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Possible positive effect of gum Arabic against the toxicity of the drug furosemide on newborn rats

Abstract. Medicinal plants received special attention due to their biological and medicinal activities, aspects of safety in their use, and low cost. This study aimed to prove gum Arabic (GA) extract's preventive and therapeutic role against furosemide toxicity. Moreover, histological findings, renal functions, and the level of MDA and GSH in the serum and kidney tissues of newborn rats were assessed. Thirty pregnant rats were divided into six groups (n = 5 per group). The first group is the control group, with no treatment. The second is the GA group, administered 15% w/v GA in drinking water daily from conception day 0 until the end of pregnancy. The third group is the furosemide group; furosemide (20 mg/k, ip) was taken daily from conception day 0 until the end of pregnancy. The fourth group is the protective group (preventive group); GA was taken daily (15% w/v in drinking water) from conception day 0 to day 10, followed by furosemide (20 mg/kg, ip) daily until the end of pregnancy. The fifth group is the therapeutic group; furosemide (20 mg/kg, ip) was taken daily from conception day 0 to day 10, followed by GA (15% w/v in drinking water) daily until the end of pregnancy. The sixth group is the mixed group; GA and furosemide were administered together from conception day 0 until the end of pregnancy.
Key words: gum arabic; furosemide; kidney failure; loop diuretics; newborn rats.

Introduction

Loop diuretics are widely utilized pharmaceutical agents in medicine and are considered the most potent diuretics [1]. The loop diuretics have been regarded as a significant advancement in medical treatment. Their effectiveness is particularly notable in patients with insufficient response to alternative medications, including individuals with chronic impaired kidney function and severe cardiovascular disease [2].

According to epidemiologic studies, oliguria increases the risk of death. Moreover, based on observational studies, many intensivists employ loop diuretics, particularly furosemide [3].

The documented mechanisms of sodium-potassium-chloride cotransporters suggest that loop diuretics exert vasodilatory effects inside the renal system [4]. Loop diuretics can potentially decrease metabolic demands and oxygen utilization in renal tubular cells, thereby protecting renal function under ischemic conditions [5].

Loop diuretics are organic anions with a negative charge, exhibiting limited lipid solubility. This characteristic enables them to firmly bind to serum albumin, with a binding capacity of over 95 percent. Consequently, their filtration at the glomerulus is restricted, leading to a reduction in their bioavailability. For instance, the bioavailability of furosemide is roughly 50 percent [6].

Furosemide may be classified as a weak organic acid. The primary route of elimination for this substance is renal excretion, accounting for around 85% of its clearance. Roughly half of the substance is metabolized, while the other half is actively secreted in its original form by the organic acid transporters in the proximal tubules [7].

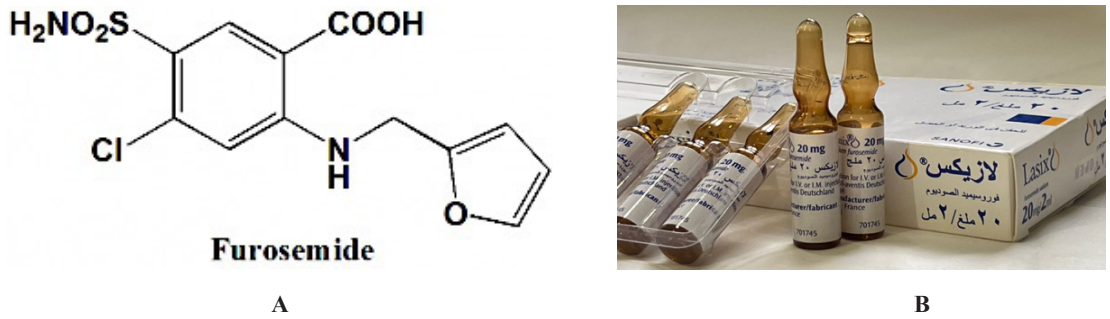
Furosemide has the potential to mitigate and lessen the intensity of renal damage. Similarly, furosemide may assist in controlling excessive fluid volume by enhancing sodium excretion and promoting diuresis [3].

Furosemide can be taken orally as tablets or as an oral solution and can be inhaled as well. The efficacy of intravenous furosemide is twice that of oral furosemide [8]. Furosemide is synthesized by a significant number of manufacturers located in Bulgaria, Brazil, China, Israel, Hungary, Italy, Switzerland, Poland, USA [9].

The furosemide chemical formula is 4-chloro-N-(2-furyl methyl)-5-sulfamoyl-anthranilic acid [10]. Figure 1A shows the structure formula of furosemide,

and a commercially available furosemide medication, named Lasix, is shown in Figure 1B.

Furosemide exerts its pharmacological effects by targeting the medullary thick ascending loop of Henle, where it inhibits the activity of the $Na^+/K^+/Cl^-$ pump located on the luminal cell membrane. This action mechanism can decrease the demand for oxygen in the renal tubules. Furosemide is further recognized for its significant adjunctive function in preserving fluid balance and enhancing nutritional provision in critically sick patients with acute kidney injury (AKI) [11].



A B

Figure 1 – Structural formula of furosemide (A) and a commercially available furosemide medication Lasix (B)

There is a correlation between the presence of furosemide in plasma and the presence of albumin. The protein-bound fraction of furosemide is transported into the proximal tubule epithelial cells by the bloodstream and then translocated to its target locations, which are the lumens of the ascending limbs of Henle's loop tubules [12].

Moreover, the kidneys regulate fluid and sodium balance, which is seen in the case of kidney failure, where fluid retention is evident [13].

However, furosemide has adverse effects, including hepatic insufficiency, diabetes, hypoproteinemia, and some infrequent symptoms such as indigestion, thirst, dizziness, dehydration, drowsiness, weakness, muscle cramps, tinnitus, and deafness [14].

In the case of individuals diagnosed with cirrhosis, furosemide is often deliberated as a treatment option. Nevertheless, its use is associated with certain risks, including the potential for severe muscle contracture and the depletion of skeletal muscle mass, as indicated in a prior study [15]. Notably, furosemide was categorized as a Pregnancy Category C medication, warranting caution among healthcare providers when prescribing it to pregnant women. This caution stems from findings in animal reproductive experiments revealing the transfer of furosemide across the placenta and its propensity to induce adverse effects [16].

Intensive diuretic therapy can potentially lead to a reduction in breastfeeding. In cases where nursing a newborn or a premature infant is involved, it might be advisable to consider an alternative medication. Markedly, administering furosemide at lower doses may not necessarily hinder or suppress lactation [16].

Furthermore, several studies have reported that the administration of furosemide in preterm newborns has been shown to significantly influence the development and advancement of renal calcification and nephrolithiasis, often referred to as the formation of renal calcifications and stones. It causes sensorineural hearing loss and is more common in premature babies [17].

