

EFFECT OF *CASSIA PODOCARPA* ON KIDNEY OF FEMALE RATS

¹ AREMU OI., ² ADEFEMI OA

1. Dept of Pharmaceutics and Pharmaceutical Technology, Olabisi Onabanjo University, Shagamu.
2. Formerly of Dept. of Pharmacology, Faculty of Pharmacy, OAU, Ile-Ife.
Now of Federal Environmental Protection Agency (FEPA, Abuja).

ABSTRACT

A study had been carried out on possible toxicity effects of commonly used laxative, *Cassia* leaves among Herbal practitioners. Since there has been no standardized dosage regime the study becomes imperative.

In this study the female rats were given a standard dose of 2.5k/kg body weight of the animals once a week for four consecutive weeks.

Histological examination revealed that the test animals had tubular degeneration showing nephrotoxicity as an adverse effect of *Cassia* leaves. Caution is hereby suggested in the indiscriminate administration of *Cassia* leaves infusion.

INTRODUCTION

Cassia podocarpa Guille and Perr is a plant related to senna. Senna is one of the oldest and best known of the wild purgatives. It stimulates the muscular coat of large intestine. Senna comes from *Cassia acutifolia* from Egypt or from *Cassia angustifolia* from India.

The genus *cassia* belongs to the family of leguminosae which is the second largest family of the flowering plants. The leguminosae family is further subdivided into three sub families namely: caesalpinaceae, mimosoidae and papilionaceae (8) Caesalpinaceae has its largest genus which contains from 450- 700 species as indigenous to west Africa (5) while about 33 species are found growing in Nigeria (3)

Cassia podocarpa is a shrub that can attain a height of 5m with paired pinnate leaves that bear light yellow flowers and green pods around the month of December. It grows in the Savannah forest around villages and in farmlands in many areas of Ghana (4)

It is known in local folk medicine as peagoro (Ashanti), Sempe (Twi) or Ashanti (Ga), the leaves, fruits or roots of the plant are boiled and drunk as a purgative. The plant and its laxative properties are well known to Ghanaian herbalists.

In the present work, an investigation into toxic effect on the kidney an organ involved in metabolism was carefully studied.

MATERIALS AND METHOD

In the process of the experiment, the following materials were used; white female albino rats, feeding bottles. Animal weighing balance (Ohaus), curved metal rat feeding needles, metallic ear punch, sieve size 60. Gallenkamp hot air circulating oven (Griffin oven). Beakers (Pyrex), measuring cylinders 50ml. Rat cages, leaves of *Cassia podocarpa*, microscope, slides, and dissecting set.

Collection and Preparation of Plant Material:

The leaves of *Cassia podocarpa* were collected at different locations outside the Obafemi Awolowo University, Ile-Ife Campus. The leaves collected were dried at 40°C in an oven for 12 hours. The dried leaves were crushed and stored in polythene bags until ready for use. The crushed leaves were finely grinded using pestle and mortar. This was then sieved using a mesh 60 sieve.

Preparation of an infusion:

15g of the ground leaves was accurately weighed into a beaker and an infusion of it was made soaking in 160ml hot water (100°C) for 15 minutes.

Procedure

Female white albino rats, weighing between 100g – 150g were used. Curved metal feeding needle attached to a syringe was used to dose the animals. This was passed down the animals throat using the

appropriate volume of infusion.

The dosing was carried out once in a week for four weeks in test animals while the control was not dosed. A standard dose of 2.5g/kg body weight of the animals was administered to the test animals once in a week.

Diet

The rats were fed daily with rat cubes (Ladokun feeds) and they were supplied with tap water regularly. The rats, four in number were divided into two group i.e. the test group which consists of three female rats and the control group which consists of only one female albino rat. The animals were supplied with water and rat cubes constantly. The bedding inside the cages was made of sawdust which was changed weekly. Those cages were kept in the Faculty of Pharmacy, animal House. Rats were permanently identified by means of the standard ear hole punch and ear numbering system.

Four weeks after the administration of *Cassia podocarpa* that is, at the beginning of the fifth week, the rats were weighed and sacrificed by anaesthesia using diethylether.

