The effective role of Arabic gum and/or vitamin E on kidney and liver injury induced by food flavor cinnamaldehyde in male albino rats.

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Abstract

Cinnamaldehyde (CNMA) as a food additive is present in low concentrations in human food.It iscommercially prepared by the condensation reaction of benzaldehyde and acetaldehyde and chemically related to toxicologically more active compounds. Accordinglyattempts have been undertaken to protect the body from such toxicity as Gum Arabic(GA) which is a natural productand Vitamin E(VE) as antioxidants.To meet this goal,sixty young adult male albino rats were used to study the therapeutic role of GA and/orVEon liver and kidneyinjury induced by overdose of CNMA.Sixty rats were divided intosixgroups each comprising 10 rats: Control group orally received distilled water,fifty CNMA rats orally received dose 73.5 mg/kg b. wt. of CNMA dissolved in distilled water daily for 3 months then they were divided into:CNMA rats at zero time, CNMA group at 30 daysfollowedwithout any treatment for another 30 days as a recovery period,CNMA+GA therapeutic group orally administered GA at a dose 7.5 g/kg b. wt. daily for another 30 days,CNMA+VE therapeutic orally administered VE at a dose 1g/kg b. wt. daily for another 30 days,CNMA+mixture therapeutic group orally administered mixture of GA and VE at doses 7.5 g/kg b. wt. and 1g/kg b. wt. of GA and VE respectively daily for another 30 days.At the end of experimental period, biochemical, histological and molecular studieswere assessed. Biochemical analysis of serum showed thatinduction with CNMA without treatment revealed a significant decrease in total protein and albumin levels and a significant increase in urea, creatinine levels andserum alanine aminotransferase (ALT),aspartate aminotransferase (AST) and γ-glutamyl transpeptidase (GGT) activities, where treatment with GA or VE or especially both of them revealed marked improvement in the previous biochemical parameters compared to normal control group.The antioxidant analysis revealed a decrease inglutathione (GSH), superoxide dismutase (SOD) and an increase in malondialdhyde (MAD) in CNMA groups, while other groups showed reverse in antioxidant level toward the normal control group. These results were confirmed with kidney histological examination and comet testfor liver tissue. The study showed thatCNMA grouprevealed marked histological lesions particularly in cortical portion of kidney. The Malpighian corpuscles lost their characteristic normal configuration and exhibited clear features of damage. On the other hand, in CNMA group a strong elevation was realized in comet percent as compared with the control group.A considerable improvement was observed in the therapeutic groups. So, the present results

clearly demonstrated the effective role of GA and VE against liver and kidney injury induced by CNMA.

Keywords: Cinnamaldehyde, Gum Arabic,Vitamin E,Comet test,Albino rats.

Introduction

Additives were used for many years for multiple purposes, to maintain product consistency, to enhance palatability, to improve the nutritional value of certain food and for flavor enhancement **(Egbuonu, *et al.*, 2010, Thomas and George, 2010and Eweka, *et al.*, 2011)**.Nevertheless, there are many food additives that exhibit toxicity**(Gowder, 2013)**. The present article is focused on certain widely used additives that have hepatotoxic and nephrotoxic potential.

CNMA as a food additive is present in low concentrations in human food. However, in some cases the use of these compounds is extended to other applications that may require higher doses. The increased exposure of humans to these compounds is a matter of concern and this is the reason why their toxicological properties are becoming of greater relevance **(Stammati*et al.*,1999)**.

CNMA is a pale-yellow liquid with a warm, sweet, spicy odor and pungent taste. It occurs naturally in the leaves and twigs of various species of the genus Cinnamomum. It is commercially prepared by the condensation reaction of benzaldehyde and acetaldehyde and chemically related to toxicologically more active compounds **(Gowder and Halagowder, 2010)**.

Cassia is replacing true cinnamon in the European food market, due to its cheaper price, being largely used in the preparation of some kinds of sweets. Several European health agencies have recently warned against consuming high amounts of cassia due to its high content of coumarin**(Lungarin*et al.*, 2008)**.So, administration of CNMA with uncontrolled values may cause oxidative stress.

Oxidative stress occurs when the homeostatic processes fail and free radical generation is much beyond the capacity of the body's defenses, thus promoting cellularinjury and tissue damage. This damage may involve DNA and protein content of the cells with lipid peroxidation of cellular membranes**(Rahal *et al.*, 2014)**

The human body has several mechanisms to counteract oxidative stress, by producing antioxidants, which are either naturally produced in situ, or externally supplied through foods and supplements. Endogenous and exogenous antioxidants act as free radical scavengers by preventing and repairing damages caused by reactive oxygen species(ROS)**(Kabel, 2014)**.

Herbal medicines have recently attracted much attention as alternative medicines useful for treating or preventing life style related disorders. There has been a growing interest in the analysis of plant products which have stimulated intense research on their potential health benefits**(Mishra*et al.*, 2015)**.Natural antioxidants contained in edible or medicinal plants often possess strong antioxidant and free radical scavenging abilities**(Li *et al.*, 2015)**.

