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Potential Effects of Papaya Seeds and Leaves on Kidney Disorder in Gentamicin-Induced Rats

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Abstract:

This study investigated the preventive effects of a food supplement with Papaya (Carica papaya) leaves and seeds either alone or in combinations at 2.5 and 5% against Gentamicin-induced Chronic kidney disease (CKD) in rats. Forty-eight rats weighing 150±5g were fed a basal diet for a week adaptation period. Rats were divided randomly into eight groups; each group included 6 rats. The first healthy group fed basal diet only as a Negative (-ve) Control, while the second group fed basal diet as a Positive (+ve) control. Other 6 pretreated groups fed basal diets containing Papaya (Carica papaya) leaves and seeds and their combinations at 2.5 and 5%, respectively, for about 4 weeks. Following the 28-day study period, gentamicin (10 ml/kg body weight) was delivered into all pretreatment groups and the positive control group to cause chronic kidney disease. The rats were weighed weekly during the trial, and their feed consumption and weight increase were noted. Following the experiment's conclusion, rats were sacrificed, blood samples were taken, and their serum was separated, centrifuged, and then frozen at -20°C until analysis. Biochemical analysis such as serum creatinine, urea nitrogen, uric acid, Serum sodium (Na), Serum potassium (K), Albumin, Blood glucose, Serum triglycerides, Total cholesterol, HDL, LDL, VLDL levels, Serum total protein (STP), L-Aspartate aminotransferase (AST), L-Alanine aminotransferase (ALT) and Alkaline phosphatase (ALP) was measured. Additionally, superoxide dismutase (SOD), glutathione reeducates (GSH), and Malondialdehyde (MDA) were measured. The groups treated with higher amounts showed the best results for Papaya leaves and seeds.

Keywords: CKD, Papaya, Kidney functions, Rats

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1. Introduction

Chronic kidney disease (CKD) is a progressively degenerative condition characterized by the gradual deterioration of renal functions, representing a significant and pressing global health issue (1). CKD is defined as abnormalities of kidney structure or functions, present for >3 months, with implications for health (2). CKD is a devastating condition that is reaching epidemic levels owing to the increasing prevalence of diabetes mellitus, hypertension and obesity, as well as ageing of the population. Regardless of the underlying a etiology, CKD is slowly progressive and leads to irreversible nephron loss, end-stage renal disease and/or premature death (3). CKD is a major public health concern affecting at least 10% of the population worldwide (>800 million people), and this prevalence is expected to increase as a result of growing rates of diabetes mellitus and hypertension (4). CKD is the fastest-growing chronic disease and is one of the most important causes of mortality (5).

Physical frailty is common in patients with CKD, which can lead to poor prognosis, as reflected by an increased risk of adverse events such as motor dysfunctions, limited mobility in daily life, and falls, leading to higher disability rates (6). Between 2017 and 2021, seven meta-analyses assessing the prevalence of physical frailty in patients with CKD or the impact of frailty on survival were published. (7-10). The inflammatory state is usually elevated in patients with CKD, and the systemic inflammatory response is a crucial factor in the progression of CKD (11). Alterations in serum electrolytes, protein, or biomarkers of kidney function show up as incidental changes in screening blood tests, perhaps presenting with minor or none of the classic clinical signs (12).

papaya (Carica papaya, Linn) is considered as a nutraceutical fruit, because of its exceptional nutritional and multifaceted medicinal properties which include antibacterial, anti-inflammatory, anti-aging, anti-proliferative, diuretic, anti-hypertensive, hypolipidemic, anti-helminthic, wound healing, anti-fungal and anti-tumor Phytochemically, the whole plant contains enzymes, lycopene, carotenoids, alkaloids, monoterpenoids, flavonoids, minerals and vitamins (13).This fruit stands out as the ultimate source of beta-carotene, due to the harmful effects it has in preventing heart disease and diabetes. Papaya actively contributes to bolstering the immune system, enhancing resistance against coughs and colds, courtesy of its rich vitamin A and C content. The diverse therapeutic properties of papaya make it a valuable addition to promoting overall health and well-being (14).papaya also contains antioxidants in the body such as vegetables A and E, flavonoids and pantothenic acids important in promoting heart and antibiotics and protects the functions of diseases such as colon cancer (15).

