Original Research

Effects of Gum Arabic in rats with adenine-induced chronic renal failure

Badreldin H Ali[1], Suhail Al-Salam[2], Isehaq Al Husseni[3], Rana R Kayed[1], Noura Al-Masroori[1], Thuriya Al-Harthi[1], Mohamed Al Zaabi[1] and Abderrahim Nemmar[4]

[1]Department of Pharmacology, College of Medicine and Health Science, Sultan Qaboos University, Muscat 123, Al-Khod, Sultanate of Oman; [2]Department of Pathology, Faculty of Medicine and Health Sciences, United Arab Emirates University, Al-Ain, United Arab Emirates; [3]Department of Physiology, College of Medicine and Health Science, Sultan Qaboos University, Muscat 123, Al-Khod, Sultanate of Oman; [4]Department of Physiology, Faculty of Medicine and Health Sciences, United Arab Emirates University, Al-Ain, United Arab Emirates

Corresponding author: Dr Abderrahim Nemmar, Department of Physiology, Faculty of Medicine and Health Sciences, United Arab Emirates University, PO Box 17666, Al-Ain, United Arabs Emirates. Emails: anemmar@uaeu.ac.ae, anemmar@hotmail.com

Abstract

Gum Arabic (GA [*Acacia senegal*]) is reputed, in Arabian medicinal practices, to be useful in treating patients with chronic renal failure (CRF), albeit without strong scientific evidence. We have previously shown that GA had no significant effect in rats with CRF induced by surgical nephrectomy. Here, we used another animal model of human CRF (feeding adenine at a concentration of 0.75%$^{w/w}$ for four weeks) to test the effect of GA on CRF. Renal morphology and measurements of plasma concentrations of urea and creatinine (Cr), and Cr clearance, in addition to urinary volume, osmolarity and protein concentrations, and *N*-acetylglucosamine and lactate dehydrogenase activities were performed. Interleukin-6 and the total antioxidant levels in urine, as well as the activity of superoxide dismutase in renal tissues, were estimated. Adenine feeding resulted in marked renal damage. GA (6%$^{w/v}$ and 12%$^{w/v}$ in drinking water for four consecutive weeks) significantly ameliorated the adverse biochemical alterations indicative of renal failure, abated the decrease in body weight and reduced the glomerular, tubular and interstitial lesions induced by adenine. Our study provides evidence that GA attenuated renal dysfunction in this model of CRF, suggesting a promising potential for it in protecting against renal failure progression. The mechanism(s) of this nephroprotection is uncertain but may involve anti-oxidant and/or anti-inflammatory actions.

Keywords: rats, chronic renal failure, adenine, Gum Arabic

Experimental Biology and Medicine 2010; **235**: 373–382. DOI: 10.1258/ebm.2009.009214

Introduction

The incidence of chronic renal failure (CRF) appears to be on the increase, especially in some developing countries, imposing a very expensive and rising demand on health-care systems already burdened by paucity of resources.[1] There is, therefore, an obvious need for alternative approaches that would reduce the cost of dialysis, drug therapy and renal transplantations. One such approach is the use of oral sorbents,[2–6] and the literature is replete with publications on the impact of nutrition on kidney disease.[2,3] Most dietary attempts to treat CRF and to decrease the consequent uremia used protein restriction regimens.[4,5] An alternative approach has recently been suggested by diet supplementation with fermentable carbohydrate.[6]

Gum Arabic (GA) is a fermentable carbohydrate derived from the dried exudates of *Acacia senegal*, and is widely used in both the pharmaceutical and food industries as an emulsifier and stabilizer (for a recent review see reference[7]). GA is used in the traditional medicinal practices in Middle Eastern countries. In Sudan particularly, GA is used to ameliorate the severity of CRF, and has been shown to reduce the frequency of dialysis from three to two times per week.[7,8] Treatment with GA has been claimed to result in a urea-lowering effect by increasing fecal urea nitrogen excretion, with a concomitant decrease in urinary total nitrogen in adults[9,10] and children.[11–13] Based on laboratory data,[14] extraction, modification and recycling of nitrogenous wastes by the gastrointestinal tract is a potentially low-cost means of substituting for missing renal function. However,

in a previous preliminary study,[8] we have shown that treatment of rats with GA (3 or 6 g/100 mL drinking water/day for five consecutive weeks) did not significantly affect the decrease in body weight or the increases in creatinine (Cr) and urea induced by CRF produced by subtotal nephrectomy.[15] In the present study, we have examined the effects of GA (6 or 12 g/100 mL drinking water/day for four consecutive weeks) on renal function impairment using a range of physiological, histological and biochemical variables in rats with CRF induced by feeding adenine, which is an established animal model of human CRF.[15,16]

Some of the findings presented in this article have been reported previously in the form of abstract at the meeting of the congress of the European Association for Clinical Pharmacology and Therapeutics, Edinburgh 2009.[17]

Materials and methods

This project was reviewed and approved by the Institutional Review Board of the Sultan Qaboos University, College of Medicine and Health Sciences, and experiments were performed in accordance with protocols approved by the Institutional Animal Care and Research Advisory Committee.

Animals

A total of 36 male Wistar rats (Taconic Farms, Germantown, NY, USA) aged six weeks and weighing initially 200–240 g were obtained from the SQU Small Animal House. They had free access to water and a feed composed of standard powder diet containing 0.85% phosphorus, 1.12% calcium, 0.35% magnesium, 25.3% crude protein and 2.5 IU/g vitamin D_3 (Oman Flour Mills, Muscat, Oman).

Chemicals

Adenine and GA powder were obtained from Sigma (St Louis, MO, USA) and were prepared freshly every day. Kits for measuring *N*-acetylglucosamine (NAG) activity were obtained from Diazyme Laboratories (General Atomics, San Diego, CA, USA), for Cr, urea, gamma glutryl transaminase and protein concentration from Human GmbH (Mannheim, Germany), for lactate dehydrogenase (LDH) from Roche Diagnostics (France), for superoxide dismutase (SOD) from Randox (Antrim, UK) and for total anti-oxidants from Cayman Chemicals (Ann Arbor, MI, USA).

Experimental design

After an acclimatization period of seven days, rats were randomly divided into six equal groups. The first group continued to receive the same diet without treatment until the end of the study (control group). The second group was switched to a powder diet containing adenine (0.75%$^{w/w}$ in feed) for four weeks. The third and fourth groups were given normal food and GA in drinking water at a concentration of 6%$^{w/v}$ and 12%$^{w/v}$, respectively, for four weeks. The fifth and sixth groups were given adenine in the feed as in group two, plus GA in drinking water at a concentration of 6%$^{w/v}$ and 12%$^{w/v}$, respectively, for four weeks.

During the treatment periods rats were weighed weekly and placed individually in metabolic cages to collect the urine voided in 24 h. Twenty-four hours after the end of the treatment rats were anesthetized with intraperitoneal injection of ketamine (75 mg/kg) and xylazine (5 mg/kg), and blood (3 mL) collected from the anterior vena cava was placed into heparinized tubes. The blood and urine were centrifuged at 900 *g* at 4°C for 15 min. The plasma obtained, together with the urine specimens, were stored frozen at −80°C pending analysis. The two kidneys were excised, blotted on filter paper and weighed. A small piece of the right kidney was placed in 10% formalin for subsequent histological processing. The rest of the kidneys were kept frozen at −80°C to await biochemical analysis within a week. The left kidney was homogenized in ice-cold Tris buffer (pH 7.4) to give a 10%$^{w/v}$ homogenate. The latter was centrifuged at 1500 *g* at 4°C for 15 min, and the supernatant obtained was used to measure SOD activity.