GA is an edible, dried gummy exudate from the stems and branches of *Acacia Senegal*and *Acacia seyal*, that is rich in non-viscous soluble fiber. It is widely used in pharmaceutical and

food industry as an emulsifier and stabilizer, and in some countriesin the traditional treatment of patients with chronic kidneydisease**(Ali*et al.*, 2013)**.
VE(α-tocopherol) is considered to be beneficial for prevention of diseases associated with oxidative stress because of its remarkable anti-oxidative properties. **(Li *et al.*, 2015)**.It is the most important lipid-soluble antioxidant and protects cell membranes against oxidation **(Kabel, 2014)**.

The aim of the present work was to assess the antitoxic effect of Gum Arabic and vitamin E in hepatotoxicity and nephrotoxicity induced by over doses of Cinnamaldehyde in male albino rats.

Material and methods

The present study was carried out at the Zoology Department, Faculty ofWomen for(Art, Science& Education)Ain Shams University.The experimental animals were 60 youngadultsmale*Wistar* albino rats, weighing about 70-90 g, obtained from the Medical research center and bilharzial researchFaculty of Medicine, University of Ain Shams. Animals were kept to be acclimatized for at least two weeks prior to initiation of experimentation. They were kept under good ventilation, balanced diet and maintained at a temperature of 25 ±5°C. Food and water were allowed *ad libitum*. All animals were handled and treated according to the guidelines for animal experiments which were approved by EthicalCommittee of Medical Research of the National Research Centre, Cairo, Egypt.

Animals were divided into six groups ten rats each according the following regimen:-

1-Normal control group,orally received distilled water.
2- CNMA group at zero time,orally received dose 73.5 mg/kg b. wt. of CNMAdissolved in distilled water daily for 3 months.
3- CNMA group at 30 days orally administrated with a dose of CNMA 73.5 mg/kg b. wt. daily for three months followedwithout any treatment for another 30 days as a recovery period.
4- CNMA+GA therapeutic group received oral doses of CNMA 73.5 mg/kg b. wt. daily for three months then orally administered GA at a dose 7.5 g/kg b. wt. daily for another 30 days.
5- CNMA+VE therapeutic group received oral doses of CNMA 73.5 mg/kg b. wt. daily for three months then orally administered VE at a dose 1g/kg b. wt. daily for another 30 days.
6- CNMA+Mixture therapeutic group received oral doses of CNMA 73.5 mg/kg b. wt. daily for three months then orally administered mixture of GA and VE at doses 7.5 g/kg b. wt. and 1g/kg b. wt. of GA and VE respectively daily for another 30 days.

Chemicals and drugs

All chemicals were obtained from Sigma-Aldrich Chemical Company.

Sample collection

At the end of treatment, the animals were sacrificed by slaughtering. Blood samples were collected through carotid artery, in a clean centrifuge tube and left at room temperature in an

oblique position for two hours to coagulate. After complete retraction of the clot, the blood was centrifuged at 3000 r.p.m for 15 min. Serum was separated and stored at -20ºC until biochemical analysis.

Biochemical studies:

Serum ALT and AST were determined according to the method described by **Murray(1984)**. and GGT by the method of **Gendler (1984)**. Blood urea was determined according to the method of **Fawcett and Scott (1960)**. Serum creatinine was determined according to the method of **Seeling and Wust (1969)**.Total protein in serum was determined colorimetrically according to **Henry *et al.* (1974)**. Serum albumin level was estimated colorimetrically according to **Doumas *et al.* (1971)**. In liver tissue GSH was determined by **Tietze (1969)**, MDA was determined according to **Botsoglou *et al.*(1994)**, SOD level according to the method of **Nishikimi *et al*. (1972)**.

Histological studies

Kidney tissue wascut and fixed in 10% buffered formalin solution **(Lillie, 1954)**then processed for H X Estaining for histological studies.

Molecular study including Comet assay

The Comet Assay, also called single cell gel electrophoresis (SCGE), is a sensitive and rapid technique for quantifying and analyzing DNA damage in individual cells **(Sanadi *et al.*,2011)**.Liver tissue were separated and homogenized for oxidative stress and molecular studies.

Statistical analysis

All data were analyzed using the SPSS for windows software, version 10.0. Analysis of variance (ANOVA) which is an indication of the dispersion or difference between more than two means to the calculated standard error of this difference was assessed **(Tello and Crewson, 2003)**.

Results

Biochemical studies**of Cinnamaldehyde(CNMA)**

The data presented in **Table (1)** show a significant increase in serum ALT, ASTand GGT activities in the CNMA group at zero time. This increase reached 65.88%, 74.50% and 159.82 %, respectively as compared to the control group.

The results in **Table (2)** revealed that serum total protein and albumin concentrations decreased significantly (-36.70% and -37.46%) in the CNMA group as compared to the control group.

The data in **Table (2)** also showed a significant increase in serum urea, creatinineand uric acid levels in the CNMA rats at zero time as compared to control group, where the percentage of changes were123.07%, 353.89% and 148.63in serum urea, creatinine and uric acid levels, respectively.

GSH and SOD declined significantly and recorded –34.08% and -40.18%, respectively,whileHighly significant increase was observed in tissue MDA which reached 91.31% in the CNMA rats at zero time as compared to the control group **(Table 3)**.