The leaves of Papaya (Carica papaya) have proven to be a rich source of vitamin C (ascorbic acid) that functions as a co-factor for the proline and lysine hydroxylases that stabilize the tertiary structure of the collagen molecule, an essential reaction in wound healing (16) Papaya leaf has proven to be a good nutritive agent because of varying concentrations of nutritional components, which include ascorbic acid (38.6%), protein (5.6%), phosphoric acid (0.225%), carbohydrates (8.3%), iron (0.0064%) and minerals like magnesium (0.035% per 100 g) of leaf portion (17). Carica papaya leaves have alkaloids, flavonoids, saponins, tannins, cardiac glycosides, anthraquinones and cardenolides (15).

Papaya (Carica papaya) seeds are part of the papaya plant which is a source of organic waste (18). Papaya seeds contain proteins, fiber, fatty acids, papaya oil, carpaine, vitamins, minerals, and an enzyme Papain (19). Papaya seeds contain minerals like magnesium, potassium, calcium, copper, and zinc, as well as phenolic compounds such as benzyl isothiocyanate, glucosinolates, tocopherols (alpha and delta), beta-cryptoxanthin, beta-carotene, and carotenoids (15). Papaya seeds, containing enzymes like papain and antioxidants, exhibit anti-parasitic properties and may aid digestion. They also possess anti-inflammatory potential, making them a promising candidate for managing inflammatory conditions (20).

2. Materials and Methods

2.1.Materials

2.1.1.papaya leaves and Seeds

papaya (leaves and seeds) were purchased from the Agricultural Research Center, Al-Dokki, Giza governorate, Egypt.

2.1.2.Gentamicin

intra-peritoneal injection of gentamicin (aminoglycosides antibiotics) obtained by Memphis Co. for Pharm. Chem. Ind .Cairo. A.R.E. at 10mg|Kg|day for 6 days in which the nephrotoxicity, one of the adverse reaction of gentamicin takes place.

2.1.3.Chemical kits

Chemicals, kits acquired from El-Gomhorya Company in Cairo, Egypt. A basal diet was received from Technogene Chemical Co., Dokki, Egypt, and included casein as the primary source of protein, cellulose, a mixture of vitamins, salt, methionine, and corn starch.

2.1.4.Experimintal animals

Forty-eight healthy adult male albino rats of Sprague Dawley strain, 10 weeks age, weighing between 140-150 grams were purchased from the Giza Memorial Institute for Ophthalmic Research, Animal House, Ministry of Health, Giza, Egypt.Impaired kidney was induced in normal healthy male albino rats by gentamicin.

2.2.Methods

2.2.1.preparation of papaya leaves

Papaya leaves were obtained from local market then removed the impurities from it and dried it in oven at low-temperature 50c for 60 minutes then put papaya leaves in the blender to get the powder (21).

2.2.2.preparation of papaya seeds:

Fruits were washed thoroughly under running tap water, then took the seeds and dried it in oven at low-temperature 50c for 60 minutes and ground to a fine powder using an air mill(21)

2.2.3.Experimental design

The Science Research Ethics Committee of the Faculty of Home Economics approved the research protocol (numbered #10-SREC-12-2021)

Forty-eight rats weighting 150±5 g were kept in cages and maintained in generally good health using 12 h light /dark cycle and permitted free access to water and a typical diet. For the purpose of adaptation, all rats were fed a basic diet for seven days. Eight groups of rats were created, with six rats in each group, using the following scheme:

Group (1): Fed basal diet (22) and used as a negative control group.

Group (2): Fed basal diet and used as a positive control group.

Groups (3 and 4): Fed basal diets containing 2.5% and 5% of Papaya leaves, respectively.

Groups (5 and 6): Fed basal diets containing 2.5% and 5% of Papaya seeds, respectively.

Groups (7 and 8): Fed basal diet containing combination with Papaya leaves and seeds at 2.5% and 5% for 4 weeks. During the experimental period, rats were weighted weekly and feed intake was recorded daily.

All rats received an injection of Gentamicin (10 ml/kg body weight) on day 26 of the trial. (23) for CKD induction, rats were thirsty eighteen hours before myoglobin-uric renal damage was induced, and they were scarified 48 hours after receiving a gentamicin injection. (24). BWG, or body weight increase, was calculated using (25), rats were fasted overnight before scarifying. After centrifuging the blood samples for 15 minutes at 3000 rpm in a satirized dry centrifuge tube, the serum was carefully separated and stored in a plastic tube at -20°C until analysis.

