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Research Article

Effects of Acute Oral Administration of *Hibiscus sabdariffa* Tea on Blood Pressure and Pulse Rate in Apparently Healthy Humans: A Randomized Double-blind Placebo-controlled Study

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ABSTRACT

Background: Although evidence suggests that acute ingestion of the aqueous calyx extract of *Hibiscus sabdariffa* inhibits sympathetic nervous system activity, it is not known if this effect is associated with a fall in blood pressure (BP). This study therefore investigated the acute effect of *Hibiscus sabdariffa* tea (HST) on BP and pulse rate using a randomized double-blind, placebo-controlled design.

Methods: Following ethical approval and informed consent, the BP and pulse rate of apparently healthy subjects were measured before (basal) and one hour after the oral administration of 200mg/kg HST (n=20) or a food colourant (16ml/L; n=20) that served as placebo. The change (Δ) in these parameters was obtained by subtracting the basal values from the values obtained in the presence of *Hibiscus sabdariffa* tea or placebo. The results are expressed as mean ± SEM. $P<0.05$ was considered significant.

Results: In the presence of *Hibiscus sabdariffa* tea, the systolic blood pressure (SBP; 106.5±3.0mmHg) fell significantly ($P<0.05$) compared to the basal value (116.7±2.9mmHg), while the diastolic blood pressure (DBP); mean arterial pressure (MAP) and pulse rate (PR) showed no significant difference. In the presence of the placebo, the blood pressure parameters (SBP, DBP and MAP) and PR showed no significant difference from the basal values. However, in the presence of *Hibiscus sabdariffa* tea, the changes (Δ) in blood pressure parameters and PR were significantly ($P<0.0001$) lowered, compared to the change induced by the placebo.

Conclusion: It is concluded that acute consumption of *Hibiscus sabdariffa* tea lowers blood pressure and pulse rate in normotensive subjects.

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INTRODUCTION

Hibiscus sabdariffa L. (roselle; family: *Malvaceae*), is a flowering plant with high nutritional importance that grows in tropical climates (Vadivel, Sriram and Brindha, 2016). It is commonly grown for the nutritional value of its various parts, which include the seed, calyx, leaf, and fibre (Dalziel, 1973). In some cultures, it is used as a blood pressure lowering agent in traditional medicine and as a soft drink (Ajay *et al*., 2007; Hopkins *et al*., 2013). It is also employed in folk medicine to treat cancer (Chang *et al*., 2005), renal dysfunction (Da-Costa-Rocha *et al*., 2014), among other ailments.

Hibiscus sabdariffa calyx extract has been reported in various studies to lower blood pressure in animals (Odigie, Ettarh and Adigun, 2003; Mojiminiyi *et al*., 2007; Mojiminiyi *et al*., 2012) and in humans (Mozaffari-Khosravi *et al*., 2009; McKay *et al*., 2010; Mozaffari-Khosravi *et al*., 2013). Furthermore, some studies suggest that its antihypertensive effect may be attributed to anthocyanins, polyphenols, and hibiscus acids (Carvajal-Zarrabal *et al*., 2005; McKay *et al*., 2010). Its biological actions include cardioprotective (Jonadet *et al*., 1990), antioxidant (Amin and Hamza, 2005; Hopkins *et al*., 2013;), diuretic (Mojiminiyi *et al*., 2000), cholesterol-lowering (Ochani and D'Mello, 2009), as well as acetylcholine-like, histamine-like, and direct vasorelaxant effects (Adegunloye *et al.*, 1996). Similarly it has angiotensin-converting enzyme inhibitory effects (Ojeda *et al.*, 2010), sympathetic nervous system (SNS) inhibitory action (Aliyu *et al*., 2014; Aliyu *et al*., 2021), inhibition of autonomic nervous system activity with an associated reduction in cardiac workload and cardiac oxygen consumption (Aliyu *et al*., 2021).

Its bioactivity may also occur through the nitric oxide-cyclic guanosine monophosphate (NO-cGMP) pathway and through the inhibition of Ca^{2+} entry into the vascular smooth muscle (Ajay *et al.*, 2007; Alsayed *et al*., 2020).

Earlier studies from our laboratory suggest that the extract of the calyces of *Hibiscus sabdariffa* acts acutely to inhibit sympathetic nervous system activation (Aliyu *et al*., 2014) as well as inhibit the activation of the autonomic nervous system (Aliyu *et al*., 2021). However, these studies provided no direct evidence that *Hibiscus sabdariffa* lowers blood pressure acutely. In addition, these studies were neither blinded nor placebo controlled. An earlier study has shown that administration of *Hibiscus sabdariffa* tea (HST) for six weeks had a hypotensive effect in pre-hypertensive and mildly hypertensive subjects in a randomized double-blind placebo-controlled study (McKay *et al*., 2010).