Treatment of the CNMA rats with GA, VE ortheir mixture showed a significant improvement in serum ALT, AST and GGT activities and the change reached 30.60%, 16.36% and 24.78% with GA, 27.69 %, 26.27% and 30.29% with VE, 20.06%, 5.58% and 12.61% with the mixture, respectively as compared to the control group. Serum total protein and albumin concentrations changed significantly by-9.43%, -9.27% with GA, -14.12%, -14.83% with VE and -3.59%, -5.93% with mixture. Serum urea, creatinine and uric acid levels also changed significantly by 27.41%, 90.45% and87.61% with GA, 36.87%, 110.67%, and 77.93% with VE, 15.34%, 33.71% and 48.59% with mixture when compared to the control group.MDA changed by 23.13%, 25.37%, 18.28% with GA, VE and their mixture, respectively as compared to the control group.On the other hand,GSH and SOD showed a significant improvement and changed by-9.05%, -13.61% with GA, -5.94%, -9.99% with VE and-2.44%, -8.01% with the mixture, respectively as compared to the control group.
These biochemical results are confirmed with comet test as shown in **Table (4)**.

Histological studies: -

Control group

Normal histological structure of the kidney was observed inthe control sections (Fig. 1). The kidney architecture is composedof a large number of nephrons each containing renal corpuscles and renal tubules. The renal corpuscles contain a vascular capillaryglomerulus that is enclosed by Bowman' capsule. Also, the renal tubule is connected the renal corpuscle to a large collected duct and composed of proximal and distal tubules.

Specimens of animals administrated daily dose of CNMA 73.5mg/ kg b.wt. for three months revealed marked histological lesions particularly in cortical portion. The Malpighian corpuscles lost their characteristic normal configuration and exhibited clear features of damage. The glomerular tufts were contracted (Fig. 2).

Examination of the kidney sections ofrats treated with CNMA by a dose 73.5mg/ kg b.wt. for three monthsthen left for 30 days without any treatment revealed severe alterations within some renal tubules where the epithelium cells were replaced by flattened cells. Focal areas of necrosis were also noticed **(**Fig. 3).

Rats treated with CNMA for 3 months followed by oraltreatmentwith GumArabic at a dose 7.5g/ kg b.wt. for 30 days revealed minimal changes in the structure of the kidney (Fig. 4) compared to normal ones. It showed vaculation in glomerular tuft and persistence of degenerative alteration in proximal and distal convoluted tubules.

Conspicuous feature of deleterious alterations was observed in the kidney sections of rat treated with CNMA for 3months followed by oral treatment with Vitamin E at a dose 1g/

kg b.wt. for 30 days. The glomeruli showed contracted capillaries with contraction and lobulation of glomerular tuft. Besides, some vacuolar areas of relatively variable sizes were frequently detected in the deformed glomeruli (Fig. 5).

The histological examination of sections of kidney from rats treated with CNMA for 3months followed by oral treatment with mixture of GumArabic and Vitamin E for 30 days revealed minimal changes in the structure of the kidney (Fig. 6) compared to normal ones. Both glomeruli and convoluted tubules showed limited regenerative changes.

Molecular studies: -

Comet assay

On measuring the comet %, the data tabulated in **Table (4)** and graphically represented in **Figure (7)** were obtained. The control rats designated more or less constant figures during the study period,the comet percent recorded 10%. On the other hand, in CNMA group a strong elevation was realized in comet percent as compared with the control group. This elevationreached22.9% and 14.6% at zero time and 30 days respectively. **(Table 4 and Fig 8,9)**.A considerable improvement was observed in CNMA+GA therapeutic group in comet percent. This improvement recorded12%after 30 days of treatment**(Table 4 and Fig 10)**.Also, minimal improvement occurred in the comet% of CNMA+VE therapeutic group as compared with the control group. The percentage was 13%**(Table 4 and Fig 11)**.On the other hand, strong improvement occurred in the comet % of therapeutic treatedrats by mixture of GA and VE scoring 11.8%after 30 days of treatment**(Table 4 and Fig 12)**.

Discussion

CNMA is considered one of the most commonly used food flavoring in many types of food while a high dose of it may induce undesirable effects**(Gowder and Halagowder, 2010**).On the other hand, GA has interesting antioxidant properties such as an efficient capacity for moderated radical scavenging capacity and deactivation of excited electronic states**(Montenegro*et al.*, 2012)**.Also, Vitamins are vital nutrients with diverse biochemical functions that are essential for maintaining health, VE is the most important lipid-soluble antioxidant and protects cell membranes against oxidation **(Kabel, 2014)**.The present study was undertaken to assess the antitoxic effect of Gum Arabic and vitamin E in hepatotoxicity and nephrotoxicity induced by CNMA in male albino rats.

In the present study a significant increase in serum ALT, ASTand GGT activities were observed in the CNMA rats at zero time beside a significant elevation in serum urea, creatinine and uric acid. The antioxidants biomarkers GSH and SOD declined significantly, while MDA level increased significantly in CNMA rats at zero time as compared to the control group. These results are in agreement with**Gowder and Halagowder (2010)** whomentioned that serum of the test rats (rats treated with CNMA at dose level of 73.5 mg/kg body weight/ day for 3 months) showed imbalance in the antioxidant status, which induced epoxidizing state in the rat liver. Another study, CNMA induced lipid peroxidation during free radical scavenging action, ascorbic acid is suggested to be transformed into semidehydro

ascorbate,reduced glutathione is required for the conversion of semidehydroascorbate back to ascorbate **(Niknahad *et al.*, 2003)**. Other studies noted that ROS directly impair mitochondrial functionand cellular repair mechanisms,oxidative stress is already found in early stages of renal disease and increases with declining kidney function**(Perazella and Moeckel, 2010)** and**(Ali et *al.*, 2013)**.**Gowder (2013)** noted that CNMA induced renal damage, is due to the ROS formed while in the free radical scavenging reactions.

