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HISTOLOGICAL STUDIES OF THE EFFECTS OF RED PEPPER ON THE STOMACH OF ADULT WISTAR RATS.

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ABSTRACT:

Histological effects of red pepper commonly used as spice in food on the stomach of adult wistar rats were carefully investigated. The rats of both sexes (n=24), average weight of 200g were randomly assigned into two treatments (n=16) and control (n=6) groups. The rats in the treatments groups received 1g and 2g of red pepper thoroughly mixed with 20g of their feeds for 7 and 14 days, while the control rats received equal amounts of feeds without the red pepper added. The rats were fed with grower's mash purchased from Edo feeds and flour mill Ltd, Ewu, Edo State and were given water liberally. The rats were sacrificed on day eight and fifteen of the experiment respectively.

The stomach was carefully dissected out and quickly fixed in 10% formol saline for routine histological procedure after H & E method.

The histological findings after H&E methods indicated that the treated sections of the stomach showed some level of cellular hypertrophy, congestion of blood vessels degenerative changes disruption and distortion of the cytoarchitecture of the stomach.

These findings indicate that red pepper may have some deleterious effects on the microanatomy of the stomach of adult wistar rat at higher doses. It is recommended that further studies aimed at corroborating these findings be carried out.

Key words: Red pepper. Histological effects. Stomach and wistar rats.

RESUMEN:

Han sido investigados los efectos histológicos de la pimienta roja, especie comúnmente utilizada como condimento de alimentos, en el estómago de ratas wistar adultas. 24 ratas de ambos sexos de un peso promedio de 200 gramos fueron asignados aleatoriamente a dos grupos: experimental (n = 16) y control (n = 6). Las ratas del grupo experimental recibieron 1g y 2g de la pimienta roja, mezclada con 20 gramos de la dosis de pienso para 7 y 14 días, mientras que el grupo de ratas control recibió iguales cantidades de alimentos, pero sin pimienta roja añadida. El pienso de rata procedía de Edo feeds and flour mill Ltd., Ewu, Edo State, y se les proporcionó a los animales agua sin restricciones. Los animales fueron sacrificados en el día ocho y quince de la prueba, respectivamente.

El estómago fue cuidadosamente disecado y fijado rápidamente en una solución salina de formol al 10% para estudio histológico

y siendo procesado rutinariamente y teñido con Hematoxilina y Eosina.

Los hallazgos histológicos en los cortes de estómago mostraron cierto grado de hipertrofia celular, congestión de los vasos sanguíneos y cambios degenerativos, con alteraciones y distorsión de la citoarquitecture del estómago.

Estos resultados indican que la pimienta roja puede tener algunos efectos perjudiciales sobre la anatomía microscópica del estómago de las ratas Wistar adultas a dosis más altas. Serían recomendables nuevos estudios encaminados a corroborar estos resultados.

Palabras clave: Pimienta roja. Efectos histológicos. Estómago y ratas wistar.

INTRODUCTION

Pathological processes frequently involve the body's normal responses to abnormal environmental influences. Such noxious external influences as pathogenic microorganisms, trauma, dietary deficiencies and hereditary factors acting alone or in a complex interaction with environmental factors, case diseases¹. Various environmental chemicals, industrial pollutants and food additives have been implicated as causing harmful effects². Most food additives act either as preservatives, or enhancer of palatability. Spices, in particular black pepper, red pepper, and chili powder, may produce indigestion, but they do not seem to seriously injure the stomach³. Some investigators have reported that there is no difference in rates of inflammation of the stomach in heavy consumers of spice and no difference in the rate of ulcer heating in those patients consuming large amount of red pepper daily^{4,5}. In experiments on rats, the active ingredient in pepper, capsaicin, was found to protect the stomach mucosa from damage caused by alcohol or aspirin^{6,7}.

