Original Research Article

Effects of methanolic leaf extracts of *Azadirachta indica* and *Spondias mombin* on the kidneys Histology of Zidovudine stress induced wistar Rats.

ABSTRACT

The kidneys play a role in the maintenance of homeostasis by ensuring the excreton of waste and toxic substances from the body.Oxidative stress could be defined as an imbalance between the production of reactive oxygen species and an inability of the body system to scavenge the presence of free radicals. Intake of certain drugs and toxic substances exposes the kidneys to oxidative stress effects and this may lead to impairment of homeostasis and malfunctioning of the kidney.This study was carried out to access the efficacy in administration of single herbal extracts of either *Azadirachta indica* or *Spondias mombin* when compared to the combination of both herbal extracts in ameliorating the effects of oxidative stress in wistar rats kidney. The study was carried out using 25 male adult wistar rats of weight 180-200 g, the animals were randomly selected and were designated into groups A (Negative control group that received Rat chow and water, group B is the positive control group that received the administration of 450 mg/kg body weight of zidovudine drug, group C is the *A. indica* group that received 450 mg weight of zidovudine drug and 500 mg/kg body weight of methanoic leaf extract /kg body, group D is the *S. mombin* group that received 450 mg/kg body weight of zidovudine drug and 500mg/kg body weight of methanolic leaf extract and group E received a 450 mg/kg body weight of zidovudine and a combination of 500 mg/kg body weight of both methanolic leaf extracts of *A. indica* and *S. mombin* leaf . ~~T~~ administration was carried out once a day using orogastric tube for a period 21 days. At the end of the admnistration, the rats were sacrificed using chloroform inhalation technique and the kidney was fixed in 10% neutral buffered formal saline. Light mcroscopic evaluation of the kidney showed normal histological appearance of the kidney in group A as witnessed by the presence of glomerulus, proximal convoluted tubule (PCT), distal convoluted tubule (DCT), bowmans space (BS), while group B witnessed alterations in the histology of the liver as shown by the presenceof haemorrhage in the glomerulus, shrinkage in the proximal and distal convoluted tubule and shrinkage of the bowmans space, group C and D witnesseed a restoration of the kidneys histology as evidenced by a reduction of haemorrhage in the glomerulus and shrinkage PCT and DCT. Group E showed an enlargement of the Bowmans space and shrinkage of the PCT and DCT. Hence the results proved the efficacy of single administration of herbal extracts in ameliorating the effects of oxidative stress when compared with the combination of the herbal extracts.

KeyWords: ***Azadirachta indica, Spondias mombin*** **leaf, kidneys, Zidovudine**

1.0 Introduction

Medicinal plants are considered as healthy sources for the prevention of various oxidative stress related diseases [1], this is because they are rich in certain phytochemical constituents having anti-oxidative activities such as phenolic compounds and carotenoids [2]. Medicinal plants derived anti-oxidants can protect renal damage through reduction of lipid peroxidation and an increase in the levels of anti-oxidants. [3].Various sections and traditions make use of native

substances as lone herbs, join of plants and union of herbs. Combination of herbs could lead to complications as numerous associations can happen within the person constituent. Complications may arise because of numerous constituent in the native extracts. [4]. However the impacts from plant-plant association are likely uncertain and complex [5], [6], [7], [8], [9], [10], [11], [13], [14]. Oxidative stress can be defined as a disproportion among the system display of active kind air and a functional body capacity to remove the active intermediate or to restore the outcome injury [15]. It is caused when the existence of liberal substance overwhelms the free scavenging mechanism of antioxidants [16]. Oxidative stress is also an important factor which can contribute to kidney damage by increasing the production of oxidants, especially insufficiency of antioxidants defense system [17]. Oxidative stress induced damage on the kidney is associated with an increase in the production of reactive oxygen species [18].

The kidneys are paired bean shaped organs located on the posterior abdominal cavity [19]. It functions in the maintenance of homeostasis through the excretion of metabolic waste products, regulation of extracellular volume, as well as regulation of electrolyte composition and acid base balance [20]. Exposure of the kidney to several drugs, toxic xenobiotics, or chemicals can cause toxic damage to the kidney due to its high rate of blood flow (21). *A. indica* (neem tree) is a native plant of South eastern Asia, and it is distributed in India and other neighboring countries [22]. It is called dogonyaro in Hausa, and Ogwuakuma in Igbo [23]. *A. indica* plays therapeutic role in the management of health due to the presence of rich source of various types of ingredients. Most important active chemical components of *A. indica* is azadirachtin, nimbolin, nimbin, nimbol, sodium nimbinate, gedunin, salannin and quercetin [24]. *A. indica* is rich in phytochemical constituents like azadirachtin, nimbolide and ascorbate which possess significant antioxidant properties, that enables it to scavenge free radicals present in the body [25].

