A COMPARATIVE STUDY ON THE EFFECTS OF HONEY AND SOME SELECTED BEVERAGES ON HAEMATOLOGICAL PARAMETERS IN MALE WISTAR RATS

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Abstract

This study investigated a comparative study of the effects of honey and some selected beverages on haematological parameters in male wistar rats. Twenty five male rats (180-220g) were assigned into 5 groups of 5 animals each, such that the rats in groups I, II, III, IV and V received orally 1mL distilled water, 0.2mg/kg body weight (BW) of honey, 0.2mg/kg BW of vitamin C, 0.3mg/kg body weight (BW) of zobo and 0.2mg/kg BW of cocoa power, respectively. White blood cells (WBC), red blood cells (RBC), neutrophils, eosinophils, pack cell volume (PVC), haemoglobin (Hb) and lymphocytes were measured using standard methods. There were no significant differences ($p>0.05$) in WBC, lymphocytes, neutrophils and eosinophils for all the treated groups when compared to the control. However, there were significant increase ($p<0.05$) in RBC, Hb and PVC for honey, zobo and cocoa powder -treated groups when compared to vitamin c-treated group and the control. There were also significant increase ($p<0.05$) in RBC and Hb for honey and zobo-treated groups when compared to cocoa powder, vitamin c-treated groups and the control. This study showed that honey, zobo and cocoa powder stimulate haematological parameters most especially, RBC, PCV and Hb respectively. It is therefore recommended that honey, zobo and cocoa powder could be taken by people with shortage of RBC, PCV and Hb respectively. In addition, honey and zobo are highly recommended because of their high stimulatory effects on haematological parameters.

Keywords: Haematological parameters, Honey; zobo, Cocoa powder, Vitamin c

Introduction

Natural honey has both medicinal and nutritional values (Abubakar 2012; Othman, 2012). Studies have shown that honey has both enzymatic and nonenzymatic antioxidants which include: catalase, flavonoids and other polyphenols, as well as vitamins such as thiamine, riboflavin, pyridoxine, pantothenic acid, ascorbic acid, and nicotinic acid (Kishore, Halim, Syazana and Sirajudeen, 2011; Abubakar, Abdullah, Sulaiman and Suen, 2012). The leaf, fleshy calyx, seed and fibre of *Hibiscus sabdariffa*, are very important herbs belonging to the family Malvaceae and is usually cultivated in all parts of the world (Dalziel, 1973). It is mostly planted in the northeast and middle belt regions in Nigeria. It has two major species which are well known in Nigeria, *H. sabdariffa* with red calyces as well as *Hibiscus rosasinesis* with green calyces (Babalola, 2000). The dried calyces of H. sabdariffa are used as medicinal herb and raw material for the production of a local soft drink commonly known as 'zobo' in Nigeria (Usoh, Akpan, Etim and Farombi, 2005).

Vitamin C (Ascorbate) is a cofactor in at least eight enzymatic reactions, including several collagen synthesis reactions that, when dysfunctional, cause the most severe symptoms of scurvy. In animals, these reactions are especially important in wound-healing and in preventing bleeding from capillaries. Ascorbate may also act as an antioxidant against oxidative stress. Food sources such as Grape fruit juice, Strawberries, Tomato, Sweet red pepper, Broccoli, Orange, Straw berries, Tomato, Sweet red pepper, juice and vegetables are the major sources of vitamin C.

Several minerals such as calcium, copper, magnesium, phosphorus, potassium, sodium and zinc are found in cocoa powder (Steinberg, Bearden and Keen, 2003). Study has shown that cocoa powder is mostly used as anti-oxidants because it contains flavonoids (Gressner, 2012). Thus, vitamin C and cocoa powder have little or no effects on haematological parameters most especially WBC, lymphocytes, neutrophils, eosinophils and PCV respectively. This study therefore, compared the stimulatory effects of honey, zobo, cocoa powder and vitamin c on some haematological parameters in male wistar rats.