Blood was obtained from jugular veins using dissecting set. Later a whole dissection of the animal body was carried out and the kidneys of the animals including the control animal were removed for weight determination. Slides photographs of the slides under microscope were taken to see the possible effect of the administered drug.

RESULTS

During the administration of *Cassia podocarpa* for four weeks, the weights of the animals were recorded as shown in Table 1. The weights at sacrifice and organ weight were determined and recorded as shown in the same table.

The variation in the weights have been properly documented. The photomicrograph of the kidney of the control animal (Fig. 1) clearly showed that the major tubules i.e. proximal convoluted tubules, distal convoluted tubules and collecting tubules remained intact. In the test animals, degeneration of the tubules occurred (Fig.2).

In addition, there was a breakdown in the architecture of the cells. Also, figure 2 shows that there was an abundance of proliferating cells in the degenerating tubules. These proliferating cells which are phagocytic in action, increased in number at the tissue sites in accordance to the degree of damage done to the tissue. Histopathological sections of the kidneys from animals that received *cassia podocarpa* (2.45g/kg) also showed that tissue degeneration had progressed so much as to cause vacuolization, as indicated by the presence of vacuoles. In addition, there was some regeneration of the tubules.

Also, one noticed that there tends to be regeneration of new tubules as can be seen in (Fig.2). But one obvious fact is that the rate of regeneration is far lower than the rate of degeneration of tubules.

Table 1. Administered *Cassia podocarpa* effect analysis.

Dates	Rats	Body weight	Volume administration (mls)	Body weight sacrifice 5 th week	Kidney weight at body weight. week	Kidney to- mean Ratio. at sacrifice 5 th week
1 st week	C	123.0	3.3	C. 138.5	0.139	0.001
	T ₁	124.0	3.3	T ₁ 93.0	0.186	0.002
	T ₂	108.5	2.9	T ₂ 124.0	0.175	0.002
	T ₃	112.0	2.9	T ₃ 139.0	0.278	0.002
2 nd week	C	113.0	3.0			
	T ₁	137.5	3.6			
	T ₂	112.5	3.0			
	T ₃	120.0	3.2			
3 rd week	C	127.0	3.4			
	T ₁	133.0	3.5			
	T ₂	-	-			
	T ₃	114.0	3.0			
4 th week	C	116.0	3.1			
	T ₁	111.0	2.9			
	T ₂	131	3.0			
	T ₃	116.5	3.1			

Fig. 1: The Photomicrograph of the kidney of the control Animal

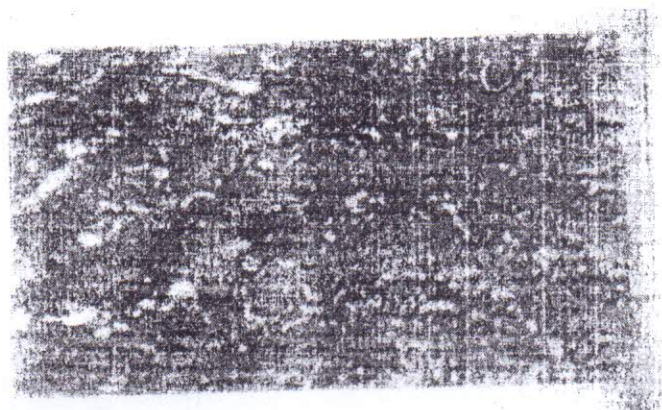


Fig. 2: The Photomicrograph of the kidney of treated Animal



DISCUSSION AND CONCLUSION

It has been observed that from the test carried out there is a tubular degeneration. It is interesting to this author to view the significance of this effect in the perspectives of the functioning capability of the tubules particularly proximal tubules. A literature review of the functions has indicated that the proximal tubular cells are involved in the transport of salt and water from lumen to the interstitials by active transport mechanism at the base of the cells.