Biochemical and physiological measurements

Creatinine (in urine and plasma), urea (in plasma) and protein (in urine) contents, and NAG and LDH activities (in urine) were measured spectrophotometrically using commercial kits (Biomerieux, Marcy l'Etoile, France). Urine antioxidant capacity was assessed by the Trolox equivalent antioxidant capacity, and was measured spectrophotometrically according to Miller *et al.*[18] using a commercial kit (Cayman Chemical).

The SOD activity in kidney supernatant homogenates was measured spectrophotometrically by a kinetic method using a commercial kit (RANSOD, by Randox Laboratories), and results were expressed in international units per milliliter. Osmolarity was measured with an Advanced Digimatic osmometer (Advanced Instruments Inc, Norwood, MA, USA) using the cryoscopic method. Creatinine clearance (CCr) was calculated by a standard method.[19] Briefly, CCr was calculated using the following equation: CCr (mL/(min kg) body weight) = [urinary Cr (mg/dL) × urinary volume (mL)/serum Cr (mg/dL)] × [1000/body weight (g)] × [1/1440 (min)].

Renal histology

The kidneys were fixed in 10% neutral formalin, dehydrated in increasing concentrations of ethanol, cleared with xylene and embedded in paraffin. Five micrometer sections were prepared from kidney paraffin blocks and stained with hematoxylin and eosin and Masson's trichrome stains using standard procedures.[20] The microscopic scoring of the kidney sections was carried out in a blind fashion by a pathologist who was unaware of the treatment groups and assigned a score, as described before,[21] which represents the approximate extent of necrotic area in the cortical proximal tubules on a scale of 0–4 (0, no necrosis; 1, a few focal necrotic spots; 2, necrotic area was about one half; 3, necrotic spots was about two-thirds; 4, nearly the entire area was necrotic).

Table 1 Effect of treatment of rats with Gum Arabic (GA, 6%$^{w/v}$ and 12%$^{w/v}$ in drinking water), with or without adenine in the feed (0.75%$^{w/w}$) on body and kidney weights

Group	Initial wt (g)	Final wt (g)	Change (%)	Kidney wt (%)
Control	191.1 ± 3.3	334.2 ± 10.2	74.9 ± 4.4	0.81 ± 0.01
GA 6%	201.3 ± 4.7	286.5 ± 6.2	42.4 ± 1.9	0.68 ± 0.02
GA 12%	183.3 ± 7.7	270.7 ± 6.3	49.0 ± 7.3	Not available
Adenine	250.8 ± 5.2	158.5 ± 7.6	−46.8 ± 10.8	2.13 ± 0.03
Adenine + GA 6%	262.9 ± 4.2	234.1 ± 5.5	−10.9 ± 2.0	1.19 ± 0.03
Adenine + GA 12%	243.3 ± 6.1	206.7 ± 6.0	−14.8 ± 2.9	1.13 ± 0.02

Each value is a mean ± SEM ($n = 6$ rats)
Kidney weights were expressed as % of the final body weight

Statistical analysis

Data are expressed as mean ± SEM and were analyzed by one-way analysis of variance followed by Tukey's multiple comparison tests (Graphpad Prism version 4.03, San Diego, CA, USA); $P \leq 0.05$ was considered statistically significant.

Results

General effects

Table 1 shows the body weight changes in the six groups studied. Adenine feeding (0.75%$^{w/w}$ for 4 weeks) caused a progressive decrease in body weight amounting to about 47% at the end of the treatment period, when compared with the initial body weight. Concomitant treatment with GA at concentrations of 6%$^{w/w}$ and 12%$^{w/w}$ in water ameliorated this effect.

The weights of the kidneys relative to the final body weights in adenine-treated rats (Table 1) indicated that the relative weights of kidneys from rats treated with adenine are significantly higher than those in control rats. Concomitant treatment with GA at the two doses incompletely, but significantly, reversed this effect ($P < 0.05$). The general appearance of the adenine-treated rats was improved by GA treatment at both doses. Kidneys of adenine-treated rats were pale, and a few adenine crystals were seen mainly in the cortex area. The morphological appearance of the kidneys of rats treated with adenine plus GA at the two doses used was improved compared with that of the kidneys of rats treated with adenine alone (Figure 1).

Biochemical findings

Plasma

As shown in Figure 2, adenine feeding (0.75%$^{w/w}$ for 4 weeks) caused significant elevations ($P < 0.001$) in the concentrations of urea and Cr in the plasma, and significant decrease in the CCr ($P < 0.001$). Treatment with GA, at both doses, significantly mitigated the adenine effect.

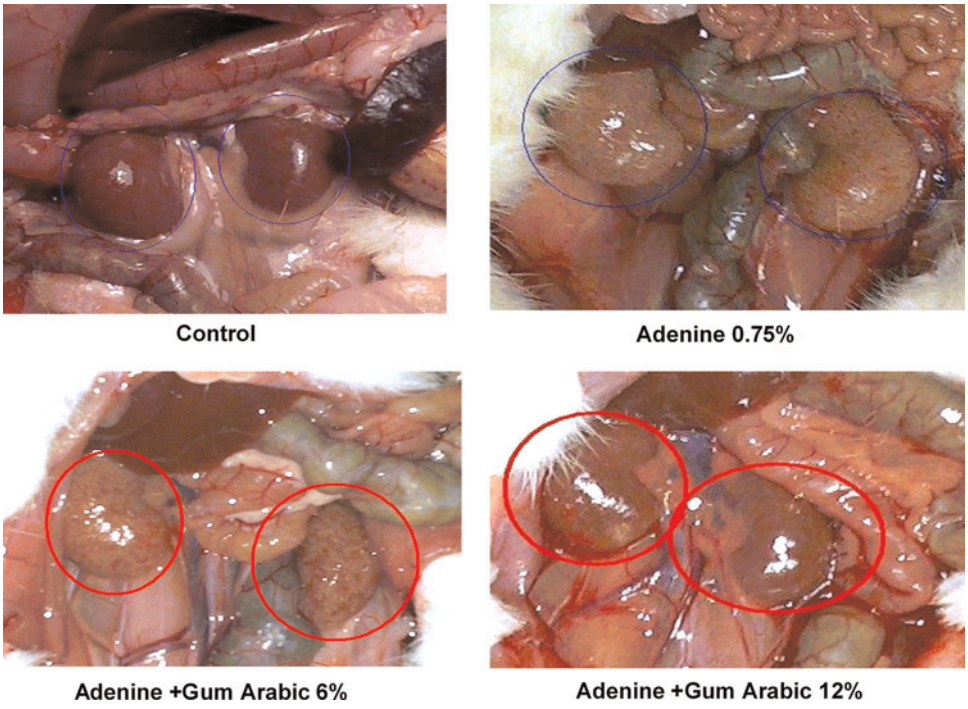

Figure 1 Gross morphology of the kidneys of a rat that has received adenine only (0.75%$^{w/w}$ in the feed), and two doses of Gum Arabic (6%$^{w/w}$ and 12%$^{w/w}$ in drinking water), compared with a control rat (normal saline) (A color version of this figure is available in the online journal)