Methodology

Animals

Twenty five male rats (180-220g) were used for this experiment. They were obtained from the Department of Biochemistry, University of Ilorin, Ilorin, Kwara State, Nigeria, were housed at room temperature with unrestricted access to diet and water and maintained on a daily light/dark cycle. Principles of laboratory animal care were followed.

Fresh red flowers of H. Sabdariffa (with the calyces) were bought directly from Kalaah farm at Mubi, Adamawa State of Nigeria. The calyces were identified using the identification key of Morton (1987). The plant materials were then air-dried for four weeks under room temperature after which the dried plant materials were weighed again to determine the appropriate moisture content and ground into powder using a laboratory milling machine (Thomas Willey model 4, USA). The method of extraction followed that of Carvajal-Zarrabal (2009). 200g of the powdered plant material was introduced into 1000 mls flat bottom flask and 250mls of distilled water was added. The content was mixed thoroughly and left for 24 hours with an occasional shaking to increase the extraction capacity. Thereafter, the soaked substance was filtered with Whatman filter paper (grade 1: 11 μm) and the resulting filtrate dried into powder using a rotary evaporator (Stuart, model RE-300, UK). The solid extract was weighed and re-dissolved in distilled water according to the body weights of the animals for oral administration.

Vitamin C (250mls) used for the study was obtained from, Momrota Pharmacy, Ilorin, kwara State.
Cocoa powder (150g) was bought from Ile-Oluji, Ondo State, Nigeria. 300mls of distilled water was added to it and shaked vigorously until it formed solution. The solution was administered orally depending on the weight of the animals. Honey (300mls) was purchased from University of Ilorin, Ilorin, Nigeria.

After 2 weeks of acclimatization, animals were randomized into five groups (I–V) of five animals each. Animals in Groups I, II, III, IV and V were given orally 1mL of distilled water, 0.2mg/kg body weight (BW) of honey, 0.2mg/kg BW of vitamin C, 0.3mg/kg body weight (BW) of zobo and 0.2mg/kg BW of cocoa power, respectively. The doses were administered twice daily (morning and evening) for 30 days. The Animals were sacrificed under ether aesthesia after the last treatment.

The male rats were anesthetized with ether, dissected and their blood was collected through cardiac puncture with a 2ml syringe into an Ethylene Diamine Tetra-Acetic Acid (EDTA) sample bottle for the determination of the haematological parameters. The red blood cell (RBC) was counted with haemocytometer, the packed cell volume (PCV) by the microhaematocrit method and the haemoglobin (Hb) concentration by cyanmethaemoglobin method. The total white blood cell count was determined manually using the improved Neubauer haemocytometer while the differential leucocytes counts were

determined by morphological identification and counting of leucocytes in Giemsa stained smears of each blood sample. Eosinophil was expressed as percentages of the total white blood cell.

Results

Results were expressed as the mean ± standard error of mean. Data were analyzed using a one-way analysis of variance, followed by the LSD post-hoc test to determine significant differences in all the parameters with Students Package for Social Science, version 20.0 (SPSS Inc., Chicago, USA). Differences with values of $P<0.05$ were considered statistically significant.

