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Hematological Effects Following Ingestion of *Allium cepa* (Onion), *Allium sativum* Garlic and Treatment with Iodine in Albino Wistar Rats

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Abstract: This study was aimed at assessing the extent to which ingestion of *Allium cepa* (onion) *Allium sativum* (garlic) and treatment with iodine affect haematological parameters of albino Wistar rats. The effect on total hemoglobin, hematocrit (PCV%), red blood cells (RBC) and mean cell hemoglobin concentration (MCHC) were carried out. Oral administration of onion and garlic extract led to a dose dependent decrease in the total haemoglobin level of normal rats which was not statistically significant ($p<0.05$). Treatment with iodine led to a statistically significant ($p<0.05$) decrease in the total hemoglobin of the positive control group (treated with iodine only). Oral administration of onion and garlic extract to the normal rats led to a decrease in the hematocrit which was not statistically significant ($p<0.05$). Nonetheless, treatment with iodine led to a statistically significant ($p<0.05$) drop in the hematocrit of the iodine treated rats administered with high dose onion when compared to the control. In the normal rats, oral administration of high dose garlic and onion extracts led to a statistically significant ($p<0.05$) increase in the RBC when compared with the control group however the iodine treated groups recorded a drop which was statistically significant ($p<0.05$) in the low dose onion and garlic groups. From these results, it becomes imperative not just to consider the effects of these *Alliums* on the haematological parameters but to consider fortification with nutrients while embarking on the various forms of fortification with iodine. This is of particular importance mainly in areas where nutritional anemia is a concern.

Key words: Iodine, onion, garlic, hematological parameters

INTRODUCTION

Iodine is essential for the production of thyroid hormones, which, in turn, are important for normal growth and development. Iodine deficiency affects nearly 2 billion people worldwide and is easily avertable with the use of iodized salt in the diet (Zimmermann, 2009). The epidemiology of iodine deficiency has improved after the implementation of universal salt iodization programs in nations where iodine deficiency was identified as a public health problem and severe iodine deficiency is now uncommon (WHO, 2007).

Onion (*Allium cepa)* and garlic (*Allium sativum*) are probably the most well-known members of the Liliaceae family. Apart from their culinary uses, therapeutic properties have been ascribed to these *Alliums* since ancient times (Brown, 2002; Arnault and Auger, 2006), provoking earlier and detailed chemical inquiry of the most characteristic active ingredients (Jones *et al*., 2004; Arnault and Auger, 2006, Lanzotti, 2006; Santas *et al*., 2008). While the health benefits of these species have been strongly demonstrated (El-Demerdash *et al*., 2005; Sankaran *et al*., 2010), *Allium* species have been associated in the stimulation of hemolytic anemia in animals such as cattle, water buffalos, sheep, horses, dogs and cats (Banerjee and Maulik, 2002).

Haematological parameters are common diagnostic tools used in evaluating the current health status of individuals (McPherson and Pincus, 2009). Haematological parameters are closely related to the response of animals to the environment. They can provide substantial diagnostic information once reference values are established under standardized conditions (Gabriel *et al*., 2004). Increased mortality and morbidity have been directly linked with abnormal hematological findings such as low hemoglobin (Jha, 2006; Pennix *et al*., 2006; Zakai *et al*., 2005), low hematocrit (Kosiborod *et al*., 2005) and elevated WBC count (Ruggiero *et al*., 2007; Weiss *et al*., 1995). Hemoglobin, hematocrit and RBC count are used to determine anemia, while RBC indices such as MCHC help in assessing and identifying the morphologic types of anemia especially in humans (Dungca *et al*., 2012).

Studies have been carried out on the effect of administration of various forms and concentrations of *Alliums* on haematological parameters (Tende *et al*., 2012; Ugwu and Omale, 2011; Samson *et al*., 2012) Unfortunately, there is a dearth of information on the effect of these *Alliums* on the haematological parameters of iodine treated animals and also the effect of iodine fortification on the haematological parameters. This study accordingly, seeks to examine, not just the effect of oral administration of onion and garlic on the haematological parameters of both normal and iodine treated rats but also the effect of the oral administration of iodine in form of potassium iodide on albino Wistar rats.

MATERIALS AND METHODS

The onion and garlic sample used for this study were purchased from Ika Ika Qua Market in Calabar, capital city of Cross River State of Nigeria. These vegetables are usually grown in the different parts of the Northern Nigeria and transported to the south. The dry scaly parts of the onion bulbs were removed and the fresh bulbs were properly washed. Three hundred gram of the onions were weighed and macerated in 300 mL of deionized water using an electric blender. The homogenous mixture obtained after maceration was filtered through a cheese cloth and the residue removed, dried and weighed. The solution left behind weighing 455.7 g was used as whole onion extract and from this stock; high and low doses were obtained for the experiments.

The dry scaly outer part of the garlic cloves was removed and the fresh cloves were properly washed. Three hundred gram of the garlic were weighed and macerated in 300 mL of deionized water using an electric blender. The solution left behind was used as whole garlic extract of 455.7 g was used as whole garlic extract and from this; high and low doses were obtained for the experiment.