2.2.4.Biological evaluation

Every week, the weight of each rat was measured, and each day's feed intake was computed. Following the conclusion of the study, body weight gain (BWG), feed efficiency ratio (FER), and relative organ weight (ROW) were used to evaluate the various diets biologically according to the subsequent equations of (26).

$$FER = \frac{\text{Feed Intake (g)}}{\text{Gain in body weight (g)}}$$

$$BWG = \text{Final weight} - \text{Initial weight}$$

$$\text{Relative organ weight (ROW)} = \frac{\text{Organ weight (g)}}{\text{Final body weight(g)}} \times 100$$

2.2.5.Blood sampling

Using kits and an enzymatic approach, serum triglycerides (T.G.) were measured in accordance with the (27, 28). The modified kinetic developed method (29) was used. VLDL-c was determined in milligrams per deciliter (mg/dl) according to (30). Serum low density lipoprotein cholesterol (LDL-c) The measurement was made in milligrams per deciliter. (mg/dl) according to (31).

The identification of serum total protein (STP) was completed in accordance with the method of (32). Using a kit from Spin React, Spain, the modified kinetic approach outlined by (33) was used to measure the blood's glucose levels. The method outlined by was used to determine the concentration of hemoglobin (HlB), red blood cells (RBCs), white blood cells (WBCs), and platelets (PLT) (34).

Serum biochemical analysis as serum urea nitrogen was measured according to (35), uric acid according to the method described by (36), creatinine (37), L-Aspartate amine transferase (AST) and L-Alanine amine transferase (ALT) activities (38), alkaline phosphatase (ALP) according to (39), serum sodium (Na) was measured according to the colorimetric method of (40) and serum potassium (K) was measured according to the colorimetric method of (41).

Antioxidant enzymes, including glutathione reeducates (GSH), were identified based on (42). SOD (superoxide dismutase) activity was measured using the (43). Malondialdehyde (MDA) was established based on (44).

2.2.6.Statistical analysis

The results were statistically assessed using a computer program called One-way ANOVA and shown as mean ± SD. According to (46) while, differences between treatments were deemed significant ($P \leq 0.05$).

3.RESULTS AND DISCUSSION

The present work was investigated effect of papaya leaves and seeds on biological parameters of nephropathic albino rats induced by Gentamicin

The results of the current study are presented as the follows:

Leaves and seeds of papaya were produced into powder and subjected to proximate analysis Table (1).The proximate analysis shows that the moisture, protein, Ash, fat, fiber and carbohydrate contents of papaya leaves as dry weight (D/W) were 57.01±0.20, 6.50±0.30, 2.18±0.08, 2.01±0.04, 3.10±0.06 and 29.20 % respectively .While, the moisture, protein, Ash, fat, fiber and carbohydrate contents of papaya seeds as dry weight (D/W) were 7.34±0.05, 9.65±0.45, 10.25±0.06, 27.72±0.02, 21.25±0.04 and 23.34% respectively.

The result of the data analysis of the samples revealed that moisture and carbohydrate in the papaya leaves were 57.01% and 29.20% respectively. The mean value for protein was 6.50% while crude fiber was 3.10%. Other proximate parameters were 2.10% and 2.18% respectively for ash and fat. The proximate composition of the papaya leaves as shown in Table (1) revealed that it is a source of carbohydrate (29.20%) and has been supported by OECD which (47)affirmed that the major nutritional component of papaya leave is carbohydrate. The high moisture

(57.01%) is in conformity with United State Department of Agriculture's [16] report, that moisture content of leaves is high and had mean value of 83%, this is however higher than the value reported in this present study.

Low protein was observed in papaya leaves (6.50%), however it could be used to supplement other sources of protein to make up the 11 RDA for children above 6 months especially children that are prone to protein energy malnutrition. Crude fibre (3.10%) was observed in this study, the presence of crude fibre makes for good bowel movement as well as aid nutrient absorption (48)

Ash content was (2.18%), ash is an indication of mineral contents of foods (49) and hence the ash content of the extract has implication for mineral values. The fat concentration in the sample was 2.01% which is quite low and does not meet daily recommended allowance of fat, this may not be adequate for body needs, as a source of fatty acids which have been associated with wound healing (50) and immunity (51), it is still considered as very useful in this regard when added to other foods that has a better source of fat .On the contrary The proximate analysis of C. papaya seed powder showed that fat is the most abundant nutrient followed by carbohydrate and fiber in that order. The sample has moderate protein and ash. Also moisture content was low(52).