Capsaicin is contained in a different spicy vegetable among which is cayenne pepper, which was used in this experiment. Cayenne pepper is a member of the family solanaceae genus: capsicum of vegetables and with botanical name - capsicum frutescence. The common name, cayenne, was actually given to this pepper because of its cultivation in a town that bears the same name in French Quiana on the North-East Coast of South America. The Scoville heat unit of cayenne pepper ranges 30k to $50k^8$. Red pepper is the mostly used spicy for food throughout the world, especially in Central America, Latin America, Africa and Asia⁹. In moderately large quantities, it is one of the most ingredients used in meals in Nigeria, especially the Southern part. Chemically, the red pepper contains approximately 0.14% capsaicin¹⁰⁻¹¹, a crystalline colourless compound which is an active principle that accounts for its pungency.

It has been reported that the gastro protection afforded by prostaglandin E2, cholecystokinin gastrin, the proton pump inhibits lansoprazole, and the antacid, hydrotalcit is reduced or abolished in capsaicin pretreated rats¹². In 1999, Holzer reported that capsaicin lessen ablation effect produced by aspirin, and that neurotoxin dosage of capsaicin defunctionalizes nociceptive afferents, including those innervating the stomach, this producing a chemical knockout of neurons that participate in the maintenance of the gastric mucosal integrity¹³. In the stomach of the rats capsaicin has been reported to cause no change on the basal mucosal blood flow¹⁴. While it was also stated by another investigator that in the stomach of the rat capsaicin do not cause any change in the basal acid and pepsin¹⁵.

The stomach is the most dilated portion of the alimentary canal, located in the epigastriun and the left hypochondruim regions of the abdomen. It is lined by simple columnar epithelial cell and functions in the degradation and digestion of food materials in the body. The stomach also functions in the preventions of gastric ulceration due to the presence of the numerous mucous secreting glands. Since red pepper contains capsaicin which was found to protects the stomach mucosa from damage caused by alcohol or aspirin, it is therefore relevant to investigate some of its histological effects on the stomach. It is probable that the adverse effects of red pepper may affect the normal histological structure of the stomach, and hence this investigation.

MATERIALS AND METHODS

Animals: Twenty four adult wistar rats of both sexes with average weight of 200g were randomly assigned into three groups, (two treatments n=16 and control n=6). The rats were obtained and maintained in the animal holdings of the department of anatomy, school of basic medical sciences, University of Benin, Edo State, Nigeria. They were fed with grower's marsh obtained from Edo feed and flour mill limited, Ewu, Edo State and given water liberally. The control group consisted of six rats while the two treatments groups consisted of six rats each red pepper was obtained from the Edaiken market, Uselu, Benin City.

Red pepper administration: Red pepper was obtained from the Edaiken market, Uselu, Benin City, dried and grinded in the department of anatomy, school of basic medical sciences, University of Benin Edo State. The rats in the treatments groups were divided into two main groups (A, B), with each group receiving 1.0g and 2.0g of red Pepper mixed with 20g of grower's march respectively. The red pepper was thoroughly mixed with the feeds in a container fixed to the floor of the experimental cage to avoid spillage of the feeds during their feeding. The quantity of feeds consumed was known by weighing before and after each day of the experiment. The control rats received equal amount of feeds without red pepper added for the same period of the experiment: the rats were sacrificed by cervical dislocation method on the eight and fifteenth days of the experiments respectively. The stomach was quickly dissected out and fixed in 10% formal saline for routine histological techniques.

Histological study: The tissue were dehydrated in an ascending grade of alcohol (ethanol), cleared in xylene and embedded in paraffin wax, serial sections of 7 microns thick were obtained using a rotator microtome. The deparaffused sections were stained routinely with haematoxylin and eosin. Photomicrographs of the desired section were obtained using digital research photographic microscope in the department of anatomy, school, of basic medical sciences, university of Benin for further

observations.

RESULTS

The stomach of the control section showed normal histological features with the mucosa lined with simple columnar epithelial cells, lamina propria showing some highly packed glandular secretory cells, and some blood vessels in the section of the control stomach.