Numerous research investigations have shown the extensive use of GA and many traditional therapeutic practices. These studies have revealed various advantageous effects associated with Gum Acacia or Gum Arabic (GA; here is shown on Figure 2), which encompasses health-related, aesthetic, nutritional, and industrial aspects [18]. It refers to a desiccated gum fluid often obtained by incisions made on the stems and branches of acacia trees, namely those belonging to the *A. senegal* and *A. seyal* species within the Fabaceae family. The gum exudate is commonly found in the shape of tears, spheres, or semi-spheres [19]. *A. senegal* is a drought-tolerant tree that grows natively in desert, subtropical, and semi-arid climates. It is a tiny, spiky shrub that grows 2-6 m or even 12 m tall from the ground [20].

Figure 2 – General view of GA

GA comprises glycoproteins, proteins that include a carbohydrate co-factor or prosthetic group, and polysaccharides. The predominant constituents of these polysaccharides are galactose and arabinose. GA is widely recognized as an environmentally sustainable and nutritionally viable option [21, 22].

The scientific classification of GA places it in the plant kingdom within the *Fabaceae* family (subfamily *Mimosoideae*), belonging to the *Acacia* genus, with the primary species being *Acacia senegal* and *Acacia seyal* [19].

Materials and methods

Chemicals. In this investigation, furosemide, a member of the loop diuretic class, was employed. Furosemide was procured as commercial vials (Lasix® 20 mg, Sanofi-Aventis Deutschland GmbH, Germany) and administered intraperitoneally at 20 mg per kilogram, following established precedents in scientific literature.

Gum Arabic. GA, obtained in solid spheres, was sourced commercially from a perfumery establishment in Khartoum, Sudan. These spheres constitute pure extracts derived from the *Senegalese acacia* tree and do not contain any additives. The specimens underwent a grinding process and subsequent preparation in potable water, with a concentration of 15% (Figure 3), following procedures delineated in previous studies.

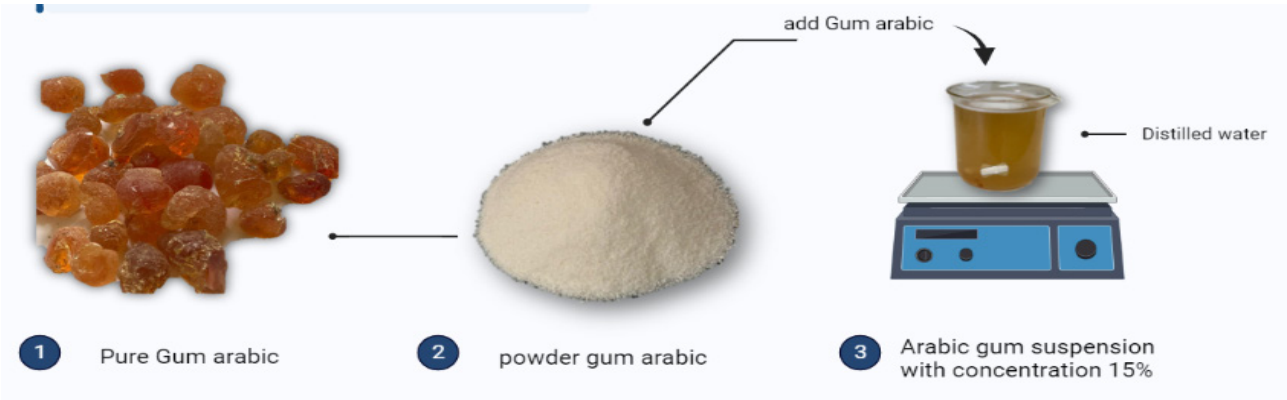


Figure 3 – Preparation of GA suspension

Experimental animals. The research employed pregnant Wistar albino rats, specifically those displaying albinism, as the subjects for experimentation. These rats were initially assessed for weight, demonstrating an average weight ranging from 200 to 250 grams at the study's outset. The specimens were sourced from the animal housing facility associated with the Faculty of Pharmacy at King Abdulaziz University. The rats were housed under carefully controlled laboratory conditions, specifically in plastic cages, where the temperature was maintained at 20 ± 22°C. They were subjected to a light-dark cycle, each lasting 12 hours, and the light phase commencing at 07:00 AM. Throughout the experiment, the rats had unrestricted access to food and water (Figure 4).

Ethical approval. The animal studies in this research adhered to the ethical guidelines established by the Committee on Bioethics for Animal Studies at the College of Pharmacy, King Abdulaziz University. Trial registration number: PH-1443-64 and date of registration: 1/6/2022.

Figure 4 – Experimental setup for housing of rats under controlled laboratory conditions

Experimental design. The implemented experimental design is summarized on Figure 5. The animal subjects were categorized into six distinct groups as follows:

1. Control Group (G 1): No specific treatment was administered.

2. Gum Arabic Group (G 2): GA (15% w/v in drinking water) was administered daily from conception (day 0) throughout the entire pregnancy period [23, 24].

3. Furosemide Group (G 3): Furosemide (20 mg/kg, intraperitoneal) was administered daily from conception (day 0) until the conclusion of the pregnancy [25].

4. Protective (Preventive) Group (G 4): GA was administered daily (15% w/v in drinking water) from conception (day 0) until day 10, followed by daily administration of furosemide (20 mg/kg, intraperitoneal) until the end of pregnancy.

5. Therapeutic Group (G 5): Furosemide (20 mg/kg, intraperitoneal) was administered daily from conception (day 0) until day 10, followed by daily administration of GA (15% w/v in drinking water) until the end of pregnancy.

6. Mixed Group (G 6): GA and furosemide were administered concurrently from conception (day 0) until the conclusion of the pregnancy period.

Morphological studies. At the age of two weeks, an assessment of the newborns was conducted across all groups. The evaluation aimed to determine mortality rates, examine the external characteristics of the newborns, and measure parameters including body weight, length, and kidney weight in the treated groups, with comparisons made against the control group for analysis.

A laboratory digital balance was used to measure the body weight and kidney weight of newborn mice. A meter was used to measure the body length of newborn mice.

Histological studies. Kidney samples were obtained from juvenile rats (aged two weeks) and immediately fixed in a 10% formalin solution. Following fixation, the specimens underwent a series of standard processing steps involving alcohol and xylol treatment, ultimately leading to their embedding in paraffin. Subsequently, the paraffin-embedded samples were sectioned into slices with a thickness ranging from 4 to 6 micrometers. These slices were then subjected to staining using the hematoxylin and eosin (H&E) stain method, facilitating the examination of histopathological changes and alterations within the kidney tissue.