Treatment of the CNMA rats with GA, VE or their mixture revealed a significant decrease in serum ALT, AST, GGT concentrations and a significant increase in total protein and albumin concentrations. Marked improvements were achieved in urea, creatinine and uric acid levels, which declined significantly after treatment with GA, VE or their mixture. Highly significant increase was observed in GSH and SOD, while marked decrease in tissue MDA was recorded in all therapeutic groups, especially with mixture group. These results corroborate the findings of **Omer *et al.* (2013)**who found that GA was widely used by Eastern folk medicine practitioners as a restorative agent and is thought to be an excellent cure for renal failure patients.Theyobserved that the concentration of ALT and AST were significantly higher in the group treated with *Aristolochia bracteata* when compared to the control, while GA treated groups recorded lower concentration of the same parameter. Also, **Al-Yahya*et al.* (2009)**found that oral administration of GA significantly palliates CCl4 induced hepatotoxicity in mice and nephrotoxicity in rats. Another study, reported that the pretreatment with GA prevented the increase of plasma urea and creatinine **(Ali*et al.*, 2015)**. Similarly, it was reported that GA was shown to reduce urinary nitrogen excretion by increasing urea disposal in the cecum and lower serum urea concentration in rats and humans**(Ali *etal*., 2013)**.Moreover,**Montenegro *et al.* (2012)** reported that GA has interesting antioxidant properties such as an efficient capacity for deactivation of excited electronic states and moderated radical scavenging capacity.**Al Za'abi*et al.* (2015)** mentioned that concomitant treatment with GA significantly reduced the inflammatory and oxidative stress induced by the administration of intraperitoneal adenine. **Venkatanarayana *et al.* (2012)** observed that simultaneous supplementation of vitamin E was found to ameliorate the renal toxicity and oxidative stress induced by CCl4 and caused a significant increase in renal GSH, SOD and a significant reduction in renal MDA in experimental exhaustive stress. In another report using mice treated with VE, a progressive decrease in MDA levels was detected **(Caetano *et al.*, 2013)**. VE is known to have antioxidant properties capable of scavenging free radicals, which have critical roles in radiation injuries as observed by**WahbaandIbrahim(2013)**,**SinghandKrishnan (2015)**.

Histological section of renal tissues in control group showed normal architecture of kidney.On the other hand, the present study indicated that CNMA induced congestion of glomerular capillaries, mild degenerative changes with appearance of fibrin casts in tubules and occurrence of high mitotic activity, focal areas of necrosis, degenerative changes within epithelial lining cells of proximal and distal convoluted tubules. Similar results, were reported by **Gowderand Devaraj(2006)**.Tissue infiltration of white blood cells as a sign of inflammation was observed on histological examination of kidneys sections of CNMA-treated animals, which was significantly suppressed in animals treated with GA**(Ali *et al*.,2013)**.GA

may work on the nephron to cause nephroprotection. Nephron is a structural protein that is expressed on the surface of glomerular podocytes and is critical in maintaining selective permeability and preventing proteinuria **(Ali et *al.*, 2010)**.Moreover, GA exert anti-inflammatory effects by interacting with intestinal dendritic cells, which are in direct contact with the intestinal lumen and decreaseits phagocytic activity**(Nasir, 2013)**.The present study indicated that VE ameliorated renal tissue injuries to mimic normal figureswith well-developed Bowman's capsule with enlarged glomerulus and convoluted tubules. Similar resultswere observed by **Derakhshanfar*et al.* (2007)**and **Stojiljković*et al.* (2014)** who noticed that VE treatment in rats prevented the renal lesions with gentamicin while moderate tubular changes were observed.Similar results were obtained after CNMA rats treated with mixture of GA+VE.

Additionally, under oxidative stress cells display various dysfunctions due to lesions caused by ROS to DNA, proteins and lipids**(Al-Attar, 2011)** ˙OH radicals react with the DNA bases resulting in impaired DNA, causing several physiological conditions such as mutagenesis and carcinogenesis**(Nimseand Pal,2015)**. The present study found that CNMA - induced hepatic cell death were characterized with changes in nuclear morphology, DNA fragmentation, and cell morphology, and it was able to induce apoptosis in a concentration-dependent manner. Similar resultswere observed by**Stammati*et al.* (1999)**and**Huang*et al.* (2007)** who reported thatthe lipophilic nature of CNMA suggests that DNA damage could follow primary damage on the membrane allowing the diffusion of the compound to the nuclear region. Treatment with GA enhanced the concentration of nuclear DNA-binding protein that had many functions dependent on its location in the cell and contribute to normal tissue development and repair (**Ali *et al.*, 2015)**. GA also enhanced the growth of intestinal normal flora and the metabolites of these microorganisms such as volatile fatty acids and polyamines that play an effective role in DNA and RNA protein synthesis regulation and enhancing proliferation of the epithelial cell **(Freigoun*et al.*, 2015)**. On the other hand,ratstreated with VE showed marked improvement in their cell structure.Similar resultswere observed by **Agarwal *et al.* (2010)**who examined the effective role of vitamin E against Mercury Hg induced acute toxicity in rats that resulted in oxidative injury and metallothionein mRNA expression.Also, the present study indicated that CNMA rats treated with mixture of GA+VE produced a strong improvement in comet parameters which near to normal values as compared with the control group.