A quantity of 400 mg of potassium iodide was dissolved in 400 mL of water. 0.8 mL of the solution corresponding 0.8 mg/kg body wt was administered to the animals.

Experimental animals: A total of 100 albino rats of the Wistar strain consisting of both male and females were obtained from the disease free stock of the departmental animal house of biochemistry department, faculty of Basic Medical sciences, University of Calabar, Nigeria. These animals weighing between 80g-120 g were used for the experiment.

The animals were housed in Perspex cage,(North Kent Plastic Cages Ltd, England) with bottom grid and a stainless steel top. The animals were kept under adequate ventilation at temperature and relative humidity of 26±20°C and 46%, respectively. Feed and water were provided *ad libitum*. There weights were taken 3 times during the course of the 14 days.

Animal grouping and treatment: The animals were grouped into ten study groups of ten animals each based on their average weight. The groups were treated as stated below:

Group 1: Control-placebo-deionized water
Group 2: Positive control-potassium iodide solution only
Group 3: Low dose of garlic only
Group 4: High dose of garlic only
Group 5: Low dose of onion only
Group 6: High dose of onion only
Group 7: Low dose of garlic+potassium iodide solution
Group 8: High dose of garlic+potassium iodide solution
Group 9: Low dose of onion+potassium iodide solution
Group 10: High dose of onion+potassium iodide solution

Administration of the extracts: The administration of the aqueous extracts to the different groups of animals was done for 14 days. One milliliter of both the onion and the garlic extracts containing 1.14 g of each sample of onion and garlic extract was designated as low dose while 1.5ml containing 1.70 g of each sample was designated as high dose.

About 0.8 mg/kg body wt of potassium iodide was administered orally to the animals taking potassium iodide at least 4hours before the respective extracts were administered to ensure iodine had been absorbed into the plasma (this is to ensure an iodine loaded state). This is same as incorporating it into the diet fed to the animals, but on account of likely nutrient-chemical interaction that may affect bioavailability, oral solution of potassium iodide was preferred.

Sacrifice of animals: Twenty-four hours after the last administration, the animals were removed, placed in a desiccator glass jar, anaesthetized in chloroform vapour and dissected. Whole blood obtained by cardiac puncture from each animal was collected into a sterile tube. The tube was treated with anticoagulant (EDTA 0.77M) to have plasma with its suspended blood cells intact for haematological studies.

Estimation of haematological parameters: Blood haemoglobin determination was carried out by the cyanmethaemoglobin method as employed by Randox Haemoglobin (Total) Kit. The haematocrit pack cell volume was determined using the method of Dacie and Lewis (1976). The mean cell hemoglobin concentration was calculated using the method of Dacie and Lewis (1976) by dividing hemoglobin concentration by the PCV. Estimation of total red blood cell and white blood cell counts by visual means (Dacie and Lewis, 1976).

Statistical analysis: Results of all the studies were expressed as mean±standard deviation. Data between groups were analyzed using SPSS 2003 (version 13).

RESULTS

The effect of the oral administration of onion and garlic extract and treatment with iodine on the haematological parameters in albino Wistar rats are presented on the Table 1.

Table 1: Effect of the oral administration of onion and garlic extract and treatment with iodine on the haematological parameters in albino wistar rats

	Haematological parameters				
Treatment groups	HB (g/100 mL)	PCV (%)	MCHC	RBC(N/mm^3) X10^6	WBC(N/mm^3) x10^6
Control(placebo)	17.30±0.17	47.25±1.39	37.16±0.97	24.75±0.75	4.63±0.15
LDG	16.09±0.69	43.00±3.63	37.55±2.10	23.80±0.58	4.73±0.12
HDG	15.63±1.49	45.00±4.60	34.79±1.25	28.12±0.66^b	4.78±0.19
LDO	17.44±2.86	44.75±2.05	38.85±5.03	23.62±0.94	3.96±0.96
HDO	15.85±1.00	44.25±3.81	36.01±3.25	28.80±1.01^b	3.21±0.39
KI (Positive control)	14.53±0.88^a	41.50±3.74^a	35.23±3.39	24.45±0.77	4.36±0.22
KI+LDG	15.06±1.42	41.00±1.30^a	37.61±1.39	20.55±1.28	3.71±0.27
KI+HDG	17.62±1.35	45.50±3.89	38.85±3.05	23.39±0.69	3.39±0.13
KI+LDO	15.69±0.76	44.50±3.89	35.10±2.73	22.64±0.40	3.28±0.19
KI+HDO	15.44±1.05	39.25±3.33^a	39.40±1.34	23.47±0.58	3.35±0.13

Superscripts a, b.... indicate the level of significance in with the respective controls
a = p<0.05: b = p<0.001 Values are presented as mean±SD LDG = Low dose garlic HDG = High dose garlic
LDO = Low dose onion HDO =High dose onion KI = Potassium iodide

The blood hemoglobin concentration in g/100 mL of the experimental animals ranged between 17.62±1.35 g/100 mL for the high dose garlic group treated with iodine and 14.53±0.88g/100 mL for the group treated with iodine only (positive control).There were changes in the hemoglobin level of both normal and iodine treated rats administered with onion and garlic but when compared to the control group these were not statistically significant (p<0.05). However, treatment with iodine led to a statistically significant (p<0.05) decrease in the hemoglobin levels in positive control group.