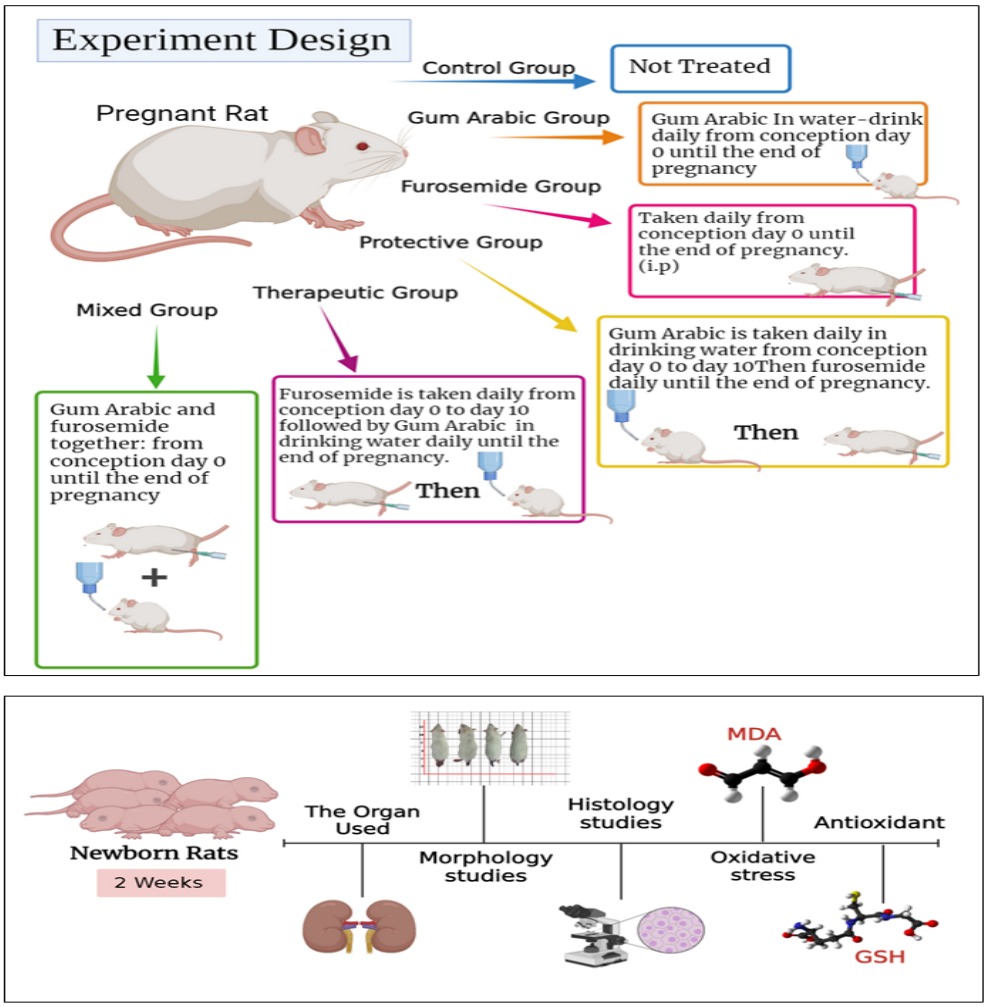


Figure 5 – Design of the experiment

Biochemical analysis. Malondialdehyde (MDA) levels were assessed to evaluate the impact of oxidative stress on both kidney tissue and serum. An enzyme-linked immunosorbent assay (ELISA) kit designed for precise quantitative detection of Rat MDA was employed. The procedure involved introducing MDA monoclonal antibodies into pre-coated wells, enabling the addition of MDA. After incubation, a biotin-conjugated anti-Rat MDA antibody was introduced and bound to Rat MDA.

Glutathione (GSH) levels were also measured to assess kidney tissue and serum antioxidant levels. In this study, an ELISA kit was utilized to determine the concentration of GSH in the serum and tissue of the rats.

Statistical analysis. Statistical analysis involved the calculation of the mean values for both the control group and the treatment groups. Furthermore, the standard deviation of these means was determined, and a T-test was performed at a significance level of 5% to assess the statistical significance of observed differences. The statistically significant data ($P < 0.05$) compared to the control group were denoted as "a," while statistically significant data ($P < 0.05$) compared to the furosemide group were denoted as "b."

Results and discussion

Morphological studies. Mortality and survival rate. Table 1 shows the number of births, deaths, and lived births as well as the mortality rate for the control group, the Arabic gum group, the furosemide group, the preventive group, the treatment group, and the mixed group at the age of two weeks. The data in Table 1 revealed an elevated mortality rate

within the mixed group compared to the control group. The results also indicated no statistically significant difference in the total number of deliveries among the GA, furosemide, protective, therapeutic, and mixed groups compared to the control group (Figure 6).

Table 1 – The number of births, deaths, lived births, and mortality rates for all groups at the age of two weeks

Group number	Number of births	Number of deaths	Number of live births	Mortality rate (%)
G1	26	0	26	0
G2	26	0	26	0
G3	25	0	25	0
G4	27	0	27	0
G5	24	9	15	37.5
G6	24	4	20	16