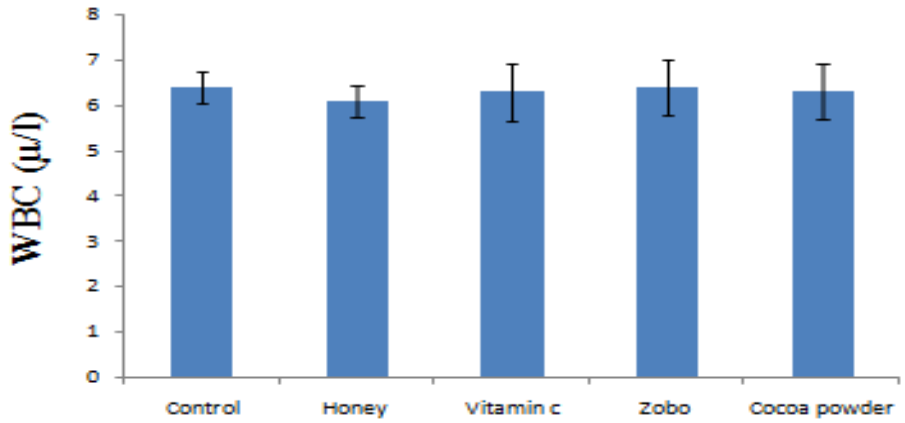

Fig. 1: WBC of rats that received honey, vitamin c, zobo and cocoa powder respectively. Values are expressed as mean ± S.E.M. p>0.05

There were no significant (p<0.05) differences among the treatment groups and the control

NOTE

WBC: White blood cell

S.E.M: Standard error of mean

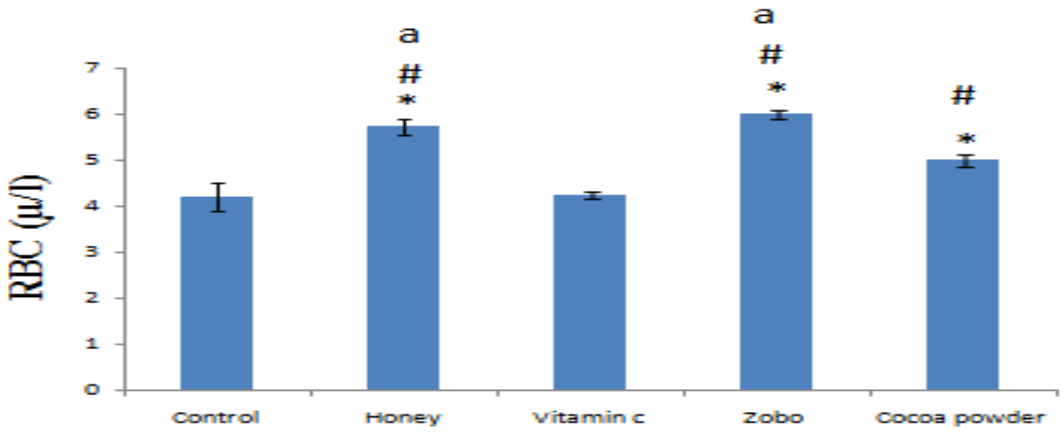

Fig. 2: RBC of rats that received honey, vitamin c, zobo and cocoa powder respectively. Values are expressed as mean ± S.E.M. *p<0.05 vs control; $^{\#}$p<0.05 vs vitamin c; ^{a}p<0.05 cocoa powder.

There were significant (p<0.05) differences among the groups treated with honey, zobo and cocoa powder when compared to the control. There were also significant (p<0.05) differences among the groups treated with honey, zobo and cocoa powder when compared to the vitamin c treated group. Also, There were significant (p<0.05) differences between the groups treated with honey and zobo when compared to the cocoa powder treated group. However, there was no significant (p>0.05) difference between the group treated with vitamin c when compared with the control.