S. mombin belongs to the family *Anarcadiaceae*, and it is one of the medicinal herbs in southern Nigeria [26]. It has several names; it is termed english in plum hog, Yoruba akika, tsardamaster in Hausa, Chabbuh in Fulani and nuskakara in Efik [27]. *Spondias* also possess anthelminthic, antioxidant, antimicrobial and anti-inflammatory actions, sedative and anxiolytic potentials [25, 26 ,27,28,29,30]. Therefore, this study was carried to evaluate the effects of oxidative stress on the histology of the kidney of adult male Wistar rats to compare the impacts of single administration of herbal extracts with the combination of herbal extracts in ameliorating the effects of oxidative stress.

2.1: Materials and methods

The leaves *A. indica* and *S. mombin* were obtained from a local community in Ugep, Yakurr local Government Area of Cross River State, Nigeria. Taxonomical identification was conducted by a botanist in the Department of Botany University of Calabar, Calabar, Nigeria. Both leaves were grounded to powdered form and extracted by cold extraction method using methanol as the solvent for a period of 72 h with the aid of a Soxhlet apparatus. The extract obtained was filtered through Whatman paper 1 and the filtrate was evaporated to dryness on rotary evaporator at (50°C). The extracts were preserved in clean glass container for further use.

1.2 : Animals

This study was approved by the Department Ethics Committee of the University of Calabar, Calabar. Twenty-five male adult Wister rats with an average weight of 200 g were bred in the animal house of the department of Anatomical Sciences and were used for this study. The rats were fed with rat chow, water ad libitum.

2.3: Experimental Protocol.

This study was carried out using twenty-five male adult Wistar rats of average weight 200 g and there were randomly distributed into five sections (A, D, E, B, C, n=5).

Group A the Negative normal group that distilled water and rat chur, **Group B** is the Positive control group that was induced with 450 mg/kg body weight of zidovudine drug for a period of three weeks. **Group C** is the Experimental group that was induced with 450 mg/kg body weight of zidovudine drug for a period of one week and received 500 mg/kg body weight of *A. indica* for a period of two weeks. **Group D** represents Experimental group that was induced with 450 mg/kg body weight of zidovudine drug for a period of one week and received 500 mg/kg body weight of *S. mombin* for a period of two weeks. While **Group E** Experimental group received 450 mg/kg body weight of zidovudine drug for one week and 500 mg/kg body weight of *A. indica* and *S. mombin* for a period of two weeks. At the end of the administration, the animals were anaesthetized using chloroform inhalation technique.

2.4: Stress Induction.

Oxidative stress was induced using Zidovudine obtained from the Plan President Emergency for Aids and liberation section, Teaching University of Calabar Hospital, Calabar town, Cross-River State, Nigeria.

The animals in all the experimental faction collected 450 mg/kg body weight of the zidovudine. The drug was dissolved in 150 ml of distilled water and administered once daily to group C, D, and E for a period of seven days, while group B received the drug for a period of three weeks.

2.5 Determination of body weights of experimental animals

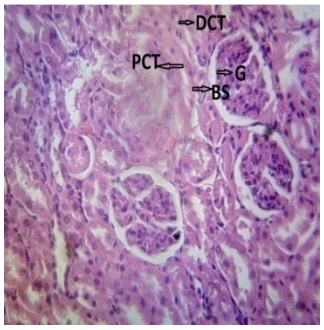
The final weights of the animals were recorded a day after the last dose of administration.

2.6: Collection of experimental specimen

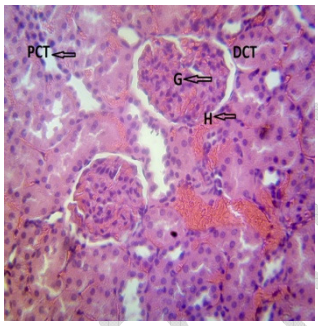
At the end of the administration, the animals were anaesthesized using chloroform inhalation technique. The abdomen was dissected out to access the kidney which was located on top of each Adrenal gland.