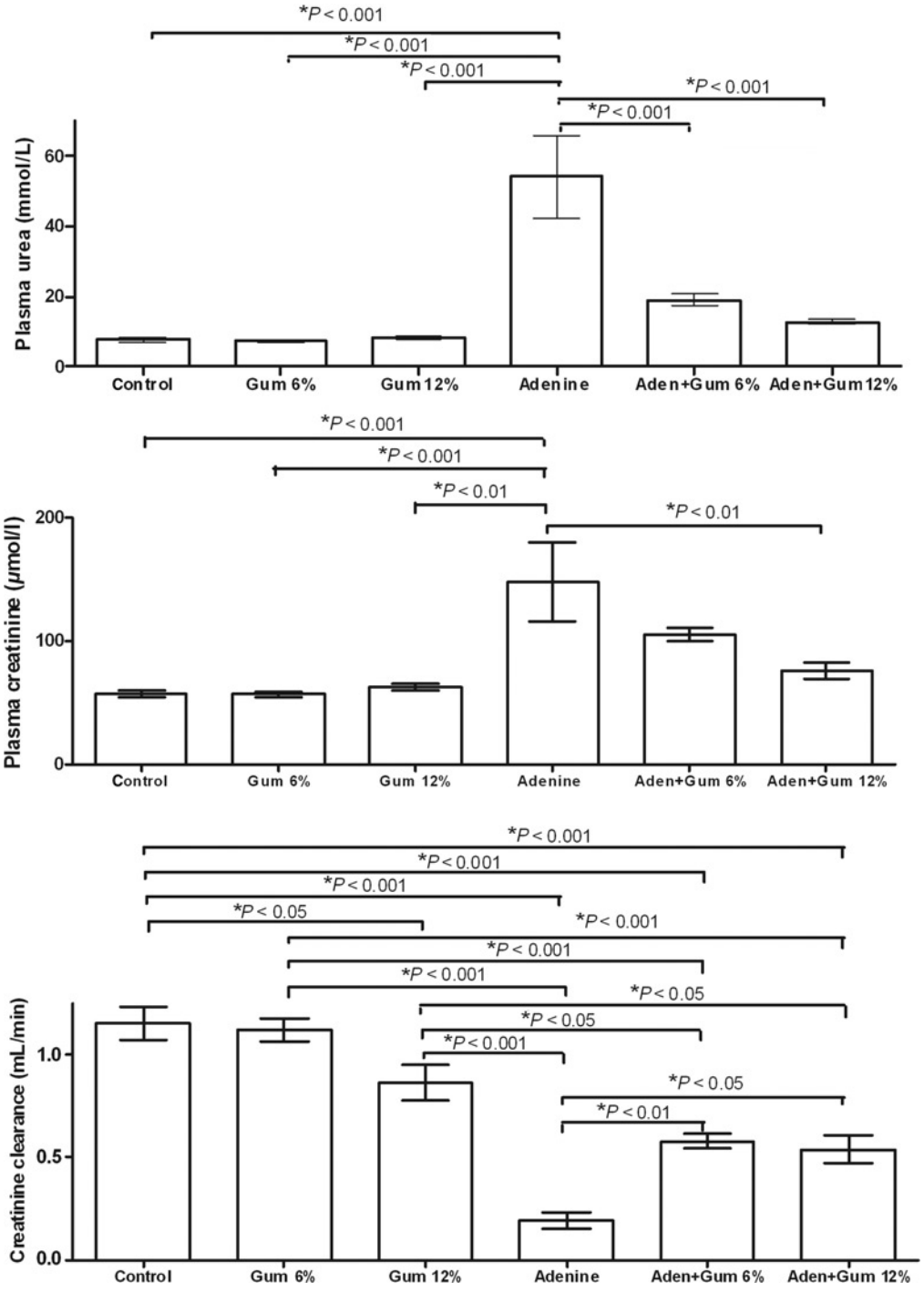

Figure 2 Effect of adenine feeding (0.75%$^{w/w}$) with or without concomitant administration of Gum Arabic (6%$^{w/w}$ and 12%$^{w/w}$ in drinking water) on creatinine and urea plasma concentrations and on creatinine clearance. Each column and vertical bar depict mean ± SEM ($n = 6$ rats)

Urine

Water intake and urine output were progressively and significantly increased by adenine treatment (Figure 3). This effect was mitigated by concomitant treatment with GA (6%$^{w/v}$ and 12%$^{w/v}$). GA, at the two doses used, did not significantly affect the urinary osmolarity or NAG activity. However, adenine feeding significantly increased the activity of the enzyme and decreased the osmolarity, an effect that was incompletely reversed by simultaneous GA treatment. At a concentration of 12%$^{w/v}$, GA was effective in significantly ameliorating the adenine-induced increase in NAG activity ($P < 0.05$). As shown in Figure 4, adenine feeding significantly suppressed total antioxidant capacity. This effect was significantly, but not completely, reversed by concomitant treatment with GA at the two doses used. The activity of LDH in urine was not measurable in the urine of rats treated with adenine. In rats treated with GA at the two doses, the LDH in urine was increased. Simultaneous treatment with GA and adenine incompletely reversed this action (Figure 4). A trend in the decrease of