In the present study, we investigated the acute effect of HST on blood pressure and pulse rate using a randomized double-blind, placebo-controlled design.

MATERIALS AND METHODS

Study design

To explore the effect of acute administration of HST on blood pressure and pulse rate in apparently healthy human subjects, a randomized, double-blind, placebo-controlled trial was carried out. Everyone who took part in the trial maintained their normal diet during the intervention, and were randomly assigned to one of two groups based on a manually constructed randomized list stratified by gender and age. The participants' weights were measured in kilograms (kg) with minimal clothing, and their heights measured without shoes. Their body mass index (BMI) was computed.

Eligibility criteria

All experiments were carried out in line with the principles laid out in the Helsinki Declaration. Students of Usmanu Danfodiyo University, Sokoto, Nigeria, who were in good health and either males or females (n=20 each) volunteered for the study after informed consent and ethical approval from the Specialist Hospital, Sokoto, Nigeria, (protocol no: SHS/SUB/133/2). Volunteers who were 18 to 25 years old with a systolic blood pressure (SBP) of less than 130 mm Hg, a diastolic blood pressure (DBP) of less than 80 mm Hg, and a BMI of 18-25 kg/m2 were recruited. In addition, they did not smoke, were not on any medication that affects the blood pressure (BP) or pulse rate (PR), and did not consume alcoholic or caffeinated beverages, or use illicit drugs. They also did not participate in physical exercise at least 24 hours before the study and had no history of cardiovascular, renal, endocrine, or gastrointestinal disease, and were non-pregnant. A questionnaire was provided to them, and the results were used to establish their health status.

Intervention

Participants who met the above requirements received either one serving (750 mL) of *H. sabdariffa* tea at a dose of 200 mg/Kg body weight or a food colourant that served as placebo.

Test beverages

H. sabdariffa calyces tea (HST) (Hadevco Resources & Investment Limited in Kano, Nigeria) was used for the study. Each tea bag contains 2g of *Hibiscus sabdariffa* calyces powder. Each serving (200mg/kg) of HST was produced by steeping appropriate number of teabags based on the subject's body weight in 750 ml of boiling water for 5 minutes. The placebo was prepared by adding 16-18 drops of an artificial pink food colouring concentrate (Preema Pink Food Colouring, Preema International Ltd., Luton, UK) in 750 mL of water to generate the same colouration as the *Hibiscus* tea. The HST and placebo were administered cold within 12 hours of production, and neither the investigators nor the subjects were aware of the drink administered during the trial.

Measurement of Blood Pressure (BP) and Pulse Rate (PR)

BP and Pulse rate (PR) were measured at the brachial artery with the cuff at the level of the heart using an automated BP sphygmomanometer (Microlife VSA, Microlife AG, Widnau, Switzerland). The mean arterial pressure (MAP) was computed using the following formula:

$\text{MAP} = \text{Diastolic BP} + \frac{1}{3}\text{ Pulse pressure}$ (Jaja *et al*., 2000)

where pulse pressure = systolic – diastolic pressure.

Each participant reported to the research laboratory of the Department of Physiology, Usmanu Danfodiyo University, Sokoto, Nigeria, and laid in a supine position on a couch in a quiet room for 30 minutes, after which blood pressure and pulse rate (PR) were measured with the cuff at heart level. A BP and PR measurement was obtained at the start of the tests. These were regarded as the casual BP and PR. Thereafter, serial BP and PR readings were measured at 10-minute intervals till three nearly identical values were obtained. The last of these readings was taken as the basal BP and PR (Wood *et al*., 1984).

After the basal BP and PR were measured, the HST or placebo was administered for one hour while the subjects rested. The BP and PR were then measured again to get the values in the presence of HST and placebo. The change (Δ) in the BP parameters and PR was calculated by subtracting the basal value of each parameter from the value obtained in the presence of HST or placebo.

Statistical analyses

The data were presented as the mean and standard error of the mean (SEM). To compare the parameters within either the placebo or HST groups, the paired t-test was employed. The unpaired t-test was utilised to compare the parameters between the placebo and HST groups. Statistical analysis was carried out using the Graphpad prism V5.0 statistics software. $P < 0.05$ was considered significant.

RESULTS

Anthropometric Parameters of participants

The anthropometric data of the study groups given HST or placebo are displayed in Table 1. These parameters showed no significant difference between the HST and placebo groups.

Table 1: Anthropometric parameters of study participants

Parameter	Placebo	Hibiscus tea
Age (years)	21.9 ± 1.9	20.8 ± 1.5
Weight (kg)	62.2 ± 7.5	61.2 ± 5.0
Height (m)	1.70 ± 0.1	1.69 ± 0.1
BMI (kg/m^2)	21.5 ± 1.3	21.4 ± 1.3
Male (n)	10	10
Female (n)	10	10

BMI = Body mass index, n = Number of subjects.