3.0 Results

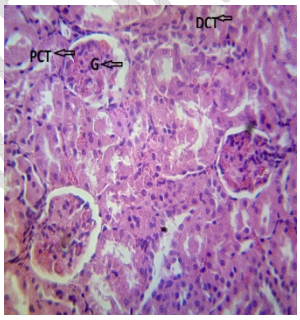
3.1 Histological Observation of the Kidney



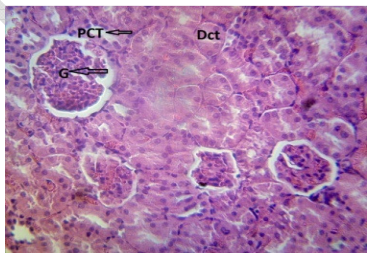
Photomicrograph of normal histology of negative control kidney group showing the presence of the glomerulus(G), distal convoluted tubule (DCT), proximal convoluted tubule (PCT), and bowmans space (BS).H and E ×400.



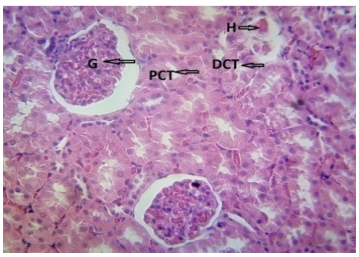
Photomicrograph of positive control group of Rat kidney showing the presence of haemorrhage, shrinkage of distal and proximal convoluted tubule and shrinkage of bowman space (BS). H and E ×400.



Photomicrograph of *A. indica* group kidney group showing Presence of glomerulus, and a restoration of haemorrhage in the distal convoluted tubule (DCT), proximal convoluted tubule (PCT), with a normal bowmans space. H and E ×400.



Photomicrograph of *S. mombin* group showing Presence of glomerulus, and a restoration of haemorrhage in the distal convoluted tubule (DCT), proximal convoluted tubule (PCT), with a normal bowmans space. H and E ×400.



Photomicrograph of combined kidney group showing the presence of dilated bowmans space, with presence of shrinkage in the glomerulus, pct and dct of Rat kidney. (H&E ×400).

4.0 Discussion

This study was carried out to assess the effect of zidovudine drug on the histology of the rat kidney in other to compare the efficacy of single administration of methanolic extracts of *A. indica* or *S. mombin* to a combination of both herbal extracts.

Exposure of the kidney to certain chemical agents or drugs could be manifested by the presence of vascular congestion (glomerulus), inflammatory cell infiltration with the presence of hyaline globule in the collecting tubule [31].

Light mcroscopic evaluation of the kidney showed normal histological appearance of the kidney in group A as witnessed by the presence of glomerulus, proximal convoluted tubule (PCT), distal convoluted tubule (DCT), and bowmans space (BS), while group B witnessed alterations in the histology of the liver as shown by the presence of haemorrhage in the glomerulus, shrinkage in the proximal and distal convoluted tubule and shrinkage of the bowmans space, group C and D witnesseed a restoration of the Kidneys histology as evidenced by a reduction of haemorrhage in the glomerulus and shrinkage PCT and DCT. Group E showed an enlargement of the Bowmans space and shrinkage of the PCT and DCT.

Results of group B and E is similar to the studies carried out by [32] which reported the presence of wider capsular space, congested glomerular tufts, and degeneration of the tubules when treated with cisplastin. Also results of group B,C,D and E is similar to works carried out by [33] on the ameliorative effect of pomegranate on the histopathology of the kidney of diabetic induced oxidative stress. The study revealed the presence of shrinkage and lesions in the bowmans capsule when exposed to oxidative stress, but intake of pomegranate herbal extract rich in anti-oxidants led to a reversal in histological changes of the kidney.

The restoration in the histology of group C, and D may be due to the presence of anti-oxidants present in the above herbal extracts, while the widening of the bowman space may be because of the drug on the kidney histology. Studies carried out by [34] showed that methanolic leaf extract of *A. indica* can ameliorate the effects of oxidative stress on the kidney. This may be due to its antinephrotoxic potential.

Conclusion

The results of the study proved that single administration *A. indica* and *S. mombin* ameliorated the effects of oxidative stress on the kidney histology of male Wistar rats when compared with the combination of both herbal extracts in ameliorating the effects of oxidative stress on the kidney. The effects of combined herbal therapy could not be compared with the single administration of the herbs, this may be due to interaction between the phytochemical components of both herbal extracts.

Ethical Approval

This study was approved by the Department Ethics Committee of the University of Calabar, Calabar.

Consent: NA

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