Table (1): Chemical composition of papaya leaves and seeds

Components	Papaya Leaves (D/W)	Papaya Seeds (D/W)
Moisture	57.01±0.20	7.34±0.05
Protein	6.50±0.30	9.65±0.45
Ash	2.18±0.08	10.25±0.06
Fat	2.01±0.04	27.72±0.02
Crude fiber	3.10±0.06	21.25±0.04
Carbohydrate	29.20±0.16	23.34±0.04

Total carbohydrates were calculated by difference DW= Dry Weight

Effect of papaya leaves and seeds on (BWG), (FI) and (FER) of nephropathic rats

Data presented in Table (2) Present the mean value of body weight gain (BWG), feed intake (FI) and feed efficiency ratio (FER) of nephropathic rats on papaya leaves, seeds and their mixtures.

It is evident that the positive control group recorded a lower value with significant differences, whereas the negative control group reported a bigger body weight gain (BWG) ; being 20.85 and 30.70 g, respectively. However, 5% of papaya leaves showed the lowest value with notable variations, whereas 5% of the combo leaves and seeds showed the largest body weight gain among the treatment groups. 34.50 and 15.00g were the mean values, respectively.

Additionally, Table 2 showed that the positive control group's mean value of FI (FI) was significantly lower than that of the negative control group, at 120.10 and 13.30.50g, respectively, which was significant (p≤0.05). It was evident that 5% of the mixture leaves and seeds had the maximum feed intake of the treatment groups, whereas 5% of papaya seeds had the lowest value, with notable variations. The average weights were 16.80g and 21.31g, respectively.

Conversely, the positive control group's mean FER value was 0.0480, which was lower than that of the negative control group, and 0.0392. displaying a notable contrast between them. Notably, the 5% of combination papaya leaves and seeds had the highest feed efficiency ratio among the treated groups (nephropathic groups), whereas the 5% of papaya leaves had the lowest value. The average values were, respectively, 0.0500 and 0.0260.

The papaya leaves and seeds (P≤0.05) affected the average daily feed intake, weight gain, feed conversion ratio and cost per kg live weight of weaner pigs (53).The higher weight gain of may be attributed to the action of papain, a natural enzyme occurring on the leaves of papaya plant which helps in the digestion of proteins in the digestive tract and synthesis of vitamins C and E (54- 58). According to (59), the major active ingredient recorded in pawpaw seed such as caprine, chymopapain, and papain are enzymes capable of enhancing appetite and metabolism and this appears to be responsible for increased daily feed intake while benzyl isothiocyanate has active anthelminthic activity.

It was shown that papaya seed meal was able to improve the production performances of poultry, including increasing growth rate, egg production, and feed efficiency of poultry. In broiler chickens, (60) showed that incorporation of 1% papaya seed powder in rations increased the final body weight and feed intake. In line with this, (61) reported that incorporation of 1.2% papaya seed in diets increased body weight gain, reduced feed intake, and improved feed efficiency of Raja ducks. Likewise, (62) documented that inclusion of 0.5, 1.0, and 2% papaya seed in rations increased daily weight gain of pullet when compared with the control.

Table (2): Effect of papaya leaves and seeds on (BWG), (Fl) and (FER) of nephropathic rats

Parameters Groups	BWG (g)	Feed Intake (g/ day/ rat)	FER (%)
Group (1): negative control	30.70±0.30a	20.10±2.20a	0.0480±.15a
Group (2): positive control	20.85±0.10e	13.30±.03e	0.0392±0.13d
Group (3): 2.5%papaya leaves	25.73±0.50c	19.35±0.20b	0.0399±0.23a
Group (4): 5%papaya leaves	15.00±0.15c	18.60±0.30b	0.0260±0.15b
Group (5): 2. 5%papaya seeds	28.83±0.30e	19.33±0.50b	0.0400±0.15a
Group (6): 5%papaya seeds	18.60±0.20e	16.80±0.40d	0.0280±0.30b
Group (7): 2.5%Mixture (leaves and seeds)	29.23±0.40a	20.10±0.93a	0.0440±0.50a
Group (8): 5%Mixture (leaves and seeds)	34.50±0.40a	21.31±0.98a	0.0500±0.71a
LSD	1.21	1.15	0.02

** Each value represents the mean ± SD of three replicates.*
Mean in the same column with different superscript letters are different significantly (p ≤ 0.05)

Effect of papaya leaves and seeds on glucose levels of nephropathic rats

The effect of papaya leaves, seeds, and their mixes on the glucose level of nephropathic rats is demonstrated by the data in Table (3). It is evident that the positive control group had the highest glucose levels, whereas the negative control group had the lowest value with notable variations. 180.5 and 98.00 mg/dl were the corresponding mean values.