Conclusion

There are numerous sources of free radicals that, in excess, may have deleterious effects on the human body. This study suggests that GA and VE have protective effects on oxidative stress caused hepatic and renalinjury induced by overdose of CNMA. The protective effect of the mixture is more potent than those of GA or VE only.

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Explanation of figures

Fig. (1): Light micrographs of control rat kidney, showing the glomerulus(G), two layers of Bowman's capsule; outer and inner visceral layers and urinary space. Notice theproximal (P) and distal(D) convoluted tubules.

Fig. (2): Photomicrograph of a section of kidney of rats treated with CNMA by a dose of (73.5mg/ kg b.wt.) for three months showing, contraction of the glomerular capillaries and fatty degenerative changes within glomeruli.

Fig. (3): Photomicrograph of a section of kidney of rats treated with CNMA by a dose of (73.5mg/ kg b.wt.) for three months then left for 30 days without any treatment revealing swelling, vaculation and necrosis of epithelial cells of distal convoluted tubules (D). Notice the degeneration of the proximal convoluted tubules (P).

Fig. (4): Photomicrograph of a section of kidney of rats treated with CNMA by a dose of (73.5mg/ kg b.wt.) for three months followed by oral treatment with GumArabic at a dose of (7.5g/ kg b.wt.) for 30 days showing vacuolation in glomerular tuft and persistence of degenerative alteration in proximal and distal convoluted tubules.

Fig. (5): Photomicrograph of a section of kidney of rats treated with CNMA by a dose of (73.5mg/ kg b.wt.) for three months followed by oral treatment with Vitamin E at a dose of (1g/ kg b.wt.) for 30 days showing erosion of bruch border of the proximal convoluted tubules.

Fig. (6): Photomicrograph of a section of kidney of rats treated with CNMA by a dose of (73.5mg/ kg b.wt.) for three months followed by oral treatment with mixture of GumArabic and Vitamin E for 30 days showing limited regenerative changes in kidney tissue.

Fig. (7): Comet of control rats designated more or less constant figures during the study period.

Fig. (8): Rats treated with CNMA by a dose of (73.5mg/ kg b.wt.) for three months,strong elevation was realized in the comet appearance as compared with the control group.

Fig. (9): Rats treated with CNMA by a dose of (73.5mg/ kg b.wt.) for three months then left for 30 days without any treatment.Ther is No improvement was observed.

Fig. (10): Comet of rats treated with CNMA by a dose of (73.5mg/ kg b.wt.) for three months followed by oral treatment with GumArabc at a dose of (7.5g/ kg b.wt.) for 30 days.A Considerable improvement was observed in GA therapeutic group in the comet appearance.

Fig. (11): Comet of rats treated withCNMA by a dose of (73.5mg/ kg b.wt.) for three months followed by oral treatment with Vitamin E at a dose of (1g/ kg b.wt.) for 30 days.Minimal

improvement occurred in the comet appearance of VE therapeutic group as compared with the control group.

Fig. (12): Comet of rats treated withCNMA by a dose of (73.5mg/ kg b.wt.) for three months followed by oral treatment with mixture of GumArabic and Vitamin E for 30 days.Strong improvement occurred in the comet appearance of therapeutic rats.

Table (1): Therapeutic role of GumArabic and Vitamin E on serum ALT (u/l), AST(u/l) and GGT(u/l) levels in control and experimental groups.

Parameters	Group / Duration	Control group	CNMA group	CNMA+GA group	CNMA+VE group	CNMA+mixture group
ALT (U/L)	Zero time	25±0.28	41.47±0.44 %65.88 a**			
	30 days	26±0.30	37.46±20 %44.08 ab**	33.96±0.25 %30.60 abc**	33.20±0.25 %27.69 abc**	31.22±0.16 %20.06 abc**
AST (U/L)	Zero time	57.85±0.75	100.95±0.43 %74.50 a**			
	30 days	59.27±0.29	92.89±0.34 %56.73 ab**	68.97±0.86 %16.36 abc**	74.84±0.32 %26.27 abc**	62.58±0.20 5.58 abc**%
GGT (U/L)	Zero time	3.26±0.03	8.47±0.02 %159.82 a**			
	30 days	3.41±0.04	6.86±0.03 %101.2 ab**	4.26±0.03 %24.78 abc**	4.44±0.03 30.29 abc**%	3.84±0.02 %12.61 abc**

Data are expressed as means ± S.E. (n = 10 in each group).

a: Significant change at p< 0.05 with respect to control group at zero time.

a: Significant change at p< 0.05 with respect to control group at 30 days.

b: Significant change at p< 0.05 with respect to CNMA at zero time- group.

c: Significant change at p< 0.05 with respect to CNMA at 30 days- group.