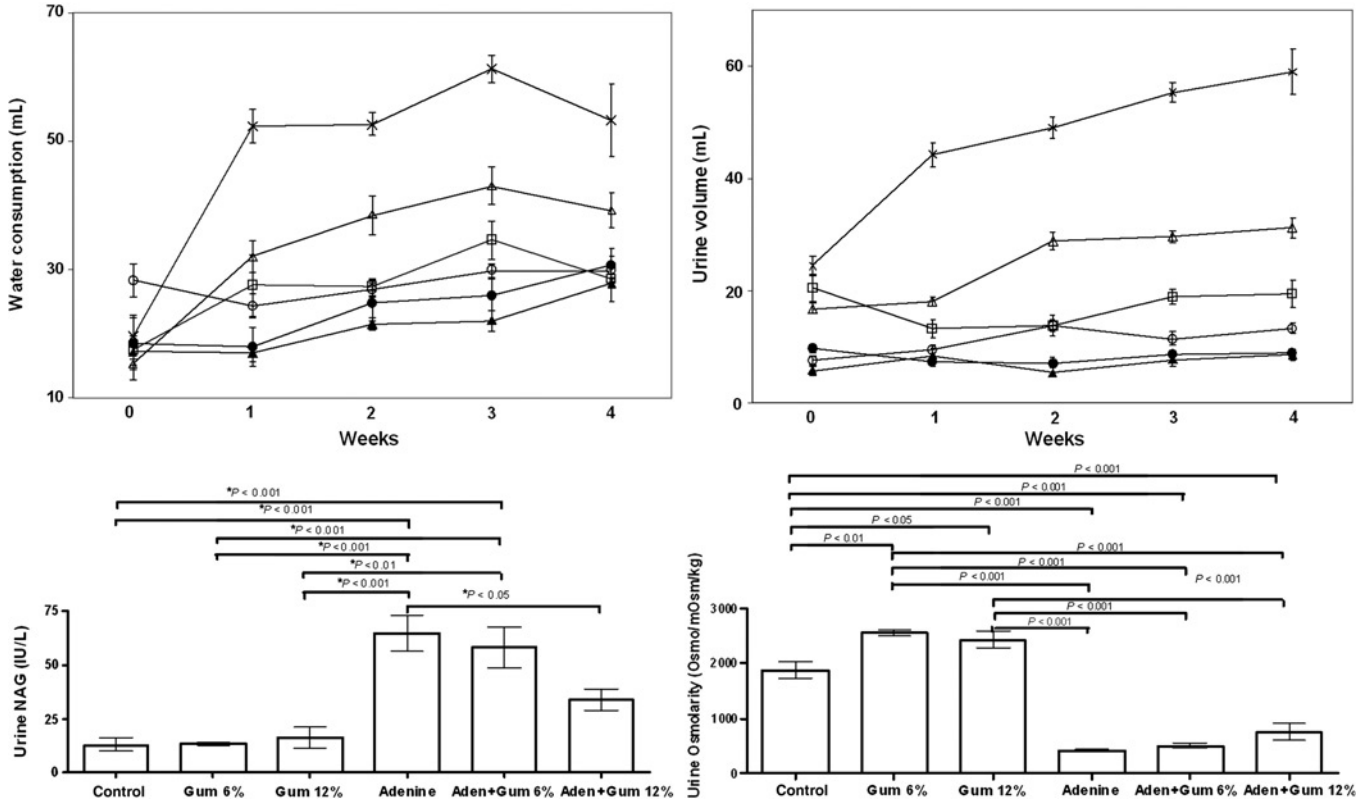

Figure 3 Effect of treatment of rats with saline (○), Gum Arabic (GA) 6%$^{w/w}$ in drinking water (•), GA 12%$^{w/w}$ in drinking water (▲), adenine 0.75%$^{w/v}$ (X), adenine + GA 6% (Δ), adenine + GA 12% (□), on water consumption, 24-h urine volume, urine *N*-acetylglucosamine (NAG) activity and urine osmolarity. Each point or column is a mean ± SEM from six rats

IL-6 concentrations in the urine in adenine-treated group, which was mitigated by GA, has been observed. However, overall, IL-6 levels measured were variable, and no statistical difference was observed between the different groups.

Kidney

The effect of adenine and GA treatments on the activity of SOD in rat renal tissue is shown in Figure 5. GA at the two doses produced a slight increase in the SOD activity (significant at a concentration of 12%$^{w/v}$), while adenine produced a marked reduction ($P < 0.01$). This reduction was incompletely but significantly reversed by GA at both doses ($P < 0.05$).

Histopathological findings

Rats in the control group showed normal kidney architecture and histology and complete absence of interstitial fibrosis (Figures 6A and 7A). Similarly, the rats treated with 6% or 12% GA had normal kidney architecture and histology, with absence of interstitial fibrosis (Figures 6B, C, 7B and C)

The adenine-treated groups showed diffuse tubular injury with neutrophil polymorph infiltration, tubular necrosis, tubular atrophy and diffuse interstitial fibrosis (Figure 6D). In this group, the Masson's trichrome stain identified the interstitial fibrous tissue by staining it green (Figure 7D).

Kidney histological sections of the rats treated with adenine plus 6% GA (Figure 6E) showed marked improvement in comparison with the adenine-treated group, reverting the histological appearance seen in this latter group to normal in about 50% of the examined tissue fields (Figure 6E). There were areas of tubular injury with neutrophil polymorph infiltration, tubular necrosis, tubular atrophy and interstitial fibrosis, but these were of less intensity than in the adenine-treated group, although they were seen in about 50% of the examined tissue fields (Figure 6E). The Masson's trichrome stain demonstrated green staining of interstitial fibrous tissue (Figure 7E). This was less in the 6% GA group than in the adenine-treated group and affected about 50% of the examined tissue field.

Kidney histological sections of the rats treated with adenine plus 12% GA showed more marked improvement, showing normal kidney architecture and histology, with the absence of interstitial fibrosis (Figures 6F and 7F). Sections from these rats showed striking histological improvement reverting the appearance seen in the adenine-treated group to normal in about 80% of the examined tissue fields (Figure 6F). There were few foci showing tubular injury with neutrophil polymorph infiltration, tubular necrosis, tubular atrophy and interstitial fibrosis, which were less intense than those seen in the adenine-treated group and the adenine plus 6% GA-treated rats, and were seen in about 20% of the examined tissue fields (Figure 6F).

The Masson's trichrome stain demonstrated the green staining of interstitial fibrous tissue (Figure 7F), which was less than that deposited in the adenine-treated group, and in the rats treated with adenine together with GA (6%), and affected about 20% of the examined tissue field (Figure 7F).