However, there were notable variations between the treatment groups' maximum and lowest glucose levels, with the former occurring for 2.5% papaya leaves and the latter for 5% papaya (leaves and seeds). 146.25 and 100.11 mg/dl were the respective mean values. This is consistent with in vivo research that shown the capacity of papaya leaf to lower hyperglycemia in diabetic rats. (63-65). The phytochemicals found in papaya, including quercetin, transferulic acid, and kaempferol, are responsible for this impact because they have a high binding affinity for the IRS-2 and GLUT-2 proteins, which in turn promotes the uptake of glucose. (66-68).

Table (3): Effect of papaya leaves and seeds on glucose levels of nephropathic rats

	Glucose (mg/dl)
Group (1): Negative control	98.00±0.30d
Group (2): positive control	180.50±0.21 a
Group (3): 2.5% papaya leaves	146.25±0. 50 b
Group (4): 5% papaya leaves	140.40±0.20 b
Group (5): 2. 5% papaya seeds	142.40±0.60 b
Group (6): 5% papaya seeds	138.20±0.11 c
Group (7): 2.5% Mixture (leaves and seeds)	130.50±0.10c
Group (8): 5% Mixture (leaves and seeds)	100.11±0.40 d
LSD	4.58

** Each value represents the mean ± SD of three replicates.*
Mean in the same column with different superscript letters are different significantly (p≤0.05)

Effect of papaya leaves and seeds on kidney functions levels of nephropathic rats

Data obtained in Table (4) explains the effect of papaya leaves, seeds and its mixture on kidney functions (on plasma urea, creatinine, and uric acid) of nephropathic rats.

As it appears the mean value of urea of positive control group was higher than negative control group, being 89.6 and 37.3 mg/dl, respectively, Which indicates that significant difference between them. On the other hand, the highest urea levels of treated groups recorded for 2.5% papaya seeds, while the lowest value recorded for 5% mixture of papaya leaves and seeds with significant differences. The mean values were 55.03 and 38.27 mg/dl, respectively.

In the instance of uric acid, the obtained results showed that the highest uric acid levels recorded for positive control group, while negative control group recorded the lowest value with significant differences. The mean values were 4.84 and 1.82 mg/dl, respectively. It could be seen that the highest uric acid levels of treated groups recorded for 2.5% papaya seeds, while the lowest value recorded for 5% mixture of papaya leaves and seeds with significant differences. The mean values were 2.50 and 1.4 mg/dl, respectively.

It was seen from the data that the positive control group had higher creatinine levels, whereas the negative control group had the lowest value with notable variations. 8.10 and 3.30 mg/dl were the corresponding mean values. Significant differences were found between the treatment groups' highest creatinine levels, which were recorded for 2.5% papaya seeds, and the lowest value, which was recorded for a 5% blend of papaya leaves and seeds. 5.60 and 3.60 mg/dl, respectively, were the mean values.

Our results undoubtedly showed that papaya had an impact on the blood levels of the renal biomarkers and, consequently, the kidney's functional performance, given that serum urea, creatinine, and uric acid levels are indicative of the degree of renal impairment (68, 69).

In specific terms, the observed significant increase (p≤0.05) in the levels of urea, creatinine, and uric acid levels across the groups is consistent with earlier findings reported by (70) that rats treated with papaya seed extract, showed increased serum levels of urea, creatinine and uric acid, but contradicts the report by (71) that renal biomarkers were not significantly affected following an exposure to aqueous papaya seed extract for 2 weeks. The contradiction with the later report however, might be due to differences in the mode administration, nature of the substance in question, and the duration of study which in this case, was 6 weeks. From the foregoing therefore, it is obvious that the dosage dependent and statistically significant increases in urea, creatinine and uric acid levels, irrespective of the type, indicates that Carica papaya's has capacity to induce alterations in renal function. Thus, there is an urgent need to regulate the inclusion of papaya seeds in herbal preparations, particularly in those used for the management of kidney diseases (72)

Table (4): Effect of papaya leaves and seeds on kidney functions levels of nephropathic rats

Parameters Groups	Urea nitrogen (mg/dl)	Creatinine (mg/dl)	Uric acid (mgdl)
Group (1): negative control	37.3±3.52 d	3.30±0.27 d	1.82±0.19 d
Group (2): positive control	239.6±9.62 a	8.10±1.56 a	4.84±1.09 a
Group (3): 2.5%papaya leaves	45.96±1.30 c	4.72±0.53 c	2.30±0.50 b
Group (4): 5%papaya leaves	41.25±0.30 c	4.00±0.14 c	2.00±0.20 b
Group (5): 2.5% papaya seeds	55.03±1.10 b	5.60±0.21 b	2.50±0.40 b
Group (6): 5% papaya seeds	53.27±0.50 b	4.73±0.70 c	2.20±0.20 c
Group (7): 2.5% Mixture (leaves and seeds)	40.96±1.30 c	3.90±0.53 d	1.83±0.20 d
Group (8): 5% Mixture (leaves and seeds)	38.27±2.30 d	3.60±0.14 d	1.4±0.1 e
LSD	3.40	1.21	1.18

