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A Comparative Study on the Effects of Excessive Consumption of Ginger, Clove, Red Pepper and Black Pepper on the Histology of the Kidney

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Abstract: Allergic reactions have been associated with cloves, black pepper and mast cells. The presence of mast cells and renal fibrosis has also been reported and in line with an ongoing investigation on the effects of excessive consumption of *Yaji* - the complex Nigerian meat sauce, this study was designed to correlate the individual and combined effects of the excessive consumption of *Yaji*-spices (ginger, clove, red pepper and black pepper) on the histology of the kidney, using 12 adult rabbits. The animals were divided into six groups of two (2) each (A, B, C, D, E and F). Groups B, C, D, E and F were the test groups whereas group A is the control. For 21 days, test group B was fed with feed and a combination of all the *yaji*-spices (3g each) while test groups C, D, E, and F were fed with feed and 3g each of the spices per day respectively. The histological observations showed that there were no effect on the kidneys in test groups C and E whereas in B, D and F, there were distinct round basophilic bodies in the interstitium of the renal cortex, which were suspected to be mast cells, and hence, implicating cloves and black pepper. This result suggests a possible mechanism of renal damage that is clove and black pepper feeding-induced, duration dependent and with mast cell mobilization into the kidney, that may then trigger renal fibrosis.

Key words: *Yaji*-spices, basophilic bodies, mast cells, renal fibrosis

Introduction

In recent years, the effect of excessive consumption of *Yaji* –the complex Nigerian meat sauce, has been under investigation (Nwaopara *et al.*, 2004; 2007a; 2007b). The potential health hazards of this meat sauce, based on the contained active principles, have been highlighted (Nwaopara *et al.*, 2007a). Its constituent spices - ginger, clove, red pepper and black pepper (Nwaopara *et al.*, 2004), have gingerol (Witchtl, 2004), eugenol (Krishnaswamy and Raghuramulu, 1998), capsaicin (Surh and Lee, 1995), and piperine (McGee, 2004) respectively, as their active principles. Recently, a study revealed that the excessive consumption of ginger, red pepper, and black pepper, particularly in their combined state, as it is the case with *Yaji*, is capable of inducing hepatic necrosis (Nwaopara *et al.*, 2007b).

In this similar study on the kidney however, the report by Soetiarto (1999) that clove and its component eugenol has been associated with allergic reactions including possible severe reactions like anaphylaxis as well as the allergic complaints by Michigan (2004) implicating black pepper, is of utmost importance. The report also, that the side effects associated with large doses of cloves ingested orally, in its diluted oil form, or used in clove cigarettes, may give rise to vomiting, sore throat, seizure, sedation, difficulty in breathing, fluid in lungs, vomiting of blood, blood disorders, and kidney failure by Consolini and Sarubbio (2002), is as well significant. Of importance also, is the fact that anaphylactic, hypersensitivity and inflammatory reactions involve mast

cells -the tissue equivalents of basophils, and are known to interact with eosinophils and macrophages in allergic reactions (Cheesbrough, 2005).

Moreover, scattered reports of mast cells in the renal interstitium dates back to 1960 (Pavone-Macaluso, 1960), and there have been a few recent studies of mast cells in the interstitium of human kidneys with primary glomerular diseases (Ehara and Shigematsu, 1988; Hiromura *et al.*, 1998; Tóth *et al.*, 1999; Roberts and Brenchley, 2000). Mast cells have also been observed in association with renal amyloidosis, diabetic nephropathy, chronic allograft rejection, and tubulointerstitial nephropathies (Eddy, 2001a). In addition, several studies on extrarenal lesions, such as the cutaneous lesions of scleroderma and keloid scars, pulmonary fibrosis, cardiac fibrosis, hepatic cirrhosis, rheumatoid arthritis, and myelofibrosis, have reported a close correlation between mast cells and fibrosis (Eddy, 2001a).

This study is therefore designed to correlate the individual and combined effects of the excessive consumption of *yaji*-spices on the histology of the kidney, since each of these spices, in large doses, are potentially harmful (Southgate, 1993).