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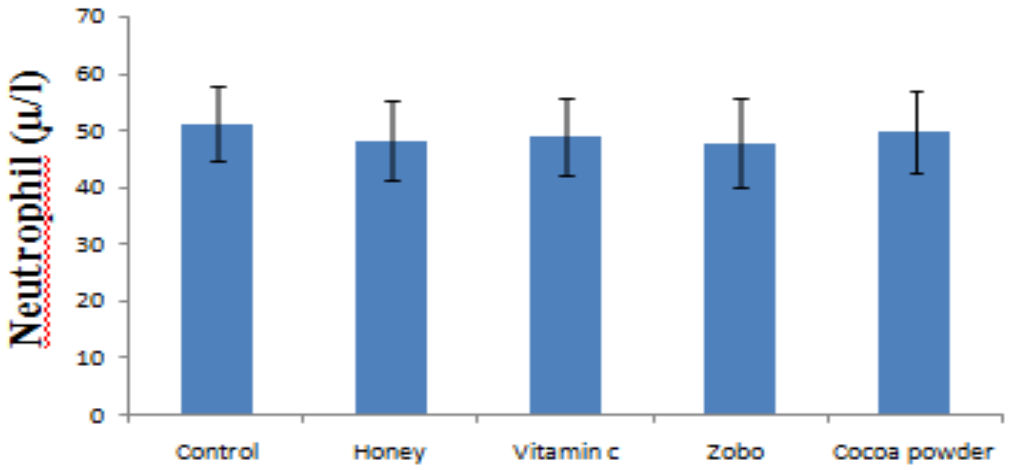

Fig. 3: Neutrophil of rats that received honey, vitamin c, zobo and cocoa powder respectively. Values are expressed as mean ± S.E.M. p>0.05

There were no significant (p<0.05) differences among the treatment groups and the control

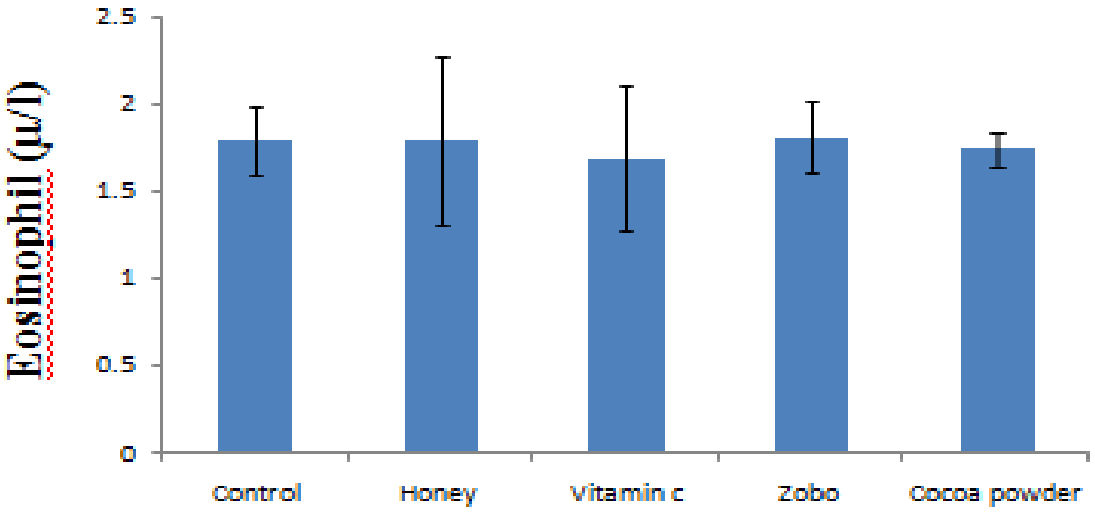

Fig. 4: Eosinophil of rats that received honey, vitamin c, zobo and cocoa powder respectively. Values are expressed as mean ± S.E.M. p>0.05

There were no significant (p<0.05) differences among the treatment groups and the control