** Each value represents the mean ± SD of three replicates.*

Mean in the same column with different superscript letters are different significantly (p ≤ 0.05)

Data obtained in Table (5) demonstrates the effect of papaya leaves, seeds and their mixtures on liver functions (AST, ALT and ALP) of nephropathic rats.

As can be seen, there was a considerable difference between the positive and negative control groups, with the positive control group's mean AST value being 80.30 and 20.00 U/L, respectively. Conversely, 2.5% papaya seeds had the highest AST liver enzyme among the treatment groups (nephropathic groups), whereas 5% papaya leaf and seed mixture had the lowest value with significant differences. The mean values were 46.90 and 25.10 U/L, respectively.

It goes without saying that the highest ALT liver enzyme levels recorded for positive control group, while negative control group recorded the lower value with significant differences. The mean values were 93.8 and 30.50U/L, respectively.

It was acquired that the highest ALT liver enzyme of treated groups recorded for 2.5% papaya seeds, while the lowest value recorded for 5% papaya (leaves and seeds) with significant differences. The mean values were 71.70 and 44.20U/L, respectively.

In the instance of ALP liver enzyme, the highest levels recorded for positive control group, while negative control group recorded the lower value with significant differences. The mean values were 53.6 and 5.50 U/L, respectively. The information that was acquired was the highest ALP liver enzyme of treated groups recorded for 2.5% papaya seeds, while the lowest value recorded for 2.5% papaya (leaves and seeds) with significant differences. The mean value were 9.93 and 5 U/L, respectively. This result agree with (73) there were a significantly decreased in serum ALT and AST at two levels for leaves and seeds powder at 10%.

Table (5): Effect of papaya leaves and seeds on liver functions levels of nephropathic rats

Parameters Groups	ALT (u/l)	AST (u/l)	ALP (u/l)
Group (1): negative control	37.5±1.7 e	20±2.20 d	5.8±0.80 d
Group (2): positive control	133.8±0.30 a	80.30±2.03 a	53.6±17.0 a
Group (3): 2.5%papaya leaves	60.00±0. 80 c	41.45±1.20 c	7.81±0. 50 c
Group (4): 5%papaya leaves	55.75±0.50 d	39.25±1.30 c	5.50±0.80 d
Group (5): 2.5% papaya seeds	71.70±0.90 b	46.90±1.10 b	9.93±0.90 b
Group (6): 5% papaya seeds	64.80±0.40 c	40.80±1.40 c	8.20±1.20 c
Group (7): 2.5% Mixture (leaves and seeds)	40.10±0.10e	28.20±3.03d	5.00±0.40c
Group (8): 5% Mixture (leaves and seeds)	39.20±0.21 e	25.31±3.6 d	5.55±0.70 d
LSD	2.40	2.56	1.21

** Each value represents the mean ± SD of three replicates.*
Mean in the same column with different superscript letters are different significantly (p ≤ 0.05)

Data obtained in Table (6) displays the effect of papaya leaves, seeds and their mixtures on total protein of nephropathic rats. It is clear to observe that the highest total protein levels were recorded for negative control group, while positive control group recorded the lower value with significant differences. The mean values were 7.80 and 3.91 g/dl, respectively. It could be seen that the highest total protein of treated groups recorded for 5% Papaya (leaves and seeds), while the lowest value recorded for 2.5% Papaya seeds with significant differences. The mean values were 6.95 and 3.60 g/dl, respectively. These findings are consistent with (74), who discovered that following six months of papaya intake, the concentration of total protein increased, albeit within the reference range. This increase was brought on by an increase in albumin, which suggests that the liver's synthetic function has improved.

