



SPLEEN INTERSTITIAL AND VASCULAR CONGESTIONS BY INGESTION OF *ALLIUM SATIVA* (GARLIC) IN ADULT WISTAR RATS

*¹Idehen I. Charles, Bankole J. Kayode¹, Airhomwanbor Kingsley¹, Dic-Ijiewere Ebenezer², Okparaku Sunday¹, Ehimara Raphael², Osarobo Eseiwi³ and Aziba Confidence¹

¹Department of Medical Laboratory Science, Faculty of Basic Medical Sciences, College of Medicine, Ambrose Alli University, Ekpoma-Nigeria.

²Department of Chemical Pathology, Faculty of Clinical Sciences, College of Medicine, Ambrose Alli University, Ekpoma-Nigeria.

³Department of Hematology, Faculty of Clinical Sciences, College of Medicine, Ambrose Alli University, Ekpoma-Nigeria.

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*Corresponding Author

Idehen I. Charles

Department of Medical
Laboratory Science, Faculty
of Basic Medical Sciences,
College of Medicine,
Ambrose Alli University,
Ekpoma-Nigeria.

ABSTRACT

This study was conducted to assess the histological effect of *Allium sativa* (Garlic) on the spleen using adult male Wistar rats. The experiment consisted of 30 adult male Wistar rats of (200 to 250g) divided into six groups; a control (group A) and 5 test groups (B to F) respectively treated on varying doses of garlic 200mg/kg, 400mg/kg, 600mg/kg, 800mg/kg and 1000mg/kg for 4 weeks. At the end of treatments, the spleens were harvested from rats for histological processing following standard histological procedures. The histological findings showed that the spleen of the control and test group showed no marked pathological or histological alteration except for the 1000mg/kg treated group where mild interstitial and vascular congestions were observed. These results suggest that garlic may not

cause any alteration in the histology of the spleen at low and moderate doses but care must be taken as the dose approaches 1000mg/kg. These findings further buttress the fact that dosage determines poison.

KEYWORDS: *Allium sativa*, Garlic, Spleen, Toxicity, Histology.

INTRODUCTION

Although most plants concoctions are not officially regulated like conventional drugs, but report has shown its use and abuse is highly prevalent.^[1] Medicinal plants have been used as sources of medicine in virtually all cultures.^[2,3] and varieties of plants are known to be consumed both for dietary and therapeutic purposes worldwide. In fact, it has long been documented that before mankind discovered the existence of microbes, the idea that certain plants had healing potentials against infections; as what we would currently characterize as antimicrobial was well accepted.^[1] One of such plants commonly consumed in Nigeria and elsewhere is *Allium sativum* commonly known as Garlic.

Garlic is one of the most frequently consumed plants worldwide, and its moderate intake has been considered to be beneficial for human health.^[4] Garlic was shown to exert a wide range of pharmacological such as antiviral, antibacterial^[5], antitumor^[6], antiatherosclerotic^[7], antioxidant^[8] and hypolipidemic activity.^[9] However, several reported effects were deviating and conflicting and depended on experimental duration, garlic dosage and mode of administration.^[10]

The spleen is known as an organ of both phagocytic and immune function. The spleen has three main functions^[11]: first, it represents a large mass of organized lymphatic tissue passed by recirculating lymphocytes, which are able to promptly elicit specific T or B lymphocyte-mediated immune reactions against antigens carried by the blood. Second, the splenic red pulp has a filtering function for the blood; a function that removes materials by red pulp macrophages, including aged or abnormal red blood cells or microorganisms and leucocytes covered with immune complexes. Third, in some mammalian species, but not in humans, the spleen serves as a reservoir of erythrocytes, which are transfused into the circulation on sympathetic stimulation. In humans, only thrombocytes are normally pooled in the spleen. The spleen is comprised of two histologically and functionally distinct tissues; one is a phagocytic tissue, the red pulp, and the other is an immune tissue, the white pulp.^[12] Considering the several reported beneficial effects of garlic couple with the conflict in relation to dose, duration and mode of administration, this study therefore evaluated the histological effect of varying doses of garlic on the spleen using adult Wistar rats as a model.