The percentage packed cell volume (PCV%) for the experimental animals ranged between 47.25±1.39% for the control group and 39.25±3.33% for the high dose onion group treated with iodine. Treatment with iodine and the *Alliums* led to a decrease in percentage PCV of the animals compared to the normal animals. However, this decrease was statistically significant (p<0.05) in the group treated with iodine only (positive control) and the high dose onion group.

The mean corpuscular hemoglobin concentration (MCHC) obtained in this study ranged between 39.40±1.34 for the high dose onion group treated with iodine and 34.76±1.26 for the high dose garlic group normal rats. Although there were changes in the MCHC in the experimental animals, the treatments did not lead to any statistically significant (p<0.05) changes in the MCHC of the experimental animals when compared to the control group.

The mean values for the red blood cell count (RBC) ranged between 28.80±1.01 x 10^6 N/mm^3 for the high dose onion group normal rats and 20.55±1.28 x 10^6 N/mm^3 for the low dose garlic group with iodine treatment. Oral administration of high dose onion and garlic led to a statistically significant (p<0.001) increase in the RBC of the normal experimental rats while oral administration of low dose onion and garlic led to a statistically significant (p<0.001) decrease in the iodine treated rats.

The mean values for white blood cell (WBC) count ranged between 4.78±0.19 x 10^6 N/mm^3 and 3.21±0.39 x 10^6 for the high dose garlic normal rats and high dose onion normal rats, respectively. Oral administration of onion and garlic did not lead to any statistically significant (p<0.001) alteration in the WBC of both normal and iodine treated rats.

DISCUSSION

The effect of oral administration of onion and garlic on the haematological parameters of both normal and iodine treated rats as well as the effect of the oral administration of iodine in form of potassium iodide on albino Wistar rats have been studied. Haematological parameters are closely associated with the reaction of animals to the environment (Fernandes and Mazon, 2003). Changes in haematological parameters may be due to modifications in cellular integrity, membrane permeability and metabolism, or even due to exposure to toxic chemicals (Aseervatham, 2010). The effect of onion and garlic on haematological parameters have been carried out extensively in many studies (Samson *et al*., 2012; Ugwu and Omale, 2011; Tende *et al*., 2012). Differing results have been recorded depending on the duration of the study, concentration of the *Alliums* administered and the method of analysis (Omotoso *et al*., 2009; Banerjee and Maulik, 2002). Samson *et al*. (2012) recorded a dose dependent significant (p<0.05) increase in Red Blood Cell count, hematocrit, hemoglobin concentration, while MCHC was significantly (p<0.05) decreased in a dose dependent manner in both onion and garlic treated groups when compared to the control. Ugwu and Omale (2011) however recorded a contrary effect. They observed no statistically significant changes in the RBC, WBC and PVC of the animals treated with onion and garlic and the control group. This is in agreement with some results recorded for the present study.

In this study however there was a statistically significant (p<0.05) increase in RBC count of the normal rats administered with high doses of onion and garlic. This observation is in agreement with the observations of Samson *et al*. (2012) and Tende *et al*. (2012).They attributed this increase in RBC to competition of the components of these *Alliums* with hemoglobin in the red

blood cell for oxygen resulting in hypoxia which then stimulates red blood cell production. This was however not observed in the iodine treated animals, this could be attributed to a masking of the effects of the *Alliums* due to the presence of potassium iodide. However, the precise mechanism for the action is yet unknown. Thus from this, it may be deduced that adding potassium iodide could help to reduce the hypoxic effect due to the consumption of the high dose of *Alliums*.

A statistically significant ($p<0.05$) decrease in both the total hemoglobin and hematocrit (PCV%) was recorded in the iodine treated group without *Alliums* (positive control group) treated with iodine. Treatment with the iodine salt in form of potassium iodide led a decrease in the hemoglobin level of these animals as well as the hematocrit of these animals. The reason for this decrease is yet unknown but may be attributed to the interaction between the components of hemoglobin and the potassium iodide. This decrease in hematocrit which was not noticed in the normal rats was also noticed in both the low dose garlic and high dose onion group treated with iodine. Although the relationship between hemoglobin or hematocrit and potassium iodide cannot easily be verified, possible molecular mechanisms relating these events still need to be studied.

Conclusion: The results of this study have shown that aqueous extracts of some *Alliums* bring about an increase in the red blood cells in normal albino Wistar rats. Conversely, iodine treated rats did not show such increase in red blood cell when treated with these *Alliums*. Furthermore, treatment with iodine in form of potassium iodide led to a decrease in hemoglobin and hematocrit of the animals. This consequently calls for the necessity for attentiveness in iodization of salt particularly in areas where nutritional anemia is already an issue.

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