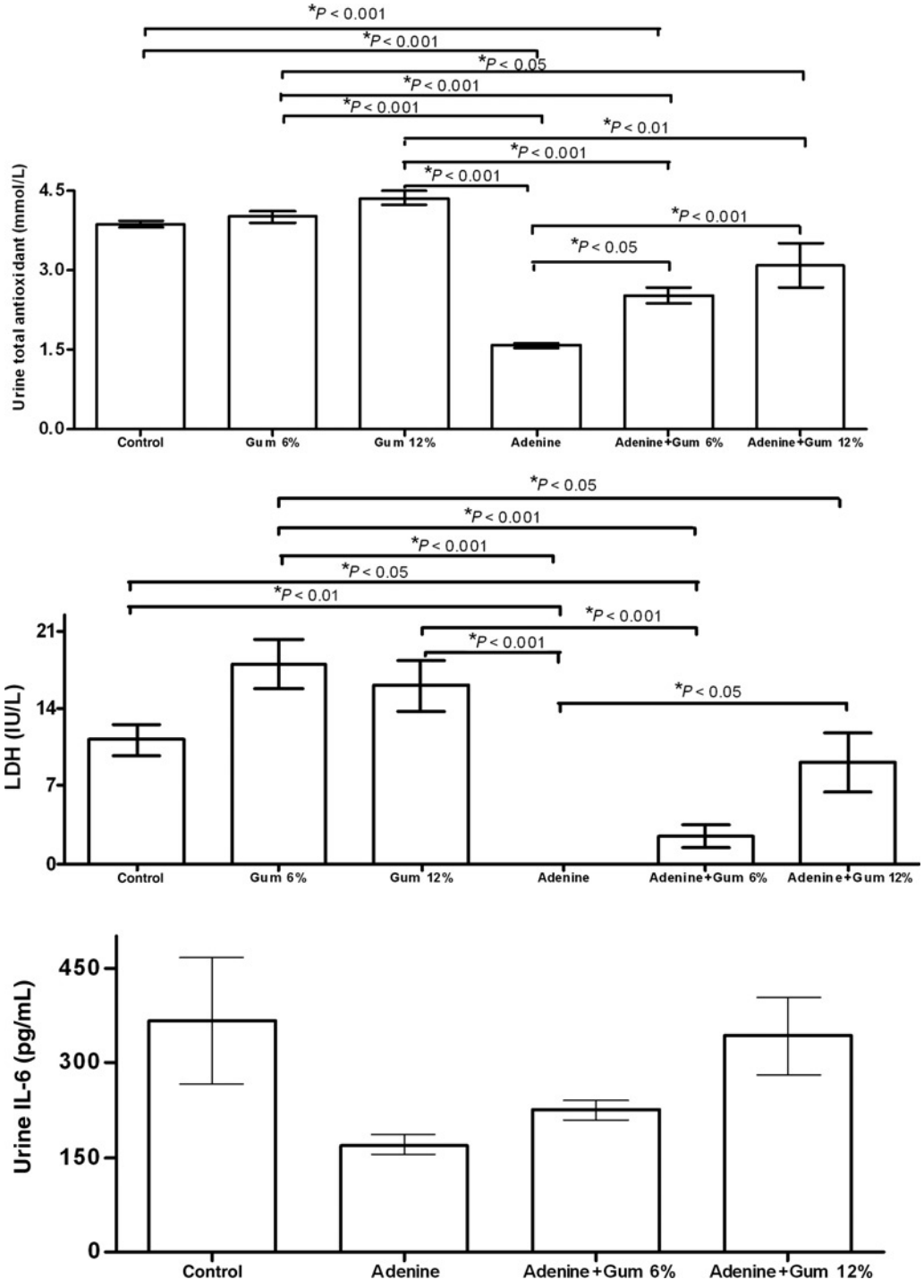

Figure 4 Effect of adenine feeding (0.75%$^{w/w}$), with or without concomitant administration of Gum Arabic (6%$^{w/v}$ and 12%$^{w/v}$ in drinking water), on urinary antioxidant, lactate dehydrogenase (LDH) activity and interleukin (IL)-6 concentration. Each column and vertical bar represent mean $\pm$ SEM ($n = 6$ rats)

Table 2 shows a semi-quantative analysis of the histology of the kidney of rats treated with adenine, with or without GA.

Discussion

It has been shown that, in rats, long-term feeding of adenine suppresses the excretion of nitrogenous compounds by means of renal tubular occlusion, and produces metabolic abnormalities resembling CRF in humans.[14] In mammalian metabolism, when adenine is present in excess, it becomes a significant substrate for xanthine dehydrogenase. This enzyme can oxidize adenine to 2,8-dihydroxyadenine (DHA). Because adenine and DHA have low solubility, they precipitate in renal tubules.

Adachi *et al.*[22] reported that adenine-rich diets increase blood urea nitrogen and Cr by decreasing the excretion of these substances, because excretion of nitrogenous

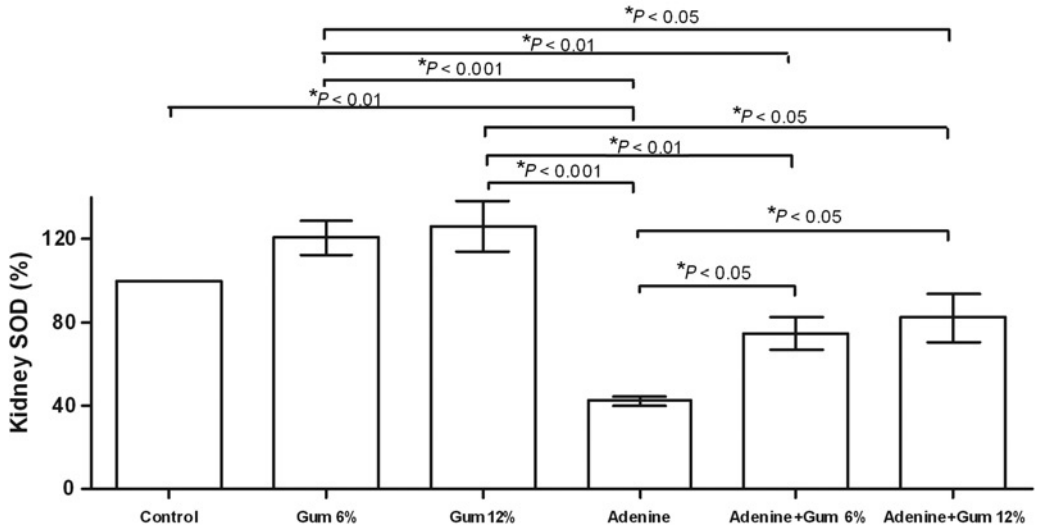

Figure 5 Activity of superoxide dismutase (SOD) in kidney homogenate from rats treated with Gum Arabic (6%$^{w/w}$ and 12%$^{w/w}$ in drinking water) with or without adenine in the feed (0.75%$^{w/w}$), expressed as % of the control values. Each column represents mean from six rats. Each column and vertical bar represent mean ± SEM ($n = 6$ rats)

compounds is suppressed by renal occlusion due to DHA. Deng *et al.*[23] also showed that the disease progression of adenine-induced CRF is time-dependent – the longer the feeding time, the more severe is the disease. In this work we opted to feed adenine (0.75%$^{w/w}$) to the rats for 28 days, which was found by some researchers to be an optimum duration of treatment that damages the kidneys but does not produce mortality.[22,23] It has been reported that feeding adenine (0.04%$^{w/w}$) for 38 days has resulted in some mortalities.[24] Others have found that the same dose that we employed (0.75%$^{w/w}$) produced an 'ideal model resembling CRF', although one-half of their treated rats died.[25] No mortality was encountered in our experiment and a biochemically and histopathologically proven CRF was observed.

Another animal model with many similar characteristics of human CRF is the remnant kidney.[15] In a previous work, we employed the latter model to test the reputed usefulness of GA in CRF.[8] GA given in the drinking water at a dose of 3 or 6 g/100 mL/day for five consecutive weeks did not offer a clear or significant nephroprotective effect. In contrast, in the current study we employed the adenine-induced CRF model and found that GA at the two doses used (6% and 12% in the drinking water) significantly mitigated the extent of the biochemical, physiological and histopathological effects of the adenine. The reason for the discrepancy between the results of the two animal models of CRF is not known. However, it can be related to the fact that CRF in the remnant kidney model appears just after the surgical operation and is slowly progressing. The discrepancy between the two models may also be a reflection of the lower dosage of GA used in the earlier experiment, or to the severity of the surgical model, which induced in our previous experiment a more drastic CRF that could not be mitigated by GA treatment at the relatively low doses used. Another possible explanation why this model may have failed (where adenine model has worked) is that the reno-protective action of GA may involve a component in the kidney. This will be mostly absent in the surgical model, and therefore, perhaps the adenine model (where all the renal tissue is present) reacts differently to GA than in the surgical model. It has been suggested that nephrin may be such a component in the kidneys that GA may work on to cause nephroprotection (Nasir, personal communication). Nephrin is a structural protein of the slit diaphragm, is expressed on the surface of glomerular podocytes and is critical in maintaining permselectivity and preventing proteinuria.[26] To what extent these animal models of CRF are applicable to CRF in humans, considering that the situations of patients suffering from CRF cannot be easily and completely reproduced, is not certain.[26] However, the fact that more than one preliminary clinical report from Sudan[10] and Iraq[13] have indicated a beneficial effect of giving GA to CRF patients lends strong support to the relevance and validity of the model used here.