MATERIALS AND METHODS

Substance of study: Adequate amount of Garlic was bought from the commercial market in Ekpoma, Edo State- Nigeria. They were stored in a cool dry place pending usage.

Experimental animals and grouping: Thirty (30) Adult Albino Wistar rats of comparable sizes and weights (200 – 250g) were procured from animal house and transferred to the experimental site where they were allowed two (2) weeks of acclimatization. The animals were weighed on the first day of the acclimatization period and fed with rat chow and water giving *ad libitum*. They were housed in well ventilated labeled cages at the site of the experiment. The cages were designed to secure the animals properly especially from wild animals/insects and cleaned daily. The experimental animals were separated into six groups (A – F). Group A shall have five rats (n = 5) while groups B- F have five rats (n = 5) each using 6 big cages to house them. Group A served as the control and received only the normal feed (grower's mash) and water with no administration of Garlic while Group B, C, D, E and F received different doses of Garlic.

Substance administration: The rats were weighed before the administration of the garlic and just before they were sacrificed and similar weight measurements were done at the end of each week and recorded accordingly. The administration of the garlic was performed via mixing with feed and given orally as follows:

- **Group A** (Control) received only normal feed (growers' mash) and water daily for 28days.
- **Group B** received 200mg /kg bwt of garlic, feed and water daily for 28days.
- **Group C** received 400mg /kg bwt of garlic, feed and water daily for 28days.
- **Group D** received 600mg /kg bwt of garlic, feed and water daily for 28days.
- **Group E** received 800mg /kg bwt of garlic, feed and water daily for 28days.
- **Group F** received 1000mg /kg bwt of garlic, feed and water daily for 28days.

Sample collection and analysis: Body weight was measured before and after acclimatization, similar weight measurements were done at the end of the treatment periods and the average weight recorded accordingly. The spleen of each rat was obtained at the end of treatment under chloroform anaesthesia and fixed in 10% formalin for histological processing.

Processing schedule: The tissues were processed according to standard histological procedures as previously documented by Idehen et al.^[13]

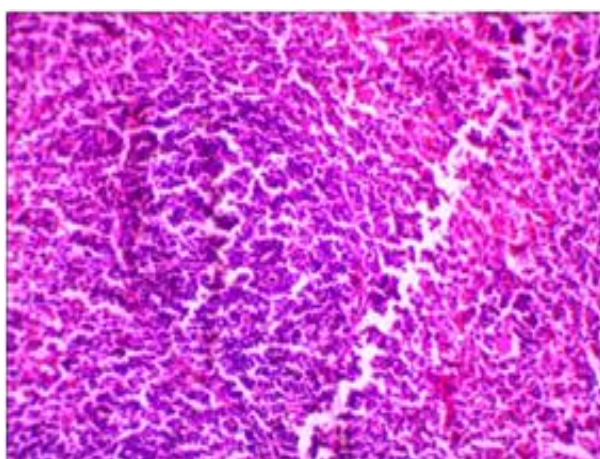
Data presentation: The obtained gross findings were presented in table while microscopic observations were presented in micrograph pictures.

RESULTS

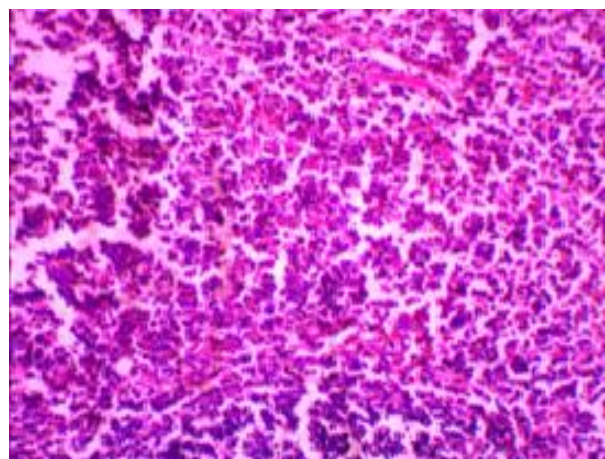
Table 1: The gross and histological observations on the effect of *Allium sativum* (garlic) on the spleen of adult Wistar rats.