Materials and Methods

Experimental animals: Twelve (12) adult rabbits bought from the animal house of the College of Medicine, Ambrose Alli University, Ekpoma, Edo, Nigeria, and moved to the experimental laboratory of Anthonio

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Research Center, Ekpoma, Edo, Nigeria, where used for this study after being allowed to acclimatize for 3 weeks. Feed was from Bendel Feeds and Flour Mills (BFFM), Ewu, Edo, Nigeria, with a daily supplementation of lemon grass procured from the immediate environment. Water was given *ad libitum*. The experimental animals were divided into six groups of two (2) rabbits each (A – F). Those in groups B, C, D, E and F, constituted the test groups whereas group A served as the control.

The Spices: For this experiment, the spices ginger, clove, red pepper and black pepper were used. They were procured from Aduwawa Market Benin City, Edo, Nigeria, where the local meat sauce –*Yaji*, containing these spices and other ingredients are produced. A hand-grinding machine was used to grind them into powdered form and then measured as appropriate.

Experimental procedure: The rabbits were weighed just before the administration of the spices and just before they were sacrificed. The administration of the spices was performed via mixing with feed as follows:

1. Group A (Control) received only normal feed with lemon grass supplementation daily for 21 days.
2. Group B received a mixture 3g of Clove, 3g of ginger, 3g of Red pepper and 3g of Black pepper in feed with lemon grass supplementation daily for 21 days.
3. Group C received a mixture 3g of Ginger in feed with lemon grass supplementation daily for 21 days.
4. Group D received a mixture 3g of Clove, in feed with lemon grass supplementation daily for 21 days.
5. Group E received a mixture 3g of Red pepper in feed with lemon grass supplementation daily for 21 days.
6. Group F received a mixture 3g of Black pepper in feed with lemon grass supplementation daily for 21 days.

At the end of the 21st day, the animals were sacrificed and the organ kidney, was obtained and immediately fixed in formaldehyde solution.

Tissue processing: Tissue sections of the organs were produced via normal histochemical methods of fixation, dehydration, impregnation, embedding, sectioning and staining (with haematoxylin and eosin).

Results

Weight gain was observed in groups A (control), C (ginger), D (clove), and E (red pepper) while there was weight loss in group B. The weight for the animals in group F (black pepper) remained the same. Using a light microscope, the histological observations on the kidneys obtained from groups A – F, are represented by micrographs labeled 'Plates A – F', as given below:

Plate A: (Control; Kidney x16): Showing normal pictures
Plate B: (Kidney x40): Showing several distinct round

basophilic bodies in the interstitium of the renal cortex.

Plate C: (Kidney x16): Showing normal pictures

Plate D: (Kidney x40): Showing several distinct round basophilic bodies in the interstitium of the renal cortex.

Plate E: (Kidney x16): showing normal pictures.

Plate F: (Kidney x16): showing normal pictures with few round basophilic bodies.

Discussion

The result of this study showed that the excessive consumption of ginger and red pepper had no effect on the histology of the kidney as observed on plates C, and E. However, there was the presence of several distinct round basophilic bodies in the interstitium of the renal cortex in plates B and D, but few of such basophilic bodies in the interstitium of the renal cortex in plate F. These histological observations require explanation.

Interestingly, the 'no-effect' result from test group C –fed with ginger (active principle –gingerol), adds 'flesh' to the findings by Kuhad *et al.* (2006) on the 'renoprotective' potential of gingerol treatment that was evident in their study involving a single dose of cisplatin on rats which resulted in marked renal oxidative and nitrosative stress and significantly deranged renal functions; whereas, gingerol treatment significantly and dose-dependently restored renal functions, reduced lipid peroxidation and enhanced the levels of reduced glutathione and activities of superoxide dismutase and catalase. Similarly, the 'no effect' result from test group E -fed with feed and red pepper (active principle-capsaicin), agrees with the findings by Jang *et al.* (1992) from a 4-week feeding study with groups of 5 male B6C3F1 mice with 0, 0.5, 1.0, 2.5, 5.0, 7.5, and 10% ground red chilli (*Capsicum annum*) in the diet, that apart from the liver, there were no lesions in the order organs (including the Kidney). Recall that group B was fed with feed and a combination of all the spices (ginger, clove, red pepper and black pepper) while group D and F was fed with feed plus clove and black pepper only, respectively. This suggest therefore, that clove and its active principle –eugenol, as well as black pepper and its active principle –piperine, are implicated with regards to the presence of distinct round basophilic bodies in the interstitium of the renal cortex, since plates A (Control) as well as plates C, and E from the groups fed with feed plus ginger and red pepper respectively, presented normal pictures.