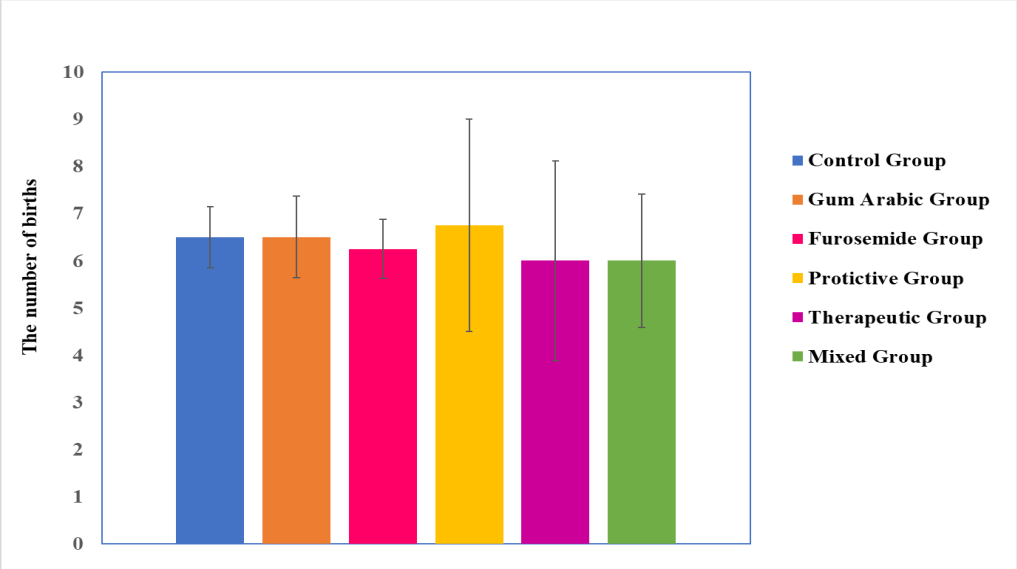


Figure 6 – The number of births for all studied groups

Malformations. In the context of our study, several noteworthy observations were made concerning malformations associated with different treatment groups. Firstly, an increased incidence of preterm labor was observed in both the furosemide and the mixed treatment groups, indicating a potential association between these treatments and the risk of preterm birth. Secondly, delayed hair growth was noted in individuals receiving furosemide and mixed treatment, suggesting a potential adverse effect on fetal development. Moreover, infants born to mothers in the furosemide and mixed groups exhibited smaller birth sizes than those in the control group, indicating a possible impact on fetal growth. Additionally, our findings indicated an elevated occurrence of bleeding in pregnant mothers within the preventive group, warranting further investigation into the safety and efficacy of the preventive treatment. Lastly, it is crucial to highlight the unfortunate occurrence of maternal mortality in the therapeutic group.

Body weight and body length. Compared to the control group, the weight of newborn rats in the GA and the treatment groups increased significantly in the second week, whereas weight decreased significantly in the furosemide and mixed groups.

The group administered with GA and the therapeutic group displayed a notable and statistically significant increase in body weight. In contrast, compared to the furosemide-treated group, the mixed group exhibited a significant reduction in body weight (Figure 7).

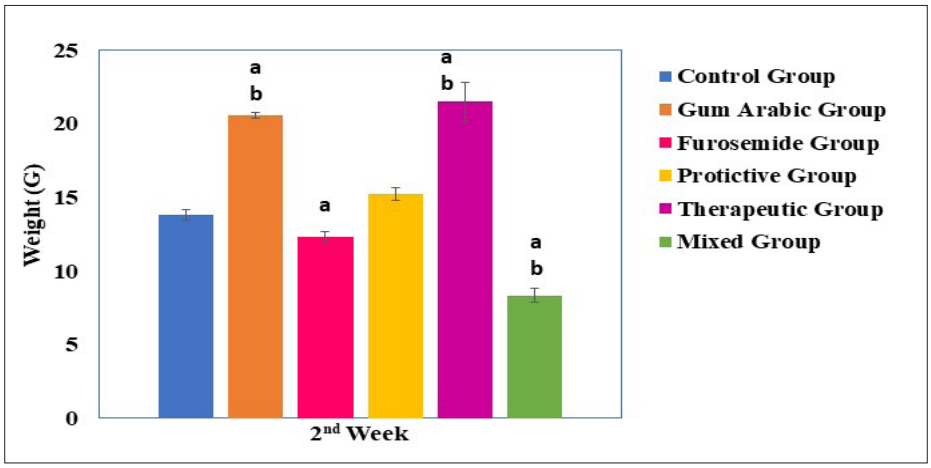


Figure 7 – The newborn rats' body weight (g) for all studied groups

In the second week of the study, noteworthy findings emerged regarding the length measurements among the various groups. The group administered with GA displayed a statistically significant increase in length. In contrast, both the group treated with furosemide and the mixed group exhibited significant decreases in length compared to the control group.

Furthermore, the group subjected to GA treatment, as well as both the preventive and treatment groups, exhibited statistically significant increases in length. Conversely, a significant decrease in height was observed in the mixed group when compared to the cohort receiving furosemide treatment (Figure 8).

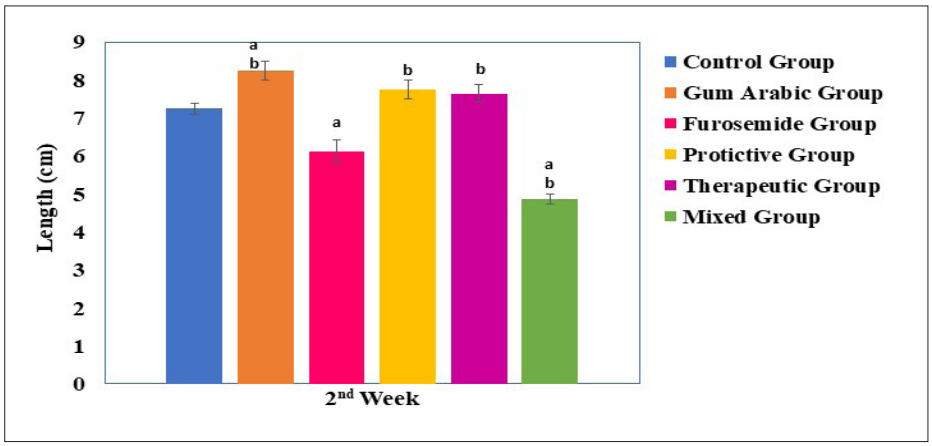


Figure 8 – The newborn rats' length (cm) for all studied groups

Figure 9 shows photographs of rats from all studied groups with their corresponding body weights and body lengths after two weeks.

Kidney weight. The study's findings indicated a statistically significant increase in kidney weight in both the group treated with GA and the therapeutic group. In contrast, a notable decrease in kidney weight was recorded in the mixed group compared to the control group. Furthermore, the study revealed a substantial increase in kidney weight within the group that received GA treatment and the therapeutic group. Conversely, a significant decrease in kidney weight was observed in the mixed group compared to the group subjected to furosemide treatment (Figure 10).

Figure 11 shows photographs representing the change in the kidney weights among all studied groups after two weeks.