The stomach of the treated sections showed some level of cellular hypertrophy associated with congestion of some blood vessels. There is also some degree of degenerative changes distortion and necrotic debris associate with the treated sections which is more marked in the treatment sections receiving 2g of red pepper

DISCUSSION

The results of the haematoxylin and eosin staining (H&E) reactions showed increased cellular hypertrophy and degenerative changes in the stomach of the treatment groups. The increased in cellular hypertrophy of the treatments group as reported in this study may have been as a result of cellular proliferation caused by the intake of food mixed with the red pepper. It may then be inferred from the present results that higher dose and prolonged intake of red pepper resulted in degenerative changes observed in the glandular epithelium of the stomach. The actual mechanism by which red pepper induced cellular degeneration observed in this experiment needs further investigation.

Degenerative changes have been reported to result in cell death, which is of two types, namely apoptotic and necrotic cell death. These two types differ morphologically and biochemically16. Pathological or accidental cell death is regarded as necrotic and could result from extrinsic insults to the cell such as osmotic, thermal toxic and traumatic effects¹⁷. In this experiment, red pepper could have acted as toxins to the cell of the stomach. The process of cellular necrosis involves disruption and distortion of membrane's structural and functional integrity which was also a landmark of this experiment.

In cellular necrosis, the rate of progression depends on the severity of the environmental insults. The greater the severity of insults, the more rapid the progression of neuronal injury¹⁸. The principle holds true for toxicological insults to the brain and other organs¹⁹. It may be inferred from the present results that prolonged intake of red pepper resulted in increase toxic effects on the stomach with that of higher dose more marked. This work is in consonance with the research work carried out by Eweka et al, (2007) that monosodium glutamate used as food additive causes cellular hypertrophy, vacuotations and distortions in the epithelia of the stomach²⁰.

The observed congestion of blood vessel in the treated groups suggested the occurrence of inflammatory, reaction which was as a result of tissue damage. This could be explained by the fact that red pepper inflicted chemical injury on the gastric mucosa which was followed by arterial relaxation around the stomach. The capillary network around the stomach then becomes engorged with rapidly flowing blood accompanied by lymphocytic infiltration. This work is in line with some studies carried out on the effect of red pepper on rat gastric mucosa resulting to some abnormality ranging from oedema submucosal haemorrhages, congestion of blood vessels and exfoliation of gastric surface epithelial cells²¹⁻²².

CONCLUSION AND RECOMMENDATION

In conclusion, our study revealed that chronic administration of red pepper to adult wistar rats caused cellular hypertrophy, congestion of blood vessels, degenerative changes, disruption and distortion of the cytoarchitecture of the stomach. This resulted in some necrotic debris associated with the treated sections.

With these results, it is probable that the functions of the stomach may be adversely affected. It is recommended that further studies aimed at corroborating these findings be carried out.

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In the present study a stomach histological investigation was carried out on the effects of feeding rats with a diet containing red pepper.

Red pepper is one of the spices that has been used all over the world from old times. It varies physiological activities, especially the suppression of blood lipid levels and the antibacterial activity, and it has an inhibitory effect on cecal bifidobacteria, etc. Furthermore, its active material, capsaicin, is irritating and inflammatory and may cause gastric ulcers and mucosal lesions. The present study reporting histological data on the effect of red pepper on the stomach can be regarded as an interesting contribution to previous studies

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Having considered the current concerns regarding the effects of diet on developing stomach pathology, the evaluation of the

dietary risk factors for stomach disorders becomes essential. The mechanism by which dietary pepper affects various gastric features is poorly understood, as is the ability of certain compounds to relieve possible symptoms. Therefore, it is worth investigating the ability of black pepper, red pepper and other relevant chilli species to determine gastric surface hydrophobicity and to induce or relieve visceral pain in rat model systems.

The study presented herein examines the effects on stomach of adult Wistar rats of red pepper consumption, one of the most popular ingredients in authors' native Nigeria, as well as elsewhere. The experimental design and results are clearly presented. The histological findings show certain level of cellular hypertrophy, congestion of blood vessels, degenerative changes and distortion of the cytoarchitecture of the stomach, following the administration of red pepper. The presented work is of interest also for comparative purposes with the results of other research groups and thus deserves to be published.

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