Group	Dosage of garlic powder (mg/kg)	Gross findings	MICROSCOPIC EXAMINATION			
			Cell degeneration	Necrosis	Loss of tissue architecture	Interstitial/vascular congestion
A	0	Normal	-	-	-	-
B	200	Normal	-	-	-	-
C	400	Normal	-	-	-	-
D	600	Normal	-	-	-	-
E	800	Normal	-	-	-	-
F	1000	Normal	-	-	-	+

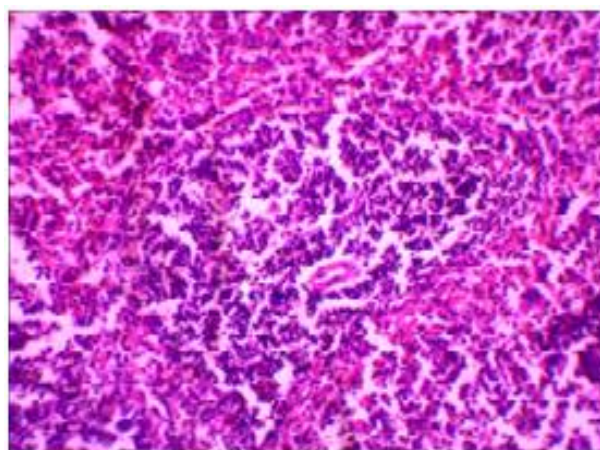
- = Absent, + = present



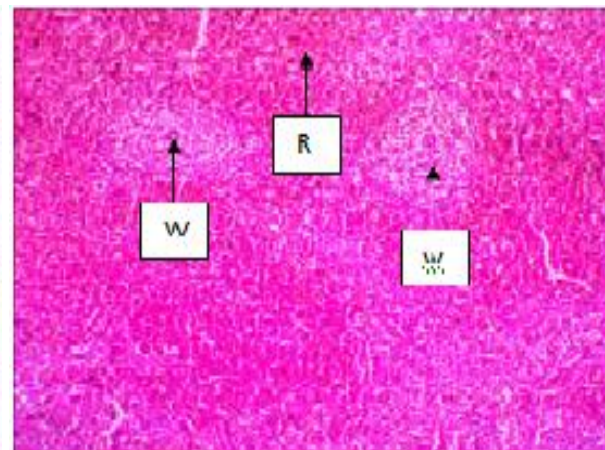
Group A. Splenic tissue section composed mainly of normal peri-arterial lymphoid sheath with germinal centre



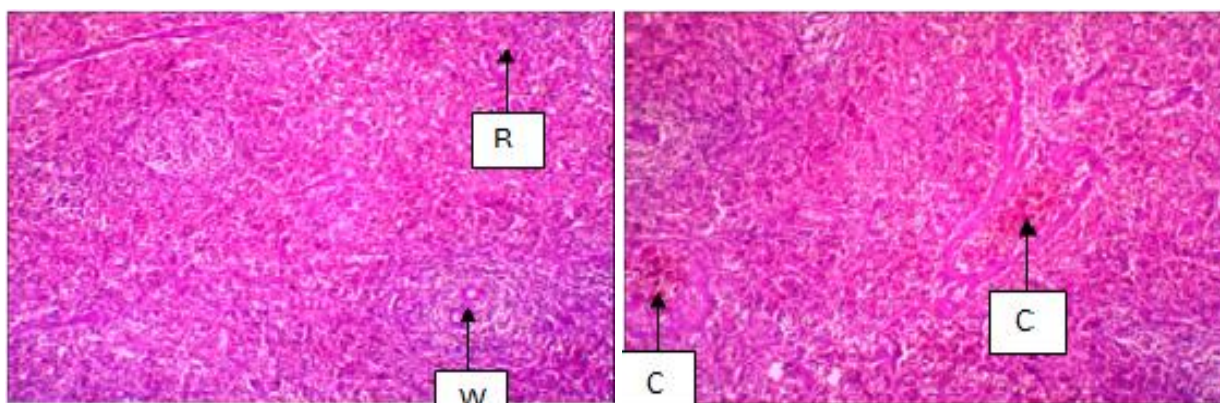
Group B (200mg/kg treated) Splenic tissue section composed mainly of normal peri-arterial lymphoid sheath with germinal centre