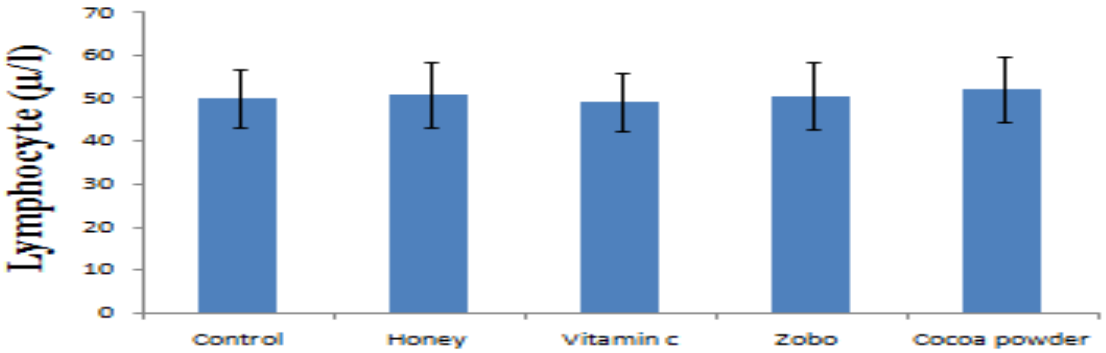

Fig. 5: Lymphocyte of rats that received honey, vitamin c, zobo and cocoa powder respectively. Values are expressed as mean ± S.E.M. p>0.05

There were no significant (p<0.05) differences among the treatment groups and the control

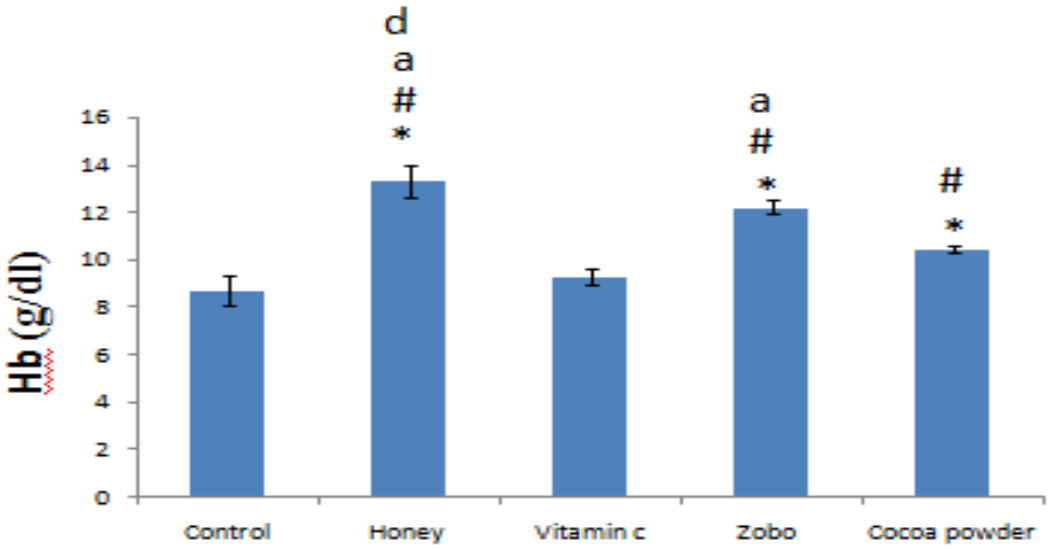

Fig. 6: Hb of rats that received honey, vitamin c, zobo and cocoa powder respectively. Values are expressed as mean ± S.E.M. *p<0.05 vs Control; $^{\#}$p<0.05 vs vitamin c; ^{a}p<0.05 cocoa powder; ^{d}p<0.05 vs zobo.

There were significant (p<0.05) differences among the groups treated with honey, zobo and cocoa powder when compared to the control. There were also significant (p<0.05) differences among the groups treated with honey, zobo and cocoa powder when compared to the vitamin c treated group. Also, There were significant (p<0.05) differences between the groups treated with honey and zobo when compared to the cocoa powder treated group. There was significant (p<0.05) difference between the group treated with honey when compared with other treatment groups and the control. However, there was no significant (p>0.05) difference between the group treated with vitamin c when compared to the control.