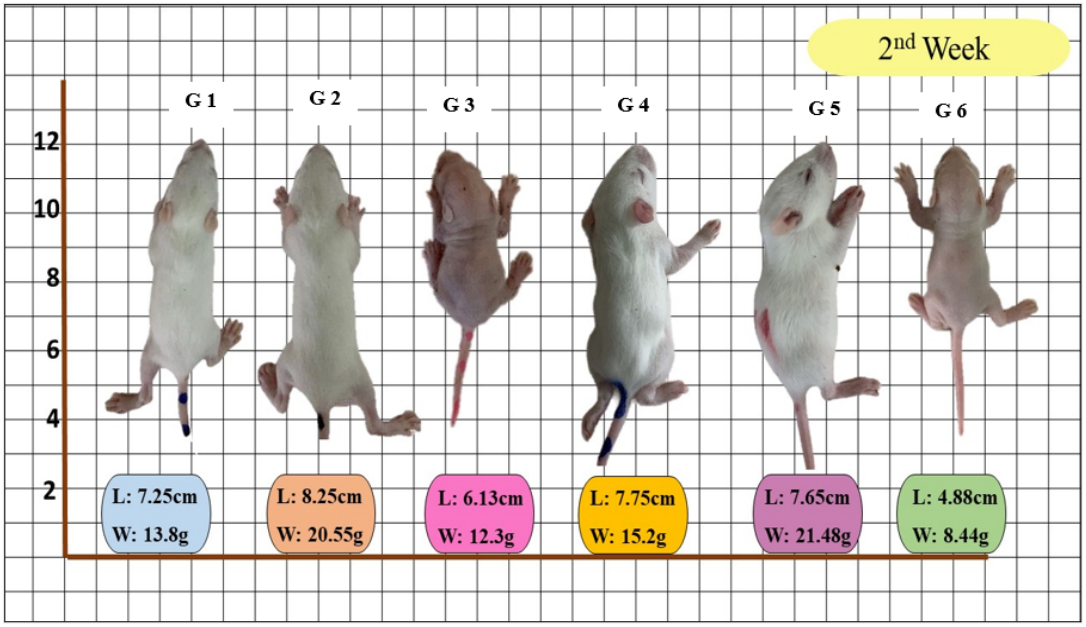


Figure 9 – Photographs of rats from all studied groups after two weeks

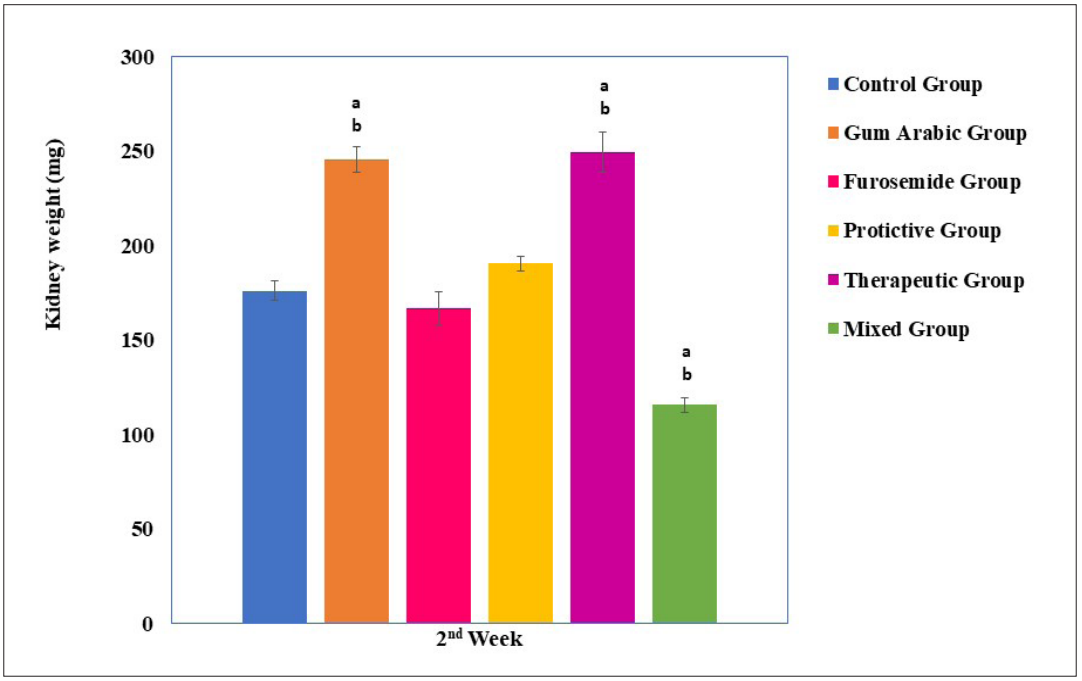


Figure 10 – The newborn rats' kidney weight (mg) for all studied groups

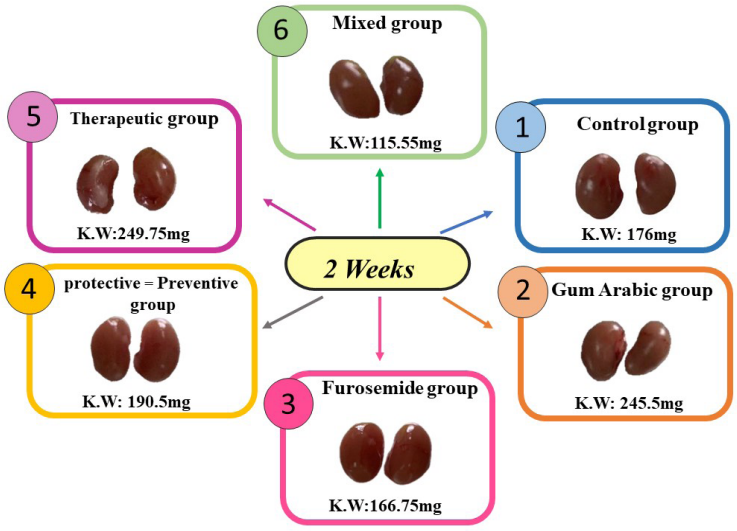


Figure 11 – Photographs of the kidneys of rats from all studied groups after the two weeks.

Histological studies. The kidney cortex of newborn rats in the control group showed normal kidney structure and tissue and a complete absence of interstitial fibrosis. The normal histological appearance of the kidney consists of the glomerulus, tufts of the glomerulus, Bowman's capsules, proximal tubule, and distal tubules (Figure 12-1). The renal cortex of the kidneys of newborn rats in the GA group was not damaged by the treatment with GA and was similar to the control group in histological structure (Figure 12-2). In contrast, the renal cortex of newborn rats in the furosemide group exhibited several notable histological changes associated with administering furosemide. These changes included the dilation of Bowman's space and increased cell pigmentation intensity. Significantly, the impact on the glomeruli exceeded that on the renal tubules, characterized by severe glomerular shrinkage, resulting in a loss of both structural integrity and function. Conversely, the effect on the renal tubules appeared relatively straightforward, with observable spaces between the tubules and the presence of fatty droplets. This appearance of droplets is a commonly recognized indicator of tissue damage in many tissues (Figure 12-3). In contrast, rats in the protective group exhibited signs of histological improvement, as evidenced by the relatively healthier appearance of the renal tubules. Notably, the damage manifested as a reduced Bowman's area size and a moderate separation of the glomerular tufts (Figure 12-4).

The rats displayed histological improvement in the therapeutic group, notably in preserving glomerular integrity. The Bowman's space exhibited a crescent shape, and the glomerular tufts remained intact. However, indications of damage were observed at the level of the renal tubules, characterized by cytoplasmic decomposition and the presence of hemorrhage (Figure 12-5).

