GSH/GSSG ratio, and increased the susceptibility of hepatocytes to in vitro lipid peroxidation [55].

Rutin, being an anti-lipoperoxidant agent, inhibits formation of lipid peroxides [53]. It acts by lowering the lipid peroxidation, scavenging free radicals and its activity is attributed to its structure [3]. This may be due to the acute antioxidant effects of the bioflavonoid rutin that showed maximum benefits, higher scavenger efficiency and more antioxidant activity, which seems to be correlated to its structure [2]. This effect may be attributable to the catechol structure of ring B, the 2, 3 double bond in conjugation with a 4-oxo function, and the presence of both 7- and 5-hydroxyl groups [53].

Table 1 Effect of Rutin and Resveratrol pretreatment on blood biochemical parameters of TAA-induced AHF in male rats

Animal groups	Ammonia µg/dl	ALT U/L	AST U/L	ALP U/L	GGT U/L	Bilirubin (mg/dl)	Albumin (g/dl)	Protein (g/dl)	Urea (mg/dl)	Uric acid (mg/dl)
Control	133.53 ± 4.27 [d]	45.48 ±2.20 [c]	162.45 ±1.96 [d]	226.00 ±2.70 [c]	1.784 ±0.111 [c]	0.395 ± 8.37 [d]	3.69 ± 9.57 [a]	6.28 ±9.22 [a,b]	40.28 ± 1.33 [c]	1.646 ±0.104 [c]
TAA-treated	323.23 ± 4.39 [a]	218.76 ±7.36 [a]	427.71 ±7.57 [a]	504.71 ±5.26 [a]	6.715 ±0.313 [a]	0.920 ± 7.22 [a]	3.23 ± 8.58 [b]	5.56 ± 9.03 [c]	86.77 ± 1.67 [b]	4.076 ±0.162 [a]
Rutin + TAA	175.02 ± 2.41 [c]	86.82 ±2.74 [b]	201.95 ±4.23 [b]	304.46 ±3.65 [b]	3.430 ±0.165 [b]	0.483 ± 1.80 [c]	3.54 ± 8.89 [a]	6.50 ± 9.28 [a]	51.73 ± 1.56 [b]	1.853 ±0.119 [b,c]
RESV + TAA	189.14 ± 2.53 [b]	91.80 ±5.12 [b]	184.64 ±3.38 [c]	311.61 ±4.41 [b]	2.271 ±0.137 [c]	0.585 ± 1.96 [b]	3.48 ± 9.11 [a,b]	6.17 ± 0.12 [b]	39.84 ± 0.99 [c]	2.100 ± 0.123 [b]

Data are presented as Mean (±S.E). Mean values with different superscript letters in the same column are significantly different at (P<0.05).

Resveratrol has been reported to prevent oxidative stress and LPO processes [15], which might be due to the phenolic moiety present in its structure. The ability of the resveratrol is to exert protective effect against intoxication by reducing the MDA production that is indicative of its antioxidant activity. Significantly Lee *et al.* [35] shows that resveratrol treatment intensively lowered MDA level in resveratrol treated rats than ischemia rats.

The obtained data in table (2) revealed a significant increase in L-MDA level and significant decreases in SOD, GPx, CAT activities and GSH level in liver and brain tissue homogenate in TAA induced AHF group, when compared with control normal group. Pretreatment with rutin and resveratrol in TAA-induced AHF in rats resulted in significant decrease in L-MDA level and significant increases in SOD, GPx, CAT activities and GSH level in liver and brain tissue homogenate, when compared with TAA treated group.

The obtained data demonstrated in table (2) revealed that, administration of TAA to normal rats exhibited a significant reduction in liver and brain SOD, GPx, CAT activities and GSH level, observed 24 hrs after induction of AHF when compared with control normal group. Studies in TAA models of liver failure indicate a higher free radical activity in the liver, as shown by the increase in mitochondrial superoxide radical and H_2O_2 and the induction of the microsomal cytochrome P-450 [37]. Higher pro-oxidant liver status in rats with TAA is likely to involve a high consumption of cellular and circulate antioxidants. This could be partly related to the decrease in liver and brain activities of CAT, GSH and GPx, otherwise lowering ROS [34]. TAA also produced oxidative stress by depleting the GSH level suggesting the presence of free radicals generated by TAA. The antioxidant enzymes (CAT, SOD and GPx) assays showed that TAA treatment caused the depletion of these enzymes; therefore, it could be said that TAA caused the cellular damage by inhibiting the activity of the antioxidant enzymes [55]. by TAA, Rats given rutin showed significant improvement in the activity of GSH, GPx, SOD and catalase thus suggesting its role in scavenging the free radicals generated. This may be due to the acute antioxidant effects of the bioflavonoid rutin that showed maximum benefits, higher scavenger efficiency and more antioxidant activity, which seems to be correlated to its structure [2]. This effect may be attributable to the catechol structure of ring B, the 2,3 double bond in conjugation with a 4-oxo function, and the presence of both 7- and 5-hydroxyl groups [53].