NOTE

HB: Hemoglobin

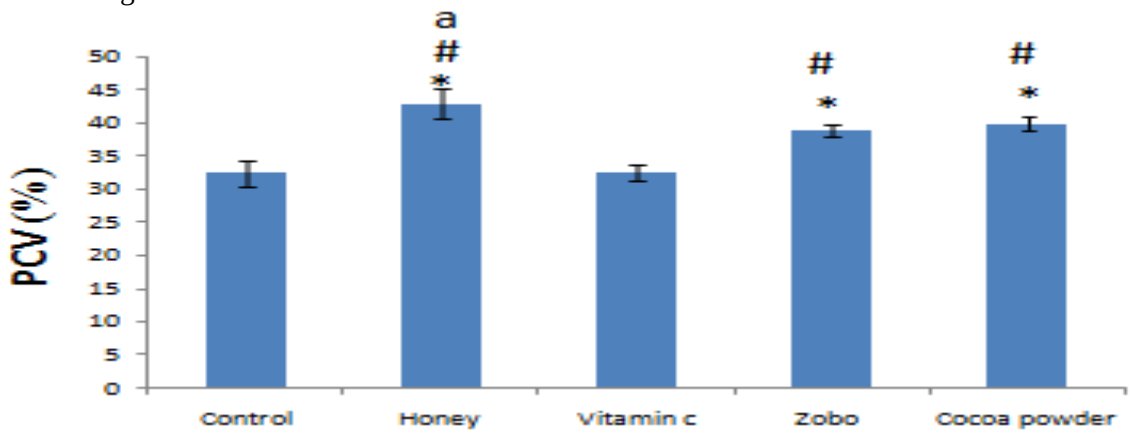

Fig. 7: PCV of rats that received honey, vitamin c, zobo and cocoa powder respectively. Values are expressed as mean ± S.E.M. *p<0.05 vs Control; $^{\#}$p<0.05 vs vitamin c; ^{a}p<0.05 zobo and cocoa powder.

There were significant ($p<0.05$) differences among the groups treated with honey, zobo and cocoa powder when compared to the control. There were also significant ($p<0.05$) differences among the groups treated with honey, zobo and cocoa powder when compared to the vitamin c treated group. There was significant ($p<0.05$) difference between the group treated with honey when compared with other treatment groups and the control. However, there was no significant ($p>0.05$) difference between the group treated with vitamin c when compared with the control.

Discussion

An increase in haemoglobin (Hb) content, packed cell volume (PCV) and red blood cell (RBC) count obtained in rats treated with honey, zobo and cocoa powder could be due to their stimulatory effects on the bone marrow. In addition, increase in Hb and RBC in the rats treated with honey and zobo suggested that they could be used in the management of anemia has reported by Adigun, Ogundipe, OAnetor and Odetunde (2006). It has also been reported that haemoglobin concentration and packed cell volume are used to detect severity of anemia to monitor an anemic patient's response to treatment (Chesbrough, 2005). Thus, this study further suggested the anti-anemic potentials of honey, zobo and cocoa powder.

However, vitamin c has no effect on haematological parameters. This could be due to the fact that it is mostly used as an anti-oxidant (Szeto, Tomlinson and Benzie 200l; Liu, 1995). In addition, there were no differences in WBC, lymphocytes, neutrophil and eosinophil between the treated groups and the control. This could be attributed to the fact that all these parameter are part of the immune cells which play important role in protection against pathogens (Ganong, 2010) and the animals used in this study were not exposed to toxic substances which could stimulate their immune system. The findings in this study suggest that honey, zobo and cocoa powder extract may be useful in improving the Hb, RBC and PCV for a long term administration.

Conclusion

This study concluded that honey, zobo and cocoa powder could be used to stimulate haematological parameters most especially, RBC, PCV and Hb respectively. However, they have no effect on WBC, lymphocytes, neutrophils and eosinophils. It also showed that vitamin C has no effect on haematological parameters. It is therefore recommended that honey, zobo and cocoa powder could be taken by people with shortage of RBC, PCV and Hb respectively. In addition, honey and zobo are highly recommended because of their powerful stimulatory effects on haematological parameters.

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