The tubules of the kidney reabsorbs or secrete a very large amount of substance, both endogenous and exogenous, and many different mechanisms and enzyme systems are involved. More than 98% of the filtered load of glucose is against concentration gradient and is therefore an active process, but there is also evidence for some passive transport. The action of phlorzoin, a glucoside which specifically blocks glucose transport, has given some information, since it apparently prevents access of intratubular glucose to the interior of the cells by its action at the luminal surface.

What must be noted from the available facts is that with the cytotoxicity of the tubules, the function of salt and

water transfer is impaired which in turn can lead to an increased in blood pressure and consequently hypertension. The Odema of the heart can also be as a result of this impaired function due to accumulation of fluid in the body.

Apparent high glucose content in the plasma is imminent Glucose is a threshold substance which has a tubular maximumTM. At normal plasma levels, it is reabsorbed almost completely in the first half of the proximal tubule (9).

Urea is one of the end - product of protein metabolism, but as well as appearing in the urine simply as substance to be excreted, it plays a part in the mechanism of the concentration of urine. It was first noticed that rats which had been fed urea were able to produce a more concentrated urine than rats fed an equivalent amount of inorganic electrolytes (2). Similarly in both animals and man, maximum concentrating ability of the kidney is reduced on a low protein diet. (1,7) In healthy adult men. It was found that feeding either urea itself or protein increased the maximum concentrating ability and also the response to Pitressin (antidiuretic hormone) (1). Urea is a toxic

waste product and in the process of dysfunction of the tubules accumulation of urea occurs in the body causing damage to other body organs.

The products of normal metabolism produce an excess of acid substances so that in order to maintain the normal pH of the internal environment it is necessary that such substances should be buffered and should be removed as rapidly involved as the carbonic acid- bicarbonate buffer system. The carbonic acid component (depending on $p\text{CO}_2$) is regulated by the respiratory system while the kidney regulates the bicarbonate concentration by its control of the reabsorption of filtered bicarbonate. The essential process concerned in the latter is the exchange of Na^+ for H^+ ions between the tubular lumen and interstitial space- (6).

This function of the kidney in buffering activities of the excess acid is impaired with tubular necrosis. A possible consequence of this is possibility of systemic acidosis in the body.

As indicated earlier, the contents of leaked tubular cells were observed to be toxic to other structures in the kidney. This generalised damage to the whole structure of the organ could lead to collapse of the kidney function, some of which have been enumerated above.

It is pertinent to look at one or two mechanisms by which nephrotoxicity occur. Nephrotoxicity may be due to any of the following mechanisms or combinations thereof direct tubular damage may occur. For example, heavy metal poisoning may cause acute tubular necrosis. If the toxic proximal exposure is quantitatively less, nephrotic syndrome or proximal tubular injury with features of the Fanconi syndrome may occur. Tubular necrosis may also result indirectly from hemodynamic changes, for example after excessive diuresis, hypersensitivity reaction, or hemolysis, or from hypotonia due to methemoglobin formation as occur with ethylene glycol dinitrate poisoning, or from myoglobinuria as it may occur after electro- shock therapy. Real tubular toxicity may also occur indirectly through such mechanisms as kaliopenic nephropathy.

The various weights recorded in table I is another parameter used to confirm the nephrotoxicity of *Cassia podocarpa*. Throughout the period of administration and even at sacrifice, the weights of the animals were fluctuating. The only explanation for this apart from hypothesising the increase or decrease in weights to be due to feeding of the animals, is that one can go further by saying that shrinkage of organs such as liver, kidney and so on might account for decrease in weights at times while accumulation of fluid as a result of kidney dysfunction which may result into Oedema is a plausible reason for increase in weights of the animals in some instances. Above all microscopic analysis confirmed without any iota of doubt the nephrotoxicity of *Cassia*

podocarpa.

The result of kidney - to - body weight ratio of the rats showed that *Cassia podocarpa* did not cause kidney enlargement in female rats in this study. The difference in kidney to body weight ratios among the test animals and the control animal were not much and they are insignificant.

In conclusion, it could be inferred from the results that *Cassia podocarpa* caused nephrotoxicity. However it must be emphasised that there are other substances that do cause nephrotoxicity in a similar manner, examples of such substances are antibiotics, metals, analgesics, leads and so on

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