Resveratrol exerts antioxidant effect not only in brain but also in liver, heart and testis of rats [30]. Since resveratrol is a potent free radical scavenger, it reduces LPO and increases cellular GSH level against traumatic brain injury [5,6].

Table 2: Effect of Rutin and Resveratrol pretreatment on liver antioxidant parameters of TAA-induced AHF in male rats:

Animal groups	L-MDA (nmol/gm. tissue)		Catalase (K/gm. tissue)		GPx (mU/gm. tissue)		SOD (U/gm. tissue)		GSH (mg/gm. tissue)	
	Liver	Brain	Liver	Brain	Liver	Brain	Liver	Brain	Liver	Brain
Control	59.36 ± 2.7[c]	51.38 ± 2.74[c]	31.17 ±0.60[a]	21.18 ±0.65[a]	359.43 ±4.66[a]	164.34 ±2.14[a]	560.18 ±5.38[a]	361.89 ±5.07[a]	75.9 ±1.53[a]	75.9 ±1.53[a]
TAA-treated	139.49 ± 3.12[a]	126.25 ± 3.51[a]	17.49 ±0.39[d]	11.48 ±0.38[d]	126.46 ±1.76[d]	76.44 ±2.49[d]	242.34 ±5.74[d]	142.92 ±8.06[c]	47.66 ±1.37[c]	47.66 ± 1.37[c]
Rutin + TAA	70.31 ± 1.37[b]	58.87 ±2.57[b,c]	25.09 ±0.35[c]	18.34 ±0.29[b]	306.52 ±2.54[b]	154.01 ±1.71[b]	454.41 ±8.02[b]	323.11 ±5.41[b]	68.68 ±1.28[b]	68.68 ± 1.28[b]
RESV + TAA	76.13 ± 1.29[b]	63.11 ± 3.17[b]	28.74 ±0.37[b]	15.66 ±0.33[c]	278.12 ±2.07[c]	138.35 ±1.04[c]	423.56 ±5.11[c]	338.57 ±5.06[b]	70.57 ±1.32[b]	70.57 ± 1.32[b]

Data are presented as Mean (±S.E). Mean values with different superscript letters in the same column are significantly different at (P<0.05).

Also Resveratrol is able to induce cellular antioxidants and phase 2 enzymes [42]. These modifications contribute to increase the resistance to hepatic cell injury elicited by ROS. It has been found that resveratrol reduces the generation of H_2O_2, and normalize levels of oxidized glutathione reductase [27].

One of the ROS that elevates in brain tissue due to AHF is NO nevertheless; its precise role in this neuropathology remains controversial [65]. Excess ammonia induces nitric oxide synthase, which leads to enhanced production of nitric oxide and other toxic free radicals in brain and leads to oxidative stress and tissue damage [36]. Rehman *et al.* [52] have shown that AHF accompanied with excess ammonia induces nitric oxide synthase, which leads to enhanced production of nitric oxide, leading to oxidative stress and liver damage. Flavonoids exerted NO production inhibitory activity in several cell lines and cultures (mouse peritoneal macrophages). This effect was probably caused by flavonoid inhibitory effect on expression of inducible NOS but not by the inhibition of its activity [44]. Flavonoids also possess the ability to directly scavenge molecules of NO [49]. Anti-inflammatory effects of flavonoids including rutin have been reported in several studies. Moreover, their role in the inhibition of NO production has also been discussed [43, 62]. Resveratrol is reported to possess significant anti-inflammatory activity in various cells and tissues and is reported to inhibit the production of NO by Kupffer cells in a dose dependent manner

that occurred at a post-transcriptional level [47].

The obtained data in table (3) revealed a significant increase in brain ammonia and nitric oxide levels and significant decrease in MAO activity in brain tissue homogenate in TAA induced AHF group, when compared with control normal group. Pretreatment with rutin and resveratrol in TAA-induced AHF in rats resulted in significant decrease in brain ammonia and nitric oxide levels and significant increase in MAO activity in brain tissue homogenate, when compared with TAA treated group.

The obtained data demonstrated in table (3) revealed that, administration of TAA to normal rats exhibited a significant decrease in brain MAO activity observed 24 hrs after induction of AHF when compared with control normal group. It is remarkable that ammonia injection induces an activation of MAO-A but not of MAO-B. It should be noted that MAO-B is mainly located in astrocytes, while MAO-A is mainly located in neurons. The results obtained indicate that ammonia intoxication does not affect MAO-B in astrocytes but increases neuronal MAO-A. The effect is completely prevented by blocking NMDA receptors with MK-801. These results indicate that activation of NMDA receptors in rat brain in vivo leads to activation of MAO-A [61].

The obtained data demonstrated in table (3) revealed that, administration of TAA to RTN protected and RSV protected rats exhibited a significant elevation in brain MAO activity, in comparison with TAA induced AHF group.