* Highly significant change at p < 0.01. ** Very Highly significant change at p < 0.001.

Table (2): Therapeutic role of GumArabic and Vitamin E on serum total protein, Albumin, urea, creatinine and uric acid levels in control and experimental groups.

Parameters	Group / Duration	Control group	CNMA group	CNMA+GA group	CNMA+VE group	CNMA+mixture group
T. Protein (g/dl)	Zero time	6.14 ± 0.01	3.95 ±0.02 %-35.67 a**			
	30 days	6.23 ± 0.01	4.219 ± 0.011 %-32.32 ab**	5.65±0.01 %-9.43 abc**	5.35±0.01 %-14.12 abc**	6.01±0.04 %-3.59 abc**
Albumin (g/dl)	Zero time	4.25±0.04	2.85±0.02 %-32.94 a**			
	30 days	4.55±0.02	3.32±0.03 %-27.07 ab**	4.13±0.03 %-9.27 abc**	3.88±0.03 %-14.83 abc**	4.28±0.02 %-5.93 abc**
Urea (mg/dl)	Zero time	30.25±0.19	67.48±0.26 %123.07 a**			
	30 days	32.51±0.20	56.61±0.27 %74.11 ab**	41.43±0.20 %27.41 abc**	44.50±0.28 %36.87 abc**	37.50±0.18 %15.34 abc**
Creatinine (mg/dl)	Zero time	0.167±0.004	0.758±0.003 %353.89 a**			
	30 days	0.178±0.003	0.546±0.005 %206.74 ab**	0.339±0.003 %90.45 abc**	0.375±0.004 %110.67 abc**	0.238±0.005 %33.71 abc**
Uric acid (mg/dl)	Zero time	1.02±0.007	2.536±0.007 %148.63 a**			
	30 days	1.03±0.006	2.14±0.008 %107.45 ab**	1.94±0.006 %87.61 abc**	1.84±0.006 %77.93 abc**	1.54±0.006 %48.59 abc**

Data are expressed as means ± S.E. (n = 10 in each group).

a: Significant change at p< 0.05 with respect to control group at zero time.

a: Significant change at p< 0.05 with respect to control group at 30 days.

b: Significant change at p< 0.05 with respect to CNMA at zero time- group.

c: Significant change at p< 0.05 with respect to CNMA at 30 days- group.

* Highly significant change at p < 0.01. ** Very Highly significant change at p < 0.001.

Table (3): Therapeutic role of GumArabic and Vitamin E on Glutathione (GSH) ((n.mol/g protein), Superoxide dismutase (SOD) (u/g) andmalondialdhyde (MDA) (n.mol/g)levels in control and experimental groups.

Parameters	Group / Duration	Control group	CNMA group	CNMA+GA group	CNMA+VE group	CNMA+mixture group
GSH (n.mol/g)	Zero time	3.11±0.01	2.05±0.01 %-34.08 a**			
	30 days	3.03±0.01	2.26±0.01 %-25.45 ab**	2.85±0.01 %-5.94 abc**	2.76±0.01 %-9.05 abc**	2.96±0.01 %-2.44 abc**
SOD (u/g)	Zero time	76.50±0.15	45.76±0.23 %-40.18 a**			
	30 days	75.71±0.34	54.41±0.27 %-27.99 ab**	68.14±0.12 %-9.99 abc**	65.40±0.32 %-13.61 abc**	69.64±0.21 %-8.01 abc**
MDA (n.mol/g)	Zero time	4.03±0.01	7.71±0.01 %91.31 a**			
	30 days	4.15±0.01	6.81±0.01 %64.06 ab**	5.11±0.01 %23.13 abc**	5.20±0.01 %25.37 abc**	4.91±0.01 %18.28 abc**

Data are expressed as means ± S.E. (n = 10 in each group).

a: Significant change at p< 0.05 with respect to control group at zero time.

a: Significant change at p< 0.05 with respect to control group at 30 days.

b: Significant change at p< 0.05 with respect to CNMA at zero time- group.

c: Significant change at p< 0.05 with respect to CNMA at 30 days- group.

*Highly significant change at p < 0.01.**Very Highly significant change at p < 0.001.

Table (4): Therapeutic role of GumArabic and Vitamin E oncomet test in control and experimental groups

Group	Comet %	Head Diameter (µm)	%DNA in Head	Tail Length (µm)	%DNA in Tail	Tail Moment
control	10%	16.1	65.9	6.4	34.1	2.2
CNMA at zero time	22.9%	12.4	55.7	9.2	44.3	4.1
CNMA at 30days	14.6%	12.8	56.6	8.1	43.4	3.5
CNMA+GA	12%	13.7	63.1	6.8	36.9	2.5
CNMA+VE	13%	13	57.7	7.5	42.3	3.2
CNMA+mixture	11.8%	14.9	64.3	6.7	35.7	2.4