Table (6): Effect of papaya leaves and seeds on total protein levels of nephropathic rats

	STP (g/dl)
Group (1): Negative control	7.80±1. 5 a
Group (2): positive control	3.91±1.36d
Group (3): 2.5%papaya leaves	5.60±1.35 b
Group (4): 5%papaya leaves	6.30±1.50 b
Group (5): 2. 5%papaya seeds	3.20±1.10 d
Group (6): 5% papaya seeds	4.45±1.50 c
Group (7): 2.5% Mixture (leaves and seeds)	6.50±1.15 a
Group (8): 5% Mixture (leaves and seeds)	6.90±1.50 a
LSD	1.50

** Each value represents the mean ± SD of three replicates.*
Mean in the same column with different superscript letters are different significantly (p≤0.05)

The obtained results indicated that the positive control group had the lowest value with notable variations, while the negative control group had the greatest red blood cell levels. 6.40 and 2.80×106/ mm3, respectively, were the mean values. It was evident that the treated groups' red blood cell levels were highest when 5% papaya leaves and seeds were combined, and lowest when 2.5% papaya seeds were used. with notable distinctions. 3.47 and 6.10×106/mm3 were the respective mean values.

Clearly, it is evident that the highest white blood cell levels were recorded for positive control group, while negative control group were recorded the lowest value with significant differences. The mean values were 12.50 and 6.20×103/ mm3, respectively. Observations revealed that the highest white blood cell levels of treated groups recorded for 2.5% papaya seeds, while the lowest value were recorded for 5% mixture of papaya leaves and seeds with significant differences. The mean values were 10.30 and 6.50×103/ mm3, respectively. These findings are consistent with (75), which found that dietary treatment with 5% dried papaya seed (DPS) did not result in any significant changes (p > 0.05) in white blood cells, leucocytes, or neutrophils. In addition, (76). who stated that the birds with the lowest WBC were fed 15% dry papaya seed. This might be because the meal contained excessive amounts of anti-nutritional factors that triggered the immune systems.

Table (7): Effect of papaya leaves and seeds on red blood cell and White blood cell levels of nephropathic rats

Parameters Groups	RBCs (10^6/mm3)	WBCs (10^3/mm3)
Group (1): negative control	5.40±0.20 a	6.20±0.60 f
Group (2): positive control	1.80±0.30 c	12.50±0.70 a
Group (3): 2.5%papaya leaves	3.20±0. 80 b	8.30±0. 90 c
Group (4): 5%papaya leaves	3.75±0.50 b	8.50±0.40 c
Group (5): 2.5% papaya seeds	2.47±0.90 c	10.30±0.30 b
Group (6): 5% papaya seeds	3.15±0.40 b	8.60±0.50 c
Group (7): 2.5% Mixture (leaves and seeds)	4.85±0.10a	7.10±0.23d
Group (8): 5% Mixture (leaves and seeds)	5.10±0.21 a	6.50±0.11 e
LSD	1.04	1.01

** Each value represents the mean ± SD of three replicates.*
Mean in the same column with different superscript letters are different significantly (p ≤ 0.05)

The collected results showed that the positive control group had the highest serum triglyceride levels, whereas the negative control group had the lowest value with significant differences. 135.61 and 65.80 mg/dl were the corresponding mean values. However, 2.5% papaya leaves gave the treated groups the greatest serum triglyceride levels, whereas 5% papaya leaves and seeds gave the lowest value. without any noticeable variations. They were 89.60 and 67.10 mg/dl, respectively, on average.

When it comes to serum total cholesterol levels, it can be said that the positive control group had the highest levels while the negative control group had the lowest. with notable distinctions. 173.90 and 80.60 mg/dl were the mean values, respectively. With notable variations, it was shown that the treatment groups' serum cholesterol levels were highest for 2.5% papaya leaves and lowest for 5% papaya (leaves and seeds). 111.92 and 88.20 mg/dl were the corresponding mean values.

Regarding blood HDL levels, it can be inferred that the negative control group had the greatest levels, while the positive control group had the lowest, with notable variations. 50.07 and 30.22 mg/dl were the corresponding mean values.

It was found that the maximum serum HDL values of treated groups recorded for 5% papaya (leaves and seeds), while the lowest value recorded for 2.5% papaya leaves with significant differences. 45.23 and 38.53 mg/dl were the corresponding mean values.

One could deduce that, in terms of serum LDL levels, the positive control group had the highest values, while the negative control group had the lowest. with notable variations. In each case, the mean values were 18.92 and 95.30 mg/dl. With notable variations, it was shown that the treatment groups' serum LDL levels were highest for 2.5% papaya leaves and lowest for 5% papaya (leaves and seeds). The average levels were, respectively, 45.20 and 25.60 mg/dl.

In case of serum VLDL levels, it could be concluded that the highest serum VLDL levels were recorded for positive control group, while negative control group recorded the lowest value with significant differences. The mean values were 29.75 and 12.58 mg/dl, respectively.