Table 3 Effect of Rutin and Resveratrol pretreatment on brain antioxidant parameters of TAA-induced AHF in male rats

Animal group	Brain Ammonia (μg/gm. tissue)	Brain NO (μmol/gm. tissue)	Brain MAO (U/L)
Control	62.07 ± 1.74 [d]	36.42 ± 1.47 [d]	38.73 ± 1.75 [a]
TAA-treated	198.76 ± 2.95 [a]	105.56 ± 2.59 [a]	14.76 ± 0.93 [c]
Rutin + TAA	91.69 ± 2.44 [c]	62.62 ± 2.09 [b]	30.68 ± 1.71 [b]
RESV + TAA	107.14 ± 4.01 [b]	54.70 ± 2.44 [c]	32.31 ± 1.24 [b]

Data are presented as Mean (±S.E). Mean values with different superscript letters in the same column are significantly different at (P<0.05).

There were no previous data studied the relation between RTN or RSV administration and its effect on MOA activity in the brain of TAA treated rats. But we suggest that the significant increase in brain MAO activity may be due to the neuroactive properties of flavonoids. Nassiri-Asl *et al.* [45] revealed that, the 4-oxo group and the 2, 3 double bond in the C ring of rutin are related to its neuroprotective action**.** Jin *et al.* [29] suggest that the neuroprotective effects of resveratrol may be related to its ability to reduce the inflammatory reaction.

4. CONCLUSIONS AND RECOMMENDATIONS

In conclusion, the findings of the present study demonstrated that rutin and resveratrol pretreatment provided an effective protection against hepatic encephalopathy and oxidative damage in liver and brain induced by TAA in rats, since these compounds were able to ameliorate serum biochemical parameters, enzymatic and non-enzymatic antioxidant defense system and to prevent the lipid peroxidation in these tissues.

We recommended that, we can take advantage of the great hepato/neuro-protective and therapeutic effects of rutin and resveratrol by administrating them for patients suffering from hepatic encephalopathy.

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مجلة بنها للعلوم الطبية البيطرية

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التأثير الكيميائى الحيوى لبعض مضادات الأكسدة على الإجهاد التأكسدى والإختلال الوظيفى للميتوكوندريا فى مخ الفئران المحدث فيها الفشل الكبدى تجريبيا

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الملخص العربي

يهدف هذا البحث إلى دراسة التأثير الكيميائى الحيوى لبعض مضادات الأكسدة على الإجهاد التأكسدى والإختلال الوظيفى للميتوكوندريا فى مخ الفئران المحدث فيها الفشل الكبدى تجريبياً باستخدام مادة ثيوأسيتاميد. وقد وقع الاختيار فى هذا البحث على استخدام اثنين من مضادات الأكسدة الطبيعية من مجموعة الفلافينويد وهما الروتن والرسفراترول، لوقاية الفئران من الفشل الكبدى والإعتلال الدماغى الناتج عنه . وقد أجريت هذه التجربة على اثنين وخمسون من فئران التجارب تتراوح أعمارهم من 8–10 أسابيع وأوزانهم من 150–180 جم، وقسمت الفئران إلى أربعة مجموعات على النحو التالى: المجموعة الأولى (المجموعة الضابطة): اشتملت على 15 فأراً ولم تعطى أية أدوية واستخدمت كمجموعة ضابطة للمجموعات الأخرى. المجموعة الثانية (المجموعة المحدث بها مرض الفشل الكبدى الحاد تجريبياً): تكونت من 15 فأراً تم حقنهم في الغشاء البروتونى بمادة الثيوأسيتاميد بجرعة مقدراها (300 مللى جرام/ كيلوجرام). المجموعة الثالثة: (مجموعة الوقاية بمادة الروتن): تكونت من 15 فأراً تم تجريعهم بالروتن بجرعة مقدراها (200 مللى جرام/ كيلوجرام) يوميا لمدة 3 أسابيع. المجموعة الرابعة: تكونت من سبعة فئران تم حقنهم في الغشاء البروتونى بمادة الرسفراترول بجرعة مقدراها (15 مللى جرام/ كيلوجرام) يوميا لمدة أسبوع. أظهرت النتائج وجود زيادة واضحة فى نشاط خمائر ودلالات وظائف الكبد والكلى فى المجموعة الثانية وعلى العكس ظهر تحسن واضح فى النتائج فى المجموعة الثالثة والرابعة. ذلك فى الانزيمات المضادة للأكسدة فى كبد ومخ الفئران أظهرت النتائج وجود نقص واضح فى تلك الانزيمات فى المجموعة الثانية وعلى العكس ظهر تحسن واضح فى النتائج فى المجموعة الثالثة والرابعة. مما سبق نستنتج أن الروتن والرسفراترول لهما تأثير وقائى واضح فى حماية الكبد والمخ من التأثير المدمر لمادة الثيو أسيتاميد ولذلك ننصح بضرورة استخدامهما كمواد فعالة فى العقاقير المستخدمة لعلاج ووقاية الكبد من مرض الفشل الكبدى.