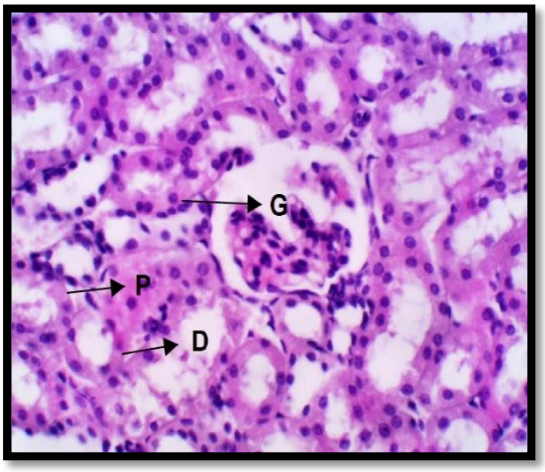


Fig. 1

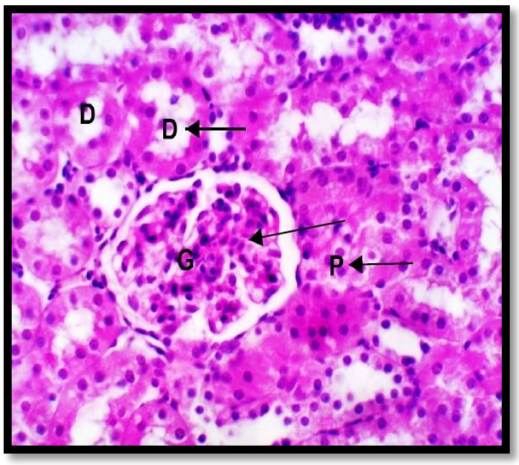


Fig. 2

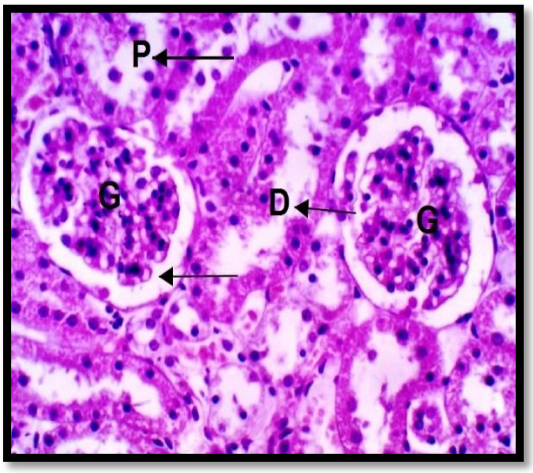


Fig. 3

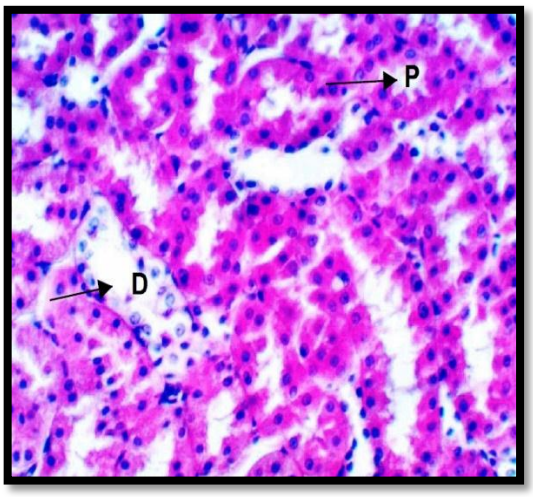
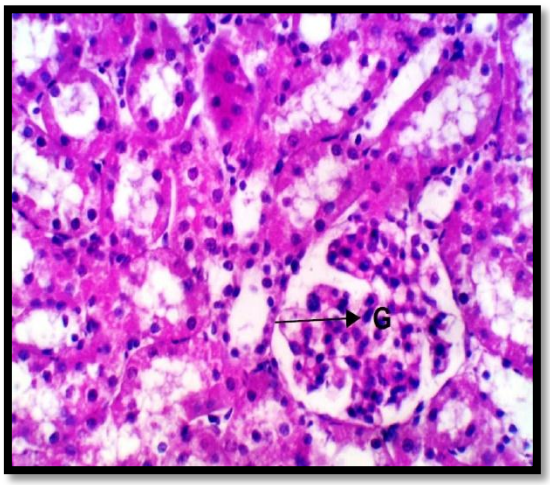


Fig. 4



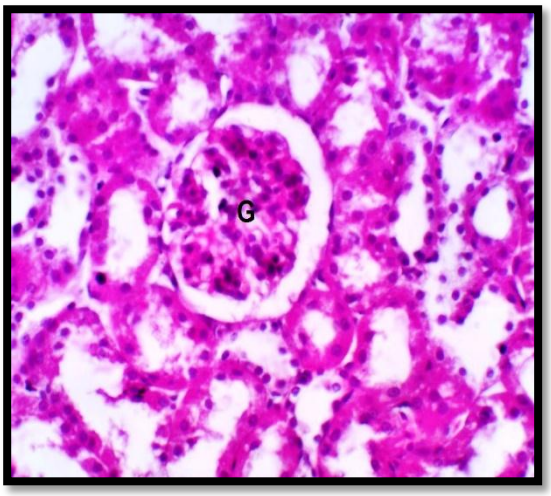


Fig. 5 **Fig. 6**

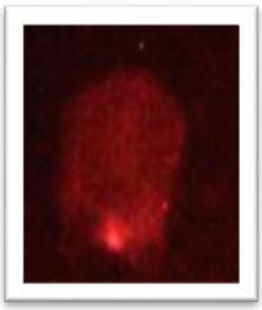
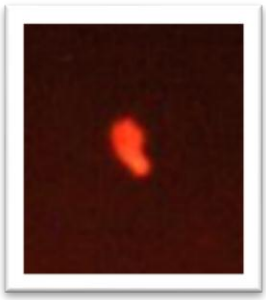
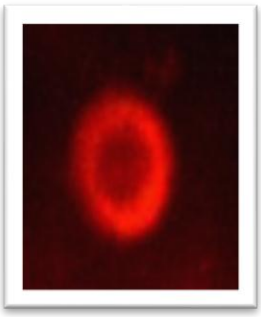