In the present work, several physiological, biochemical and histopathological parameters have been used to find out if GA consumption would mitigate CRF, as claimed in some traditional medicinal practices,[8] and reported in a few papers from Sudan and Iraq.[10–12] Our results suggest that GA at the doses tested and for the duration used was, in fact, effective in significantly (but incompletely) reversing most of the indices of CRF measured.

The mechanism(s) by which GA improves renal function in CRF are not certain. It has been postulated that GA enhances the amount of energy available to the colonies of bacteria that ferment dietary fibers and absorb nitrogen as they grow.[7–9] These bacterial colonies are also capable of degrading urea to ammonia, excreting it in feces and taking some of the body nitrogen waste with them.[9,14] It has also been shown that consuming fermentable fibers can lead to enlargement of the cecum by increasing its contents and its wall thickness and blood flow. These changes resulted in concomitant enhancement of urea nitrogen uptake into the cecum and a decrease in plasma urea

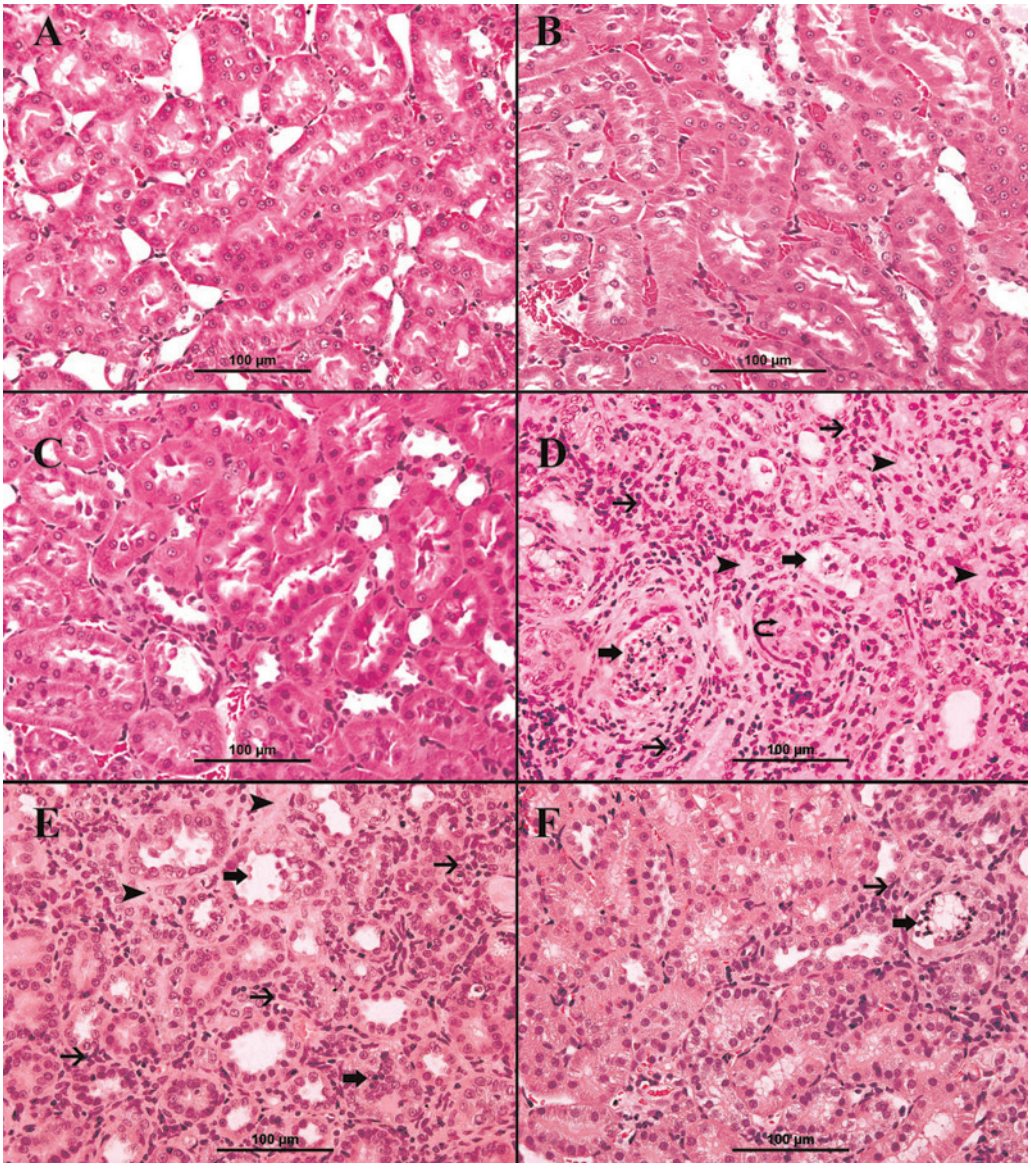

Figure 6 Representative photograph of sections after haematoxylin and eosin staining (H&E, 100×) of renal tissue under light microscope of rats treated with saline, Gum Arabic (GA), adenine and adenine + GA. (A) Control group, showing normal kidney architecture and histology, H&E. (B) GA 6% treated group, showing normal kidney architecture and histology, H&E. (C) GA 12% treated group, showing normal kidney architecture and histology, H&E. (D) Adenine-treated groups, showing diffuse tubular injury (thick arrow) with neutophil polymorph infiltration, tubular necrosis, tubular atrophy, interstitial inflammatory cells infiltration (thin arrow), interstitial fibrosis (arrowhead) and a sclerosed glomerulus (curved arrow), H&E. (D) Adenine + GA 6% treated group, showing histological improvement with reverting the histological appearance seen in (D) to normal in about 50% of the examined tissue fields. Areas of tubular injury (thick arrow), tubular atrophy, interstitial inflammatory cell infiltration (thin arrow) and interstitial fibrosis (arrowhead) which are of less intensity than (D) and are seen in about 50% of the examined tissue fields, H&E. (F) Adenine + GA 12% treated group, showing striking histological improvement with reverting the histological appearance seen in (D) to normal in about 80% of the examined tissue fields; focal area of tubular injury with neutophil polymorph infiltration (thick arrow) and interstitial inflammatory cell infiltration (thin arrow) which are less in intensity than (D, E) and are seen in about 20% of the examined tissue fields, H&E (A color version of this figure is available in the online journal)

concentration by about 30%.[2] It has also been proposed in the 'protein metabolite hypothesis' that circulating endogenous protein metabolites such as indoxyl sulfate (IS) play an important role in the progression of CRF by stimulating glomerular sclerosis and interstitial fibrosis.[27,28] GA at the two doses used ameliorated the significant increase in the concentrations of IS in plasma that was induced by adenine feeding (data not given in this work). It is known that IS, a metabolite that is derived from dietary protein, is a uremic toxin that accelerates the progression of CRF,[27,28] and has a pro-oxidant effect.[29] It is also known that pathogenesis of experimental CRF is known to be associated with generation of damaging free radicals[29] and also with an inflammatory response.[30] It was of interest to find out in this work whether GA would influence the oxidant status and/or inflammatory response of rats treated with adenine. We measured the activity of SOD in renal tissues as an indication of the oxidant status of the kidney tissue, and found that the activity of the enzyme was significantly decreased in adenine-treated rats, possibly suggesting that this CRF model involves generation of free radicals in the renal tissue. GA treatment ameliorated this action and