In contrast, the rats in the mixed group exhibited distinct histological differences in their kidney tissues. Notably, there were alterations in the shape of the glomeruli, with evidence of atrophy and fragmentation in some of the glomerular tufts. A noticeable degree of cytoplasmic decomposition was observed at the renal tubule level (Figure 12-6).

Note: (1) Control group, showing normal kidney architecture and histology. (2) Gum Arabic group, showing normal kidney architecture and histology. (3) Furosemide group, showing several effects on the histological structure of the kidney: ( ) space expansion in Bowman space, ( ) the intensity of the pigmentation of the cells, ( ) the appearance of spaces between the installation of renal tubules, and (<) severe shrinkage of many glomeruli and pyknotic. (4) Preventive group, an effect appeared on the histological structure of the kidney. (*) Relative decrease in space around the glomerulus (Bowman's distance). Many glomeruli and renal tubules appear intact. (5) Therapeutic group, the glomeruli appear intact, the crescent-shaped void appears and the tufts intact, damage appears on the tubes, (0) where cytoplasmic decomposition appears, and (□) hemorrhage. (6) Mixed group, several effects appeared on the histological structure of the kidney. (>) A few of the glomeruli showed shrinkage and nuclear atrophy, and many of them were intact. ( ) Protrusion of the nuclei into the tubular cavity and (*) relative fragmentation of some of the glomerulus tufts.

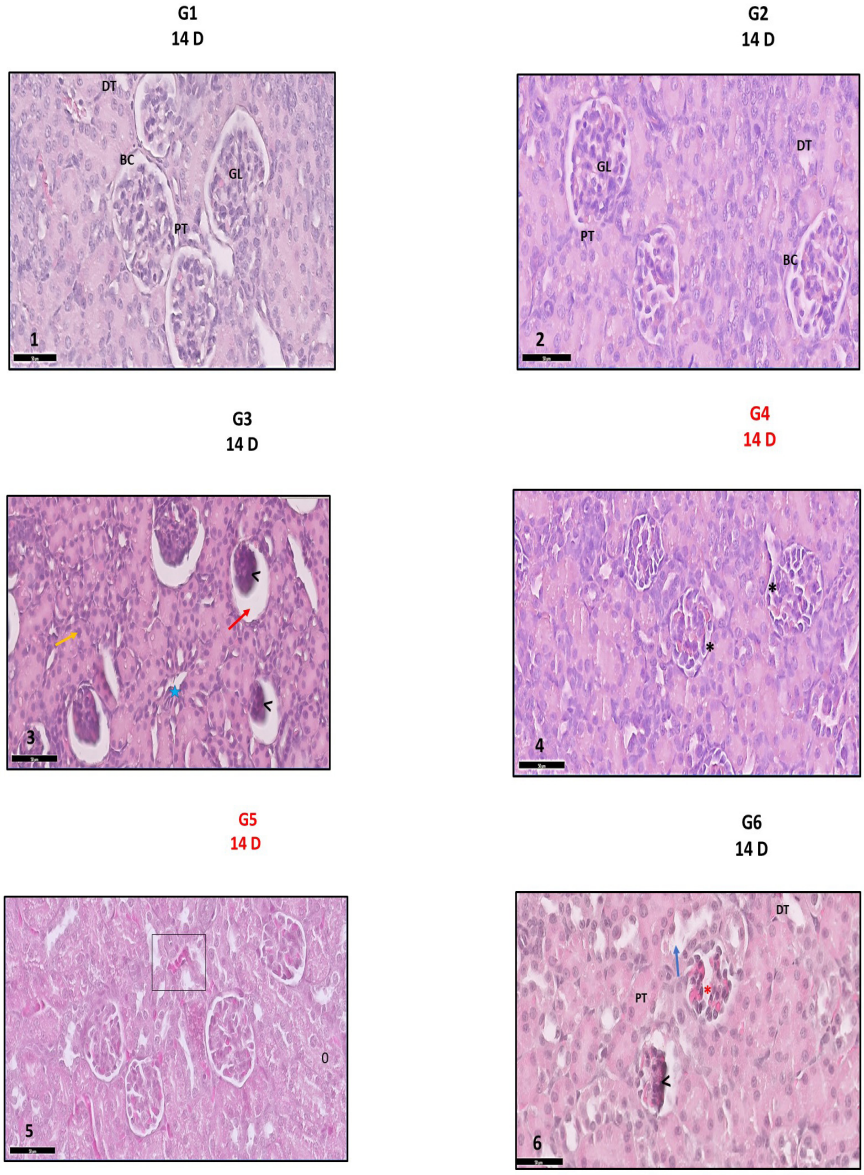


Figure 12 – Representative photograph of renal tissue under a light microscope of newborn rats at age 2 weeks. Hematoxylin and eosin staining (H&E). Scale bars: 50 μm

Malondialdehyde (MDA). The study's findings revealed noteworthy variations in the serum MDA levels across the different treatment groups during the second week of observation. Specifically, a significant elevation in serum MDA levels was observed in the group receiving furosemide treatment and in the preventive and therapeutic groups compared to the control group. Conversely, a significant reduction in serum MDA levels was noted in the group treated with GA, along with the preventive, therapeutic, and mixed groups, when contrasted with the group subjected to furosemide treatment (Figure 13).

The study's findings unveiled notable alterations in tissue MDA levels among the various treatment groups during the second week of investigation. Specifically, compared to the control group, a significant increase in tissue MDA levels was observed in the group treated with furosemide and the preventive group. In contrast, there was a significant reduction in tissue MDA levels noted in the mixed group compared to the control group.

Additionally, the results indicated a significant decrease in tissue MDA levels in the GA, preventive, therapeutic, and mixed groups during the second week compared to the group treated with furosemide (Figure 14).

Glutathione (GSH). The findings indicated a notable reduction in serum GSH levels during the second week in the group administered furosemide and in the preventive and therapeutic groups compared to the control group. Conversely, the results documented a substantial elevation in serum GSH levels within the group treated with GA, the preventive group, the therapeutic group, and the mixed group during the second week when contrasted with the furosemide-treated group (Figure 15).

The findings revealed a marked reduction in tissue GSH levels during the second week in the group treated with furosemide and in the preventive and therapeutic groups compared to the control group. In contrast, a significant increase in tissue GSH levels was observed in the group treated with GA, the preventive group, the therapeutic group, and the mixed group during the second week compared to the furosemide-treated group (Figure 16).