Fig. 7 **Fig. 8** **Fig. 9**





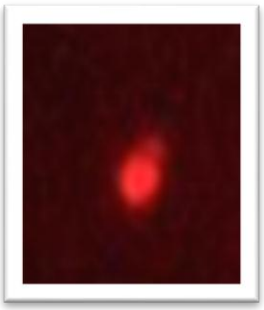


Fig. 10 **Fig. 11** **Fig. 12**

Visual scoring of DNA damage according to comet appearance

الملخص باللغة العربية

الدور الفعال للصمغ العربى وفيتامين (ه) على تسمم الكبد والكلية المحدث بمكسبات الطعم السينمالدهيد فى صغار ذكور الجرذان البيضاء

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جامعة عين شمس –قسم علم الحيوان – كلية البنات للآداب والعلوم والتربية

تعتبر مادة السينمالدهيدنوعاً من مكسبات الطعم التى تتواجد فى طعام الإنسان بنسبة بسيطة جداً، وهى تصنع كيمائياً بواسطة تكثيف البنزالدهيد مع الأسيتالدهيد وهى بذلك تنتمى لأنشط المواد السامة كيمائياً، ومن المحاولات التى أجريت للحماية من هذه السموم إستخدام الصمغ العربى وهو منتج طبيعى مع فيتامين (ه) واستخدامهما كمضادات أكسدة . وقد تم تقسيم ستين جرذاً من الجرذان البيضاءإلى ستة مجاميع حيث احتوت كل مجموعة على عشرة جرذان بحيث تمثل المجموعة الأولى و التى أعطيت عن طريق الفم ماء مقطر (المجموعة الضابطة). وأعطى الخمسون جرذاً الباقينعن طريق الفم جرعات يومية من مادة السينمالدهيد المذابة فى ماء مقطر تعادل٧٣.٥ مجم/كجم لمدة ثلاثة أشهر ثم قسموا إلى خمسمجموعات: مجموعة السينمالدهيد عند الصفرومجموعة السينمالدهيد عندالیوم الثلاثين و قد تركت بدون أى معالجة لمدة ثلاثين يوماًأخربومجموعة الصمغ العربى + السينمالدهيدوعولجتعن طريق الفم بجرعات يومية منالصمغ العربى تعادل ٧.٥ جم/كجم لمدة ثلاثين يوماًأخربو مجموعةفيتامين (ه) + السينمالدهيد وعولجتعن طريق الفم بجرعات يومية من فيتامين (ه)تعادل١جم/كجم لمدة ثلاثين يوماًأخربو مجموعة الصمغ العربى +فيتامين (ه) + السينمالدهيدوقد عولجت عن طريق الفم بجرعات يومية من خليط منالصمغ العربى تعادل ٧.٥ جم/كجم+ فيتامين (ه) تعادل١جم/كجم لمدة ثلاثين يوماًأخرى. وقد أجريت بعض الدراسات البيوكيمائية والنسيجية و الجزيئية فى نهاية التجربة وقد أظهرت النتائج أن مادة السينمالدهيد أدت إلى نقص ملحوظ فى مستويات الألبيومين و البروتين وأدت الى زيادة ملحوظة فى مستويات اليوريا و الكرياتينين ووظائف الكبد. بينماأظهرت المعالجة بالصمغ العربى وفيتامين (ه) تحسناً واضحاً فى جميع التحاليل السابقة مقارنةً بالمجموعة الضابطة. أما تحاليل مضادات الأكسدة أظهرت نقصاً فى محتويات الجلوتاثيونو إنزيم السوبر أكسيد ديسميوتاز و زيادةً فى الأكسدة الفوقية للدهون (المالوندهيد) فى مجموعتى السينمالدهيد (المجموعة الثانية والثالثة) بينما باقى المجاميع فقد اقتربت النتائج من المجموعة الضابطة. وقد تم تأكيد هذه النتائج بالدراسات النسيجية للكلية وتحليل الكوميت لنسيج الكبد وأوضحت الدراسة أن مادة السينمالدهيد تسببت فى أضرار على نسيج الكلية وخاصة منطقة القشرة حيث أظهرت كريات مالبيجي تغيرات فى مظهرها الطبيعى . ومن جانب آخر زادت نسبة الكوميت بهذه المادة مقارنةًبالمجموعة الضابطة بينما حدث تحسناً واضحاً فى باقى المجاميع العلاجية. وبذلك تكون النتائج قد أوضحت أن للصمغ العربى وفيتامين(ه) دوراً فعالاً فى تقليل أضرار مادة السينمالدهيد على كلية وكبد الجرذان البيضاء.