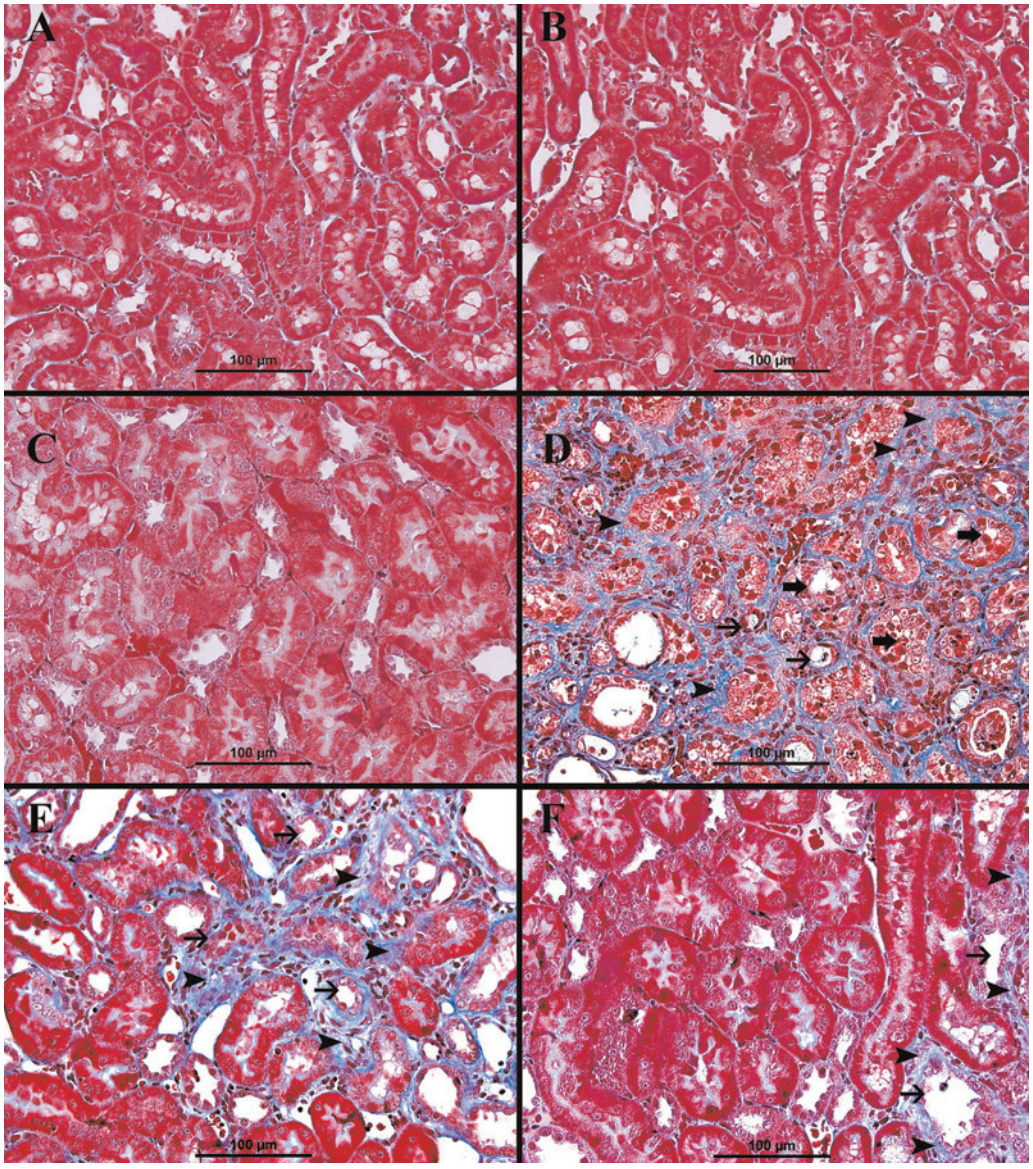

Figure 7 Representative photograph of sections after Masson's trichrome staining (100×) of renal tissue under light microscope of rats treated with saline, Gum Arabic (GA), adenine and adenine + GA. (A) Control group, showing normal kidney architecture and histology, Masson's trichrome stain. (B) GA 6% treated group, showing normal kidney architecture and histology, Masson's trichrome stain. (C) GA 12% treated group, showing normal kidney architecture and histology, Masson's trichrome stain. (D) Adenine treated groups, showing diffuse tubular injury (thick arrows), tubular atrophy (thin arrow) and interstitial fibrosis (arrowheads), Masson's trichrome stain. (E) Adenine + GA 6% treated group, showing histological improvement with reverting the histological appearance seen in (D) to normal in about 50% of the examined tissue fields. Areas of tubular atrophy (thin arrows) and interstitial fibrosis (arrowheads) which are of less intensity than (D) and are seen in about 50% of the examined tissue fields, Masson's trichrome stain. (F) Adenine + GA 12% treated group, showing significant histological improvements with reverting the histological appearance seen in (D) to normal in about 80% of the examined tissue fields; focal area of interstitial fibrosis (arrow heads) and tubular atrophy (thin arrows) which are less in intensity than (D, E) and are seen in about 20% of the examined tissue fields, Masson's trichrome stain. (A color version of this figure is available in the online journal)

Table 2 Semi-quantative analysis of histology of the kidney of rats treated with adenine with or without Gum Arabic (GA)

Group	Treatments	Score
1	Saline for four weeks (control)	+0
2	GA (6% in drinking water for 4 weeks)	+0
3	GA (12% in drinking water for 4 weeks)	+0
4	Adenine (0.75% in feed for 4 weeks)	+3
5	Adenine (as above) + GA (as in group 2)	+2
6	Adenine (as above) + GA (as in group 3)	+1

significantly, but not completely, reversed the adenine-induced decrease in SOD activity, possibly indicating that GA may have acted as an antioxidant by itself, or more likely, by another (yet unidentified) mechanism. In a preliminary *in vitro* experiment, we could not find a significant effect for GA on some free radical scavengers.[31] We also measured IL-6 cytokine (an inflammatory cytokine), which is an indicator of inflammatory response.[32] Although statistically insignificant, a trend to a decrease in the concentration of IL-6 was found in urine, suggesting that its level in plasma has increased. This action was mitigated by GA

treatment, a possible indication that the adenine-induced CRF involves an inflammatory response. Both of these two possible mechanisms need to be investigated in more detail by measuring more cytokines and reactive oxygen species.

In conclusion, this study provided experimental evidence that GA attenuated adenine-induced renal dysfunction, suggesting a promising potential of GA in protecting against renal failure progression. The mechanism(s) of this nephroprotection is uncertain but may involve anti-oxidant and/or anti-inflammatory actions. Further work on the effects of GA on some consequences of adenine-induced CRF (e.g. hypertension and anemia) is in progress.