Based on existing scientific facts, the presence of basophilic inclusion bodies in the kidney may be associated with microorganisms as reported in birds by Bernier *et al.* (1981) and in humans by Nickleit *et al.* (1999; 2000) and Drachenberg *et al.* (1999). However, there is an obvious probability that the distinct round basophilic bodies in the interstitium of the renal cortex in B, D and F, might not be micro - organism - related

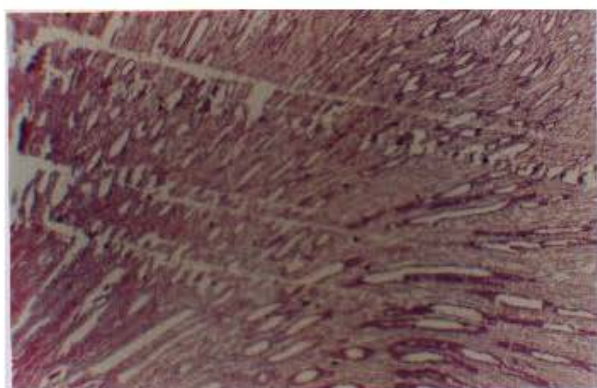


Plate A

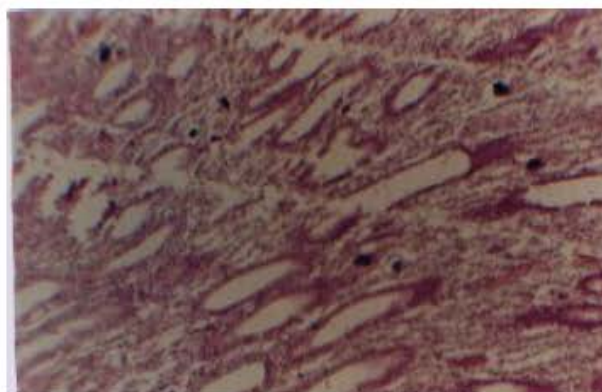


Plate D

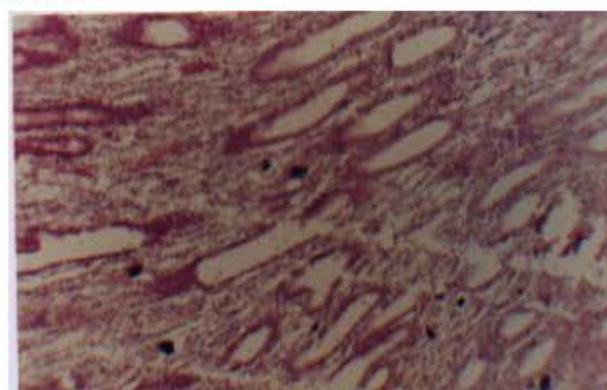


Plate B

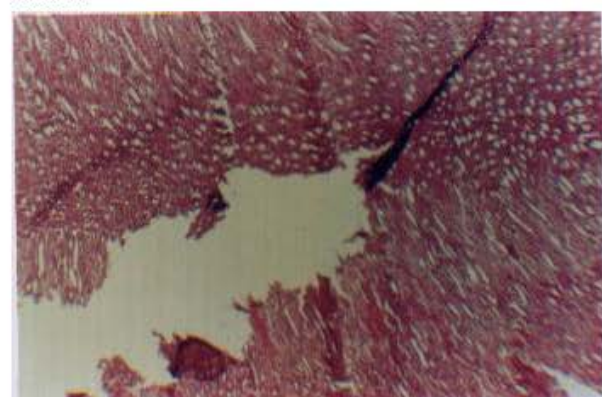


Plate E

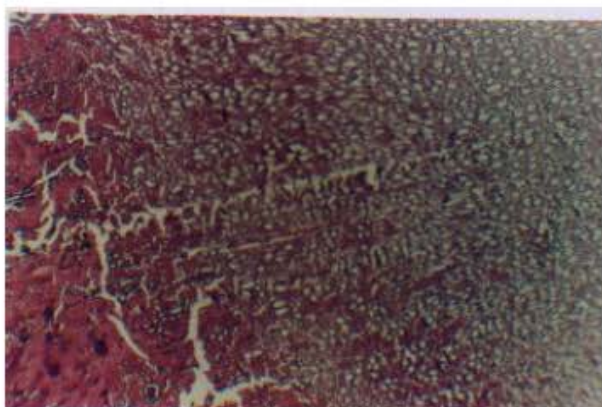


Plate C

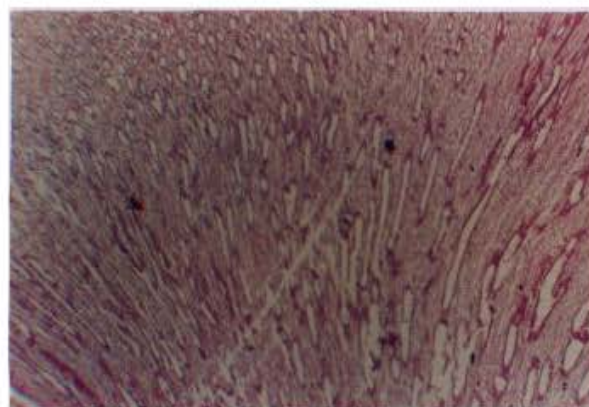


Plate F

considering the overwhelming evidence that are available to substantiate the antimicrobial potentials of the spices under study as reported by Grieve, 1971; Saber, 1982; Gughani and Ezenwanze, 1985; Nanir and Kadu, 1987; Meena, 1992; Kapoor, 1997; Kurokawa *et al.*, 1998; James *et al.*, 1999; Amaechi *et al.*, 2000; Ghelardini *et al.*, 2001; Kalemba and Kunicke, 2003.

Another line of thought considers the complains by Michigan (2004) about allergic reactions to the consumption of foods containing black pepper as well as the reports by Yunginger (1992) that though the

incidence of fatal food-induced anaphylaxis is unknown and rarely reported, it has been associated with elevated mast cell tryptase levels; by Soetiarto (1999) that cloves have been implicated in allergic reactions and anaphylaxis; and by Consolini and Sarubio (2002) that large doses of cloves ingested orally, in its diluted oil form, or used in clove cigarettes, may give rise kidney failure.

Indeed, the importance of these reports in the present study is that as mast cells have been known to be involved in anaphylactic, hypersensitivity and