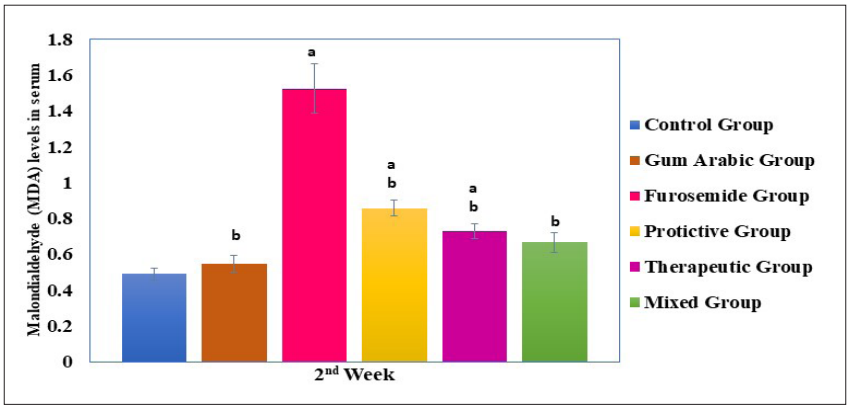


Figure 13 – The malondialdehyde (nmol/ml) levels in the serum of newborn rats for all studied groups.

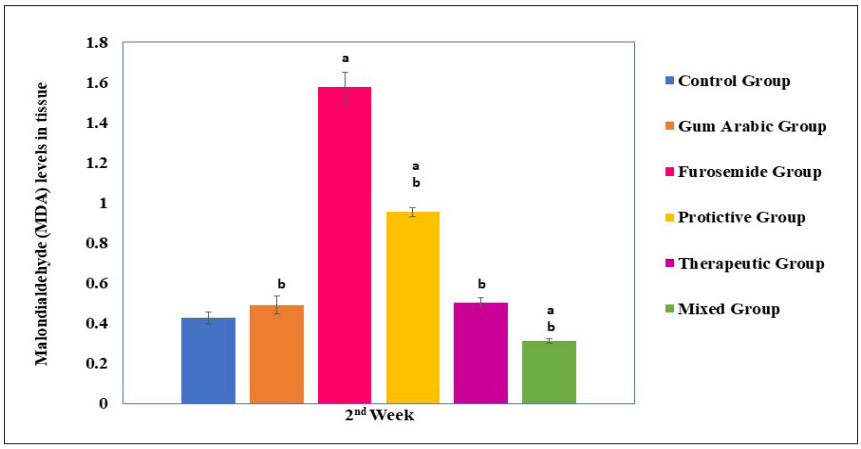


Figure 14 – All studied groups' malondialdehyde levels in newborn rats' kidney tissue.

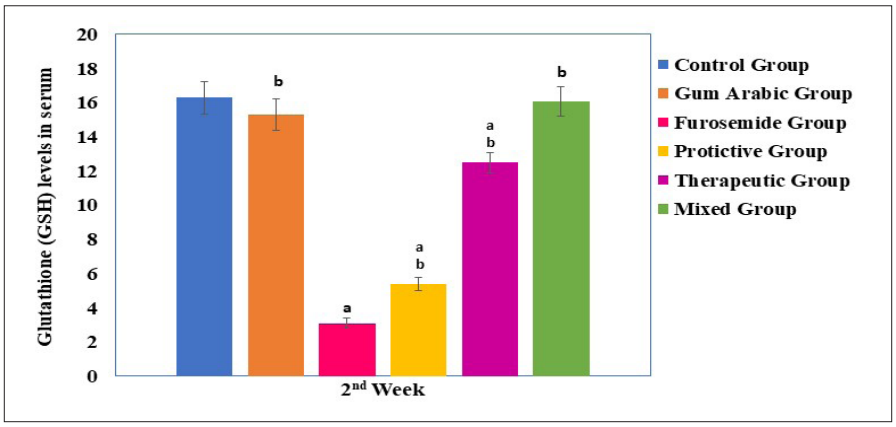


Figure 15 – The glutathione level in the serum of newborn rats for all studied groups

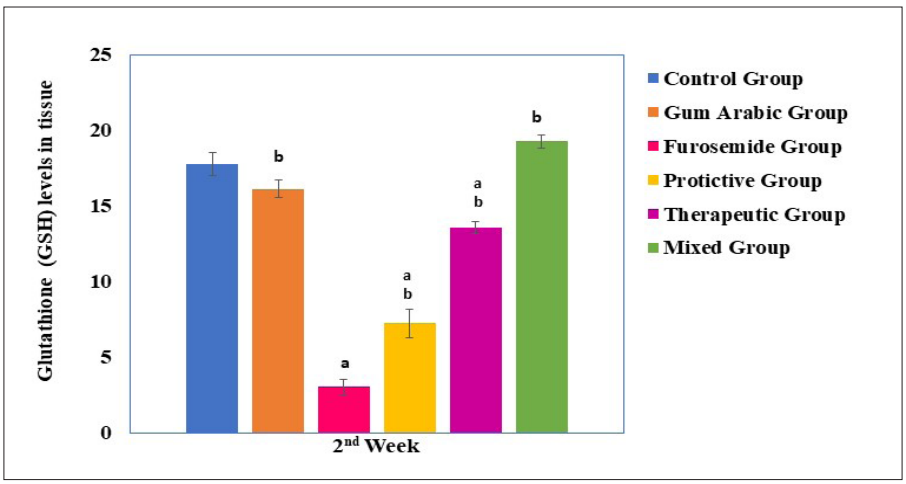


Figure 16 – Glutathione levels in the kidneys of newborn rats for all studied groups

As demonstrated in a rat model investigation conducted by Rokutan and colleagues [26], chronic administration of furosemide is associated with fatal outcomes. Furthermore, in a study involving elderly patients, furosemide contributed to increased mortality rates, thus underscoring the significance of this factor for healthcare providers [27].

In alternative investigations, loop diuretics, notably furosemide, have been employed during pregnancy to address conditions such as pulmonary edema, severe hypertension in the context of chronic kidney disease, or congestive heart failure, despite the associated risk of hyperbilirubinemia in newborns. Additionally, it has been demonstrated that loop diuretics can traverse into breast milk, potentially impeding breastfeeding [28].

Animal reproduction investigations have revealed unfavorable fetal effects when diuretics, specifically furosemide, are administered during pregnancy. The findings indicated a higher incidence of premature birth within both the furosemide and mixed groups, in contrast to the control group. Additionally, instances of maternal bleeding were observed in the preventive group.

Nakatsuka and colleagues, in their study from 1988 [29], reported that administering furosemide via intraperitoneal injection to pregnant rats leads to the relaxation of uterine muscles, potentially resulting in various fetal deformities. Furthermore, the administration of diuretics during pregnancy was found to be linked to differences in fetal birth weight and the incidence of preterm delivery [30].