It was noticed that the highest serum VLDL levels of treated groups recorded for 2.5% papaya leaves, while the lowest value recorded for 5% papaya (leaves and seeds) with significant differences. The mean values were 18.66 and 13.95 mg/dl, respectively. This results are agreement with (77) who found the greatest VLDL in rats fed 5% papaya leaves

Table (8):Effect of papaya leaves and seeds on serum triglycerides, total cholesterol, HDL, LDL and VLDL levels of nephropathic rats

Parameters Groups	T.G (mg/dl)	Chol. (mg/dl)	HDL (mg/dl)	LDL (mg/dl)	VLDL (mg/dl)
Group (1): Negative control	65.80±0.35e	80.60±0.70e	50.07±2.02a	18.92±0.98h	12.58±0.005d
Group (2): positive control	135.61±0.65a	173.90±0.34a	30.22±1.68e	95.30±1.60a	29.75±0.008a
Group (3): 2.5%papaya leaves	89.60±0.15b	111..92±0.15b	38.53±1.11d	45.20±2.40b	18.66±0.061b
Group (4): 5%papaya leaves	84.70±0.30b	100.10±0.60c	41.40±1.60c	35.90±2.10c	17.78±0.001b
Group (5): 2.5%papaya seeds	82.20±0.60b	105.58±0.29c	40.23±1.12c	40.05±1.80b	17.22±0.004b
Group (6): 5%papaya seeds	71.45±0.58c	100.00±0.90c	42.26±0.60b	36.05±0.86c	15.09±0.091c
Group (7): 2.5% Mixture (leaves and seeds)	76.50±0.15c	95.10±0.50d	45.23±2.00b	25.60±2.10d	16.30±0.009c
Group (8): 5% Mixture (leaves and seeds)	67.10±0.50d	88.20±0.25d	47.15±2.00a	19.80±2.10e	13.95±1.003d
LSD	3.60	3.98	3.6	3.5	2.90

** Each value represents the mean ± SD of three replicates.*
Mean in the same column with different superscript letters are different significantly (p≤0.05)

4. Conclusion

Papaya leaves and seeds have antioxidant activities and protective effect against Gentamicin-induced CKD that they reduced elevated levels of serum creatinine, urea nitrogen and uric acid. However, further human studies are needed.

Conflict of Interest

The authors declare that there is no conflict of interest.

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التأثير المحتمل لبذور وأوراق البابايا في الفئران المصابة بخلل في الكلي المستحث بالجنتاميسين

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الملخص العربي:
تهدف هذه الدراسة إلى معرفة التأثير الوقائي للتغذية على وجبات قياسية تحتوي على كلا من أوراق وبذور البابايا وخلطيهما بنسبة 2,5، 5% من حدوث الالتهاب الكلوي الحاد. وقد استخدمت في هذه الدراسة 48 فأر من نوع الألبينو أوزانهم 150±5 جم تم تقسيمهم إلى 8 مجموعات متساوية يحتوي كل منها على (6 فئران) تم تغذية المجموعة الأولى على الوجبة القياسية العادية واستخدمت كمجموعة ضابطة سالبة. كما تم تغذية المجموعة الثانية على الوجبة القياسية واستخدمت كمجموعة ضابطة موجبة. المجموعات الست الأخرى تم تغذيتهم على الوجبة القياسية بالإضافة إلى أوراق وبذور البابايا وخليطيهما بنسبة 2.5، 5% على التوالي لمدة 4 أسابيع. وخلال فترة التجربة تم تسجيل المتناول الغذائي اليومى ووزن الفئران المكتسب. تم حقن المجموعة الضابطة الموجبة والمجموعات الست التى تغذت على أوراق وبذور البابايا وخليطيهما بنسبة 2.5، 5% بالجنتاميسين (8مل/كجم من الوزن) وذلك لإحداث الالتهاب الكلوي الحاد. تم ذبح الفئران وجمع عينات الدم وتحليلها وتقدير كلا من وظائف الكلى والكبد والألبيومين وأملاح الصوديوم والبوتاسيوم وإنزيمات الأكسدة. وأظهرت الوجبات المختبرة تأثيرا إيجابيا على وظائف الكلى وتحسن الخلل الناتج عن الحقن بالجنتاميسين وخاصة في حالة الخليط بنسبة 5%.

الكلمات الكاشفة: الالتهاب الكلوي الحاد، البابايا، وظائف الكلي، الفئران

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