inflammatory reactions; black pepper implicated in allergic reactions; and cloves proven to be involved in allergic and anaphylactic reactions as well as in kidney failure, there is therefore a common relation in this regard, between clove, black pepper and mast cells. It suggests also that the presence of the distinct round basophilic bodies in the interstitium of the renal cortex as seen in plates B and D might be cloves and black pepper feeding-induced. In addition, there is the possibility that a more marked histological alteration would have been observed if the spice-feeding period had lasted much longer. Of course, this might sound speculative but we hinge this speculation on the statement by Eddy (2001b) that "histologically, progressive renal disease is characterized by an interstitial infiltrate of inflammatory cells in association with tubular atrophy and interstitial fibrosis".

Presently, there are many reasons to believe that mononuclear cells, especially those of monocytic lineage, play an active role in fibrogenesis even though definitive proof of their role is still lacking (Eddy, 2001a). This is of great concern, considering the strong correlation between interstitial mast cell number and interstitial fibrosis that has been reported by El-Koraie *et al.* (2001). According to Eddy (2001b), mast cells have been largely ignored by the nephrology community and that basophilic leukocytes are characterized by the presence of cytoplasmic granules enriched with proteoglycans and proteases, including the mast cell-specific proteases, chymase and tryptase. Eddy (2001b) also pointed out that mast cell precursor cells originate in the bone marrow, circulate in small numbers in the blood, and mature into tissue mast cells within most organs.

Considering the common involvement of cloves, black pepper and mast cells in allergic and anaphylactic reactions, the presence of the distinct round basophilic bodies in the interstitium of the renal cortex of plates B, D and F, are suspected to be mast cells and this might have been clove and Black pepper feeding induced. The result of the present study therefore suggests that the excessive consumption of cloves or black pepper alone, or in combination with other spices, are capable of inducing a duration dependent but progressive renal damage, which may be through a mechanism that is associated with mast cell mobilization into the kidney, and subsequently the triggering of renal fibrosis.

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