Author contributions: All authors have read and approved the final manuscript. BHA planned, supervised, performed the animal experimentations and wrote the article. SA performed and wrote the histopathology part of the article. IA, RK, NA and TA performed the biochemical analysis of the study. MA contributed to the biochemical analysis and writing up of the article. AN supervised, performed the experiments and wrote the article.

ACKNOWLEDGEMENTS

The authors thank the College of Medicine and Health Sciences at SQU for their partial financial support, and the staff of the Animal House for looking after the rats. We thank Professor R Bayoumi for his interest. The work performed in the UAE University, FMHS, Department of Physiology was supported by the funds of the UAE University individual grant (01-05-8-11/08) and Faculty of Medicine and Health Sciences grant (NP/09/04). The authors wish to thank Dhanasekaran for his technical help.

REFERENCES

1 Hossain MP, Goyder EC, Rigby JE, El Nahas M. CKD and poverty: a growing global challenge. *Am J Kidney Dis* 2009;**53**:166–74
2 Younes H, Egret N, Hadj-Abdelkader M, Remesy C, Demigne C, Gueret C, Deteix P, Alphonse JC. Fermentable carbohydrate supplementation alters nitrogen excretion in chronic renal failure. *J Ren Nutr* 2006;**16**:67–74
3 Lacson E Jr, Ikizler TA, Lazarus JM, Teng M, Hakim RM. Potential impact of nutritional intervention on end-stage renal disease hospitalization, death, and treatment costs. *J Ren Nutr* 2007;**17**:363–71
4 Fouque D, Guebre-Egziabher F. An update on nutrition in chronic kidney disease. *Int Urol Nephrol* 2007;**39**:239–46
5 Chaturvedi S, Jones C. Protein restriction for children with chronic renal failure. *Cochrane Database Syst Rev* 2007:CD006863
6 Winchester JF, Salsberg JA. Sorbents in the treatment of renal failure. *Minerva Urol Nefrol* 2004;**56**:215–21
7 Ali BH, Ziada A, Blunden G. Biological effects of gum arabic: a review of some recent research. *Food Chem Toxicol* 2009;**47**:1–8
8 Ali BH, Alqarawi AA, Ahmed IH. Does treatment with gum Arabic affect experimental chronic renal failure in rats? *Fund Clin Pharmacol* 2004;**18**:327–9
9 Bliss DZ, Stein TP, Schleifer CR, Settle RG. Supplementation with gum arabic fiber increases fecal nitrogen excretion and lowers serum urea nitrogen concentration in chronic renal failure patients consuming a low-protein diet. *Am J Clin Nutr* 1996;**63**:392–8
10 Ali AA, Ali KE, Fadlalla AE, Khalid KE. The effects of gum arabic oral treatment on the metabolic profile of chronic renal failure patients under regular haemodialysis in Central Sudan. *Nat Prod Res* 2008;**22**:12–21
11 Al Mosawi AJ. The use of acacia gum in end stage renal failure. *J Trop Pediatr* 2007;**53**:362–5
12 Al Mosawi AJ. Acacia gum supplementation of a low-protein diet in children with end-stage renal disease. *Pediatr Nephrol* 2004;**19**:1156–9
13 Al Mosawi AJ. The challenge of chronic renal failure in the developing world: possible use of acacia gum. *Pediatr Nephrol* 2002;**17**:390–1
14 Younes H, Garleb K, Behr S, Remesy C, Demigne C. Fermentable fibers or oligosaccharides reduce urinary nitrogen excretion by increasing urea disposal in the rat cecum. *J Nutr* 1995;**125**:1010–6
15 Yokozawa T, Zheng PD, Oura H, Koizumi F. Animal model of adenine-induced chronic renal failure in rats. *Nephron* 1986;**44**:230–4
16 Ormrod D, Miller T. Experimental uremia. Description of a model producing varying degrees of stable uremia. *Nephron* 1980;**26**: 249–54
17 Ali BH, Al-Salam S, Nemmar A, Al-Zaabi M, Al-Husseini I. The effect of Gum Arabic on an animal model of chronic renal failure. *Basic Clin Pharmacol Toxicol* 2009;**105**:128
18 Miller NJ, Rice-Evans C, Davies MJ, Gopinathan V, Milner A. A novel method for measuring antioxidant capacity and its application to monitoring the antioxidant status in premature neonates. *Clin Sci (Lond)* 1993;**84**:407–12
19 Duarte CG, Preuss HG. Assessment of renal function – glomerular and tubular. *Clin Lab Med* 1993;**13**:33–52
20 Prophet E, Mills B, Arrington J, Sobin J. *Laboratory Methods in Histotechnology*. Armed Forces Institute of Pathology, American Registry of Pathology, 1994
21 Ali BH, Al Moundhri M, Eldin MT, Nemmar A, Al Siyabi S, Annamalai K. Amelioration of cisplatin-induced nephrotoxicity in rats by tetramethylpyrazine, a major constituent of the Chinese herb *Ligusticum wallichi*. *Exp Biol Med (Maywood)* 2008;**233**:891–6
22 Adachi Y, Sasagawa I, Tateno T, Tomaru M, Kubota Y, Nakada T. Influence of adenine-induced chronic renal failure on testicular function in the rat. *Andrologia* 1998;**30**:115–8
23 Deng H, Jin W, Liao X, He Y. Methodology for designing pathological models of acute renal failure. *Zhongguo Zhong Yao Za Zhi* 1998;**23**: 48–52, 64
24 Ngai HH, Sit WH, Wan JM. The nephroprotective effects of the herbal medicine preparation, WH30+, on the chemical-induced acute and chronic renal failure in rats. *Am J Chin Med* 2005;**33**:491–500
25 Aoyama I, Miyazaki T, Niwa T. Preventive effects of an oral sorbent on nephropathy in rats. *Miner Electrolyte Metab* 1999;**25**:365–72
26 Hauser PV, Collino F, Bussolati B, Camussi G. Nephrin and endothelial injury. *Curr Opin Nephrol Hypertens* 2009;**18**:3–8
27 Bankir L, Bouby N, Trinh-Trang-Tan MM. Vasopressin-dependent kidney hypertrophy: role of urinary concentration in protein-induced hypertrophy and in the progression of chronic renal failure. *Am J Kidney Dis* 1991;**17**:661–5
28 Bouby N, Trinh-Trang-Tan MM, Laouari D, Kleinknecht C, Grunfeld JP, Kriz W, Bankir L. Role of the urinary concentrating process in the renal effects of high protein intake. *Kidney Int* 1988;**34**:4–12
29 Tumur Z, Niwa T. Indoxyl sulfate inhibits nitric oxide production and cell viability by inducing oxidative stress in vascular endothelial cells. *Am J Nephrol* 2009;**29**:551–7
30 Dou L, Jourde-Chiche N, Faure V, Cerini C, Berland Y, Dignat-George F, Brunet P. The uremic solute indoxyl sulfate induces oxidative stress in endothelial cells. *J Thromb Haemost* 2007;**5**:1302–8
31 Nistala R, Whaley-Connell A, Sowers JR. Redox control of renal function and hypertension. *Antioxid Redox Signal* 2008;**10**:2047–89
32 Zoja C, Garcia PB, Remuzzi G. The role of chemokines in progressive renal disease. *Front Biosci* 2009;**14**:1815–22

(Received July 19, 2009, Accepted October 17, 2009)