The present investigation revealed a significant reduction in both body weight and length during the second week in the group treated with furosemide compared to the control group.

Our findings align with prior research [13, 31], which reported that the administration of furosemide reduced body weight and decreased extracellular water due to increased diuresis. This is consistent with the observed positive correlation between the decrease in extracellular water and body weight. Additionally, our results concur with the findings from other studies [32, 33].

The study demonstrated a noteworthy increase in both body weight and length during the second week in the group treated with GA, in contrast to the control group. This finding is consistent with the results reported in another study [34].

The study revealed a substantial increase in kidney weight during the second week in the group treated with GA compared to the control group.

Upon histological examination of the kidney composition in the control group, it is evident that the cortex region contains numerous renal or urinary corpuscles and renal tubules. Each renal corpuscle comprises a glomerulus, characterized by a tuft of capillaries resting on the basement membrane of the glomerulus and Bowman's capsule. The Bowman's capsule consists of two distinct layers: the outer parietal layer, lined with simple flat squamous cells featuring thin nuclei and a delicate basement membrane, and the inner visceral layer. The visceral layer is lined with large circular cells containing prominent, circular, basal-shaped nuclei known as foot cells resting on the outer surface of the glomerulus's capillaries. The space between these two layers, the parietal and visceral layers, is termed the urothelial space. Within the cortical region, the urinary tubules can be differentiated into proximal convoluted tubules, characterized by cuboidal cells with central nuclei and a narrow lumen, and distal convoluted tubules. The distal convoluted tubules are lined by cuboidal cells with apical nuclei, and their lumens appear broader and more irregular. These findings follow prior observations reported in other studies [35-37].

The tissue composition in the group treated with GA exhibited similarities to that of the control group, which aligns with the findings reported in reference [38].

The tissues in the group treated with furosemide exhibited notable effects on the histological structure of the kidney. These effects included the expansion of Bowman's space and an increase in cell pigmentation intensity. The impact on the glomeruli was more pronounced than the renal tubules, as there was significant shrinkage of the glomeruli, resulting in a loss of their normal shape and function. The effect on the tubules was relatively straightforward, characterized by the presence of spaces between the renal tubules and the appearance of fatty droplets. These findings are consistent with the observation that furosemide adversely affects organs, particularly the kidneys.

Dilken et al. 2023, as reported in their study [39], asserted that furosemide induces damage to both the renal cortex and medulla, elevates the risk of renal injury and diminishes oxygen levels even under control conditions.

In a study conducted by Araujo et al. in 2020 [40], they documented that furosemide impacts the renal tubules of rats. This effect can be elucidated by considering that the thin tubule represents the location of the highest flow resistance within the Henle loop.

The findings from both the preventive and therapeutic groups indicated that renal tissue was affected to a lesser extent than the group treated with furosemide yet to a greater extent than the group treated with GA.

According to Hammad and colleagues in 2019 [41], the impact of GA on inflammatory markers and oxidative stress was associated with robust histological protective attributes.

As reported by Said and colleagues in 2019 [42], the utilization of GA effectively alleviated renal damage. This beneficial effect can be attributed to the antioxidant and anti-inflammatory properties of GA.

In his 2019 review [43], Jaafar reported that GA had substantiated its pharmacological effects and therapeutic advantages in numerous studies conducted across various diseases.

The preventive effect of GA on renal function significantly reduces renal function markers in patients with diabetic renal failure and exerts a direct antioxidant effect [44].

In the mixed group (comprising treatment with both GA and furosemide simultaneously), noticeable differences were observed in the shape of the glomeruli, including atrophy and fragmentation of certain glomerular tufts. Furthermore, relative cytoplasmic degeneration was noted at the renal tubule level. However, these effects were milder than those observed in the group treated with furosemide alone and more pronounced than those observed in the preventive and therapeutic groups.

In another study, it was reported that GA helps preserve the normal structure of the renal cortex, except for some tubules showing degeneration and expansion [42].

MDA is a critical marker for assessing oxidative levels in biological fluids and tissues. It is an organic

compound considered one of the foremost indicators of lipid peroxidation, particularly polyunsaturated fatty acids [45].

The findings revealed a substantial increase in MDA levels in both the kidney tissues and serum of the group treated with furosemide compared to the control group.

As elucidated by Silbert and colleagues in their 2019 study [46], furosemide diminishes the metabolic activity in the ascending loop of Henle. This, in turn, can result in an enduring elevation of renal oxidative stress via multiple mechanisms. Furosemide's inhibition of glomerular tubular feedback leads to increased nephron blood flow and filtration, necessitating elevated solute transport. Consequently, the stimulation of renin release may further contribute to oxidative stress. Therefore, it is affirmed that using furosemide acts as a catalyst for oxidative stress.

Studies have consistently demonstrated that urinary furosemide administration increases renal oxidative stress in all patients, with the most severe cases of acute kidney injury (AKI) exhibiting the most significant increase. Notably, in this study, the group treated with GA showed no statistically significant change in MDA levels in the second week following birth compared to the control group.

GSH is a significant antioxidant enzyme within the body, serving as a vital defender of cells. It plays a pivotal role in facilitating the conversion of GSH from its oxidized form to the reduced form, thereby contributing to apoptosis regulation and cell development. Furthermore, GSH assumes a crucial protective function in the detoxification processes in various organs, including the liver, kidneys, lungs, intestines, and epithelia. The GSH redox cycle offers valuable insights into the mechanisms underlying injuries caused by toxic substances, diseases, or the regulation of redox-sensitive pathways within organisms [47-49].

The results indicate a significant decrease in the level of GSH in both kidney tissue and serum of the furosemide-treated group compared to the control group. However, in the group treated with GA in this study, no statistically significant change was observed in GSH levels during the second week following birth compared to the control group.

In another study [50], it was demonstrated that the treatment with GA at doses of 2.5%, 5%, and 10% w/v in water did not result in any significant changes in the concentrations of GSH. This suggests that GA does not exert an antioxidant effect in the tissues of both treated and normal rats. Therefore, its potential beneficial effect in patients with chronic renal failure is unlikely to be attributed to an antioxidant action.

Ahmed et al.'s perspective, as presented in reference [51], contradicts the previously mentioned finding. According to Ahmed et al. 2015, the use of GA to prevent nephrotoxicity in chronic kidney disease is based on the belief that it contains a high percentage of antioxidants. This difference in interpretation highlights the ongoing debate and research regarding the exact mechanisms of GA's effects on kidney health.

Conclusion

A high dose of furosemide can result in toxicity, leading to renal and histopathological changes and disturbances in renal function tests in newborn rats. However, the administration of gum Arabic as a protective and therapeutic agent mitigates the toxic manifestations induced by furosemide.

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