Group C (400mg/kg treated) Splenic tissue section composed mainly of normal peri-arterial lymphoid sheath with germinal centre



Group D (600mg/kg treated) Splenic tissue section composed mainly of normal peri-arterial lymphoid sheath (white pulp W) with germinal centre and red pulp (R)



Group E (800mg/kg treated) Splenic tissue section composed mainly of normal peri-arterial lymphoid sheath (white pulp W) with germinal centre and red pulp (R)

Group F (1000mg/kg treated) Splenic tissue section showing mild interstitial and vascular congestion C

Figure 1: Spleen histological sections of adult Wistar rats treated on varying doses of Garlic (H&E x 100).

DISCUSSION

Garlic has been used as spices in foods and for medicinal purposes as it has antibiotic, antiviral and antifungal properties.^[14] There has been documentation that some of the common plants consumed by man and animals as food and medicine possess deleterious systemic effects.^[15] In the present study, the effect of garlic extract on the spleen histological architecture was investigated and findings showed that doses between 200mg/kg and 800mg/kg have not toxic effect on the spleen. However, at 1000mg/kg dose of garlic ingestion for 28 days, there were mild interstitial and vascular congestion in the spleen. These findings suggest that low doses of garlic may has no deleterious effects on the spleen but higher doses (>1000 mg/kg body weight/day) can cause severe threat to the spleen. In agreement with the findings of this study, significant alteration to the cells of the spleen (white pulp, red pulp, lymphoid tissue, trabeculae) and even the outer capsule have been reported in rats treated with 300mg/kg aqueous garlic extract for 21 days.^[16] Although several studies have reported the pharmacological effects of garlic in the immune system to include suppression of inflammatory cytokines production, immunomodulatory and proliferation of human lymphocytes^[17,18], others have also documented the effect of garlic on human system, opined that garlic possessed toxic substances.^[19,20]

In comparison with the findings of this study, Fowotade et al.^[21] has reported 38 days treatment with 400mg/kg and 500 mg/kg of *Allium sativum* extract to cause varied histological changes in different organs. In the study by Fowotade et al.^[21], 38 days treatment

with 400mg/kg and 500 mg/kg of *Allium sativum* extract cause widening of the cardiac interfibre spaces, nuclei irregularity of cardiac muscles, widening of the Bowman's space, loss of flattened squamous tubular epithelial cells lining the Bowman's space of the kidney, and in the liver dilated sinusoids and cellular hypertrophy with mild congestion of central vein. Also, gastrointestinal tract injury in healthy subjects after intragastric infusion of a single dose of raw garlic of over 0.75 g has been reported.^[22]

The observed spleen histological changes as induced by high dose of garlic extract in this study may cause the inability of the spleen to play its physiological function. However, in a prior study by Hamlaoui-Gasmi *et al.*^[23], it was demonstrated that high dosage garlic exhibited dual effects in rat that is antioxidant or prooxidant depending on the mode of administration. In the spleen, oral garlic was reported to exerted antioxidant effect whereas intra-peritoneal garlic was pro-oxidant as assessed by MDA and H₂O₂ levels.^[23] It was concluded by the study of Hamlaoui-Gasmi *et al.*^[23] that the prooxidative or antioxidant effect of high dosage garlic is linked to route of administration and to the extent it modulates (excess or deficiency) labile iron pool, the threshold of which is organ specific.

The findings of this study highlight the possible potential ability of garlic extract to induce morphological changes in the spleen at high doses of ingestion and in indiscriminate medicinal uses. The observed spleen interstitial and vascular congestions by high dose of garlic can hamper normal function and lead to several adverse effects considering the physiological role of the spleen. Based on the histological observations presented in this study, daily garlic consumption should be limited to a safe margin far below 800mg/kg to prevent the likely spleenotoxic effect which can be dose dependent. The findings of this study further buttress the fact that dosage determines poison.

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