The systolic blood pressure (SBP), diastolic blood pressure (DBP), mean arterial blood pressure (MAP), and pulse rate (PR) with and without *H. sabdariffa* tea and placebo are presented in Table 2. All these parameters tended to increase in the presence of placebo compared to their corresponding basal values, although the increase was not significant. In contrast, the parameters tended to fall following administration of *H. sabdariffa* tea compared to their corresponding basal values.

However, none of these falls was significant except the fall in systolic blood pressure ($P < 0.05$) in the presence of HST compared to the basal value.

The changes (Δ) in blood pressure and pulse rate in the presence of HST compared to placebo are shown in Figures 1 – 4. In the presence of HST, the changes (Δ) in systolic, diastolic, and mean arterial pressures as well as pulse rate differed significantly ($P<0.0001$ each) compared to the changes (Δ) in the presence of placebo.

Table 2: Blood Pressure and Pulse Rate in healthy human subjects following acute administration of *Hibiscus sabdariffa* tea or placebo beverage (Mean ± SEM)

PARAMETER	PLACEBO (n = 20)			HST (n = 20)		
	BASAL	PLACEBO	P-value	BASAL	HST	P-value
SBP (mm Hg)	105.5 ± 2.7	118.9 ± 2.3	NS	116.7 ± 2.9	106.5 ± 3.0	< 0.05
DBP(mm Hg)	65.7 ± 1.7	71.8 ± 1.9	NS	70.9 ± 1.9	63.5 ± 2.3	NS
MAP (mm Hg)	78.9 ± 1.9	87.5 ± 1.9	NS	86.1 ± 2.1	78.1 ± 2.4	NS
PR (beats/min)	69.7 ± 2.5	75.1 ± 2.5	NS	65.5 ± 2.4	60.7 ± 2.3	NS

HST=*Hibiscus sabdariffa* tea; SBP=systolic blood pressure; DBP=diastolic blood pressure; MAP=mean arterial pressure; PR=pulse rate; NS= Not significant.

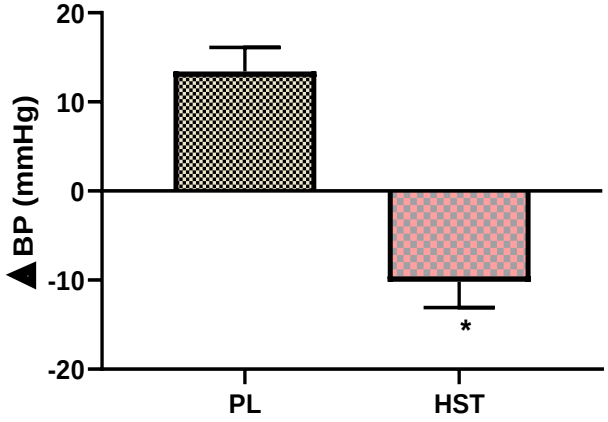

Figure 1: Comparison of changes (Δ) in systolic blood pressure induced by *H. sabdariffa* tea or placebo beverage. BP= Blood pressure; HST = *H. sabdariffa* tea; PL = Placebo, ***=** P < 0.0001 HST vs PL

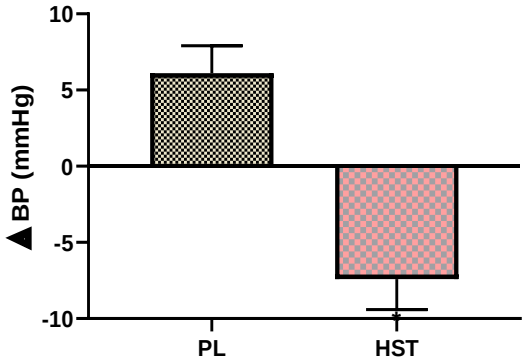

Figure 2: Comparison of changes (Δ) in diastolic blood pressure induced by *H. sabdariffa* tea or placebo beverage. BP= Blood pressure; HST = *H. sabdariffa* tea; PL = Placebo; ***=** P < 0.0001 HST vs PL

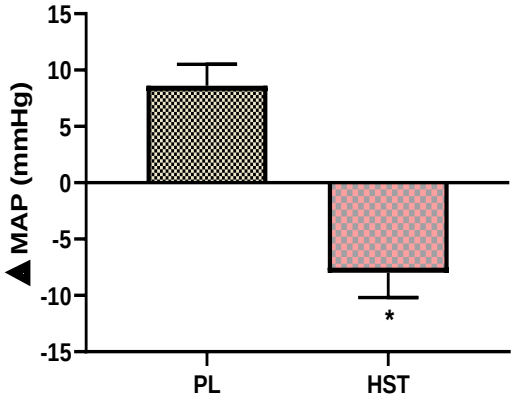

Figure 3: Comparison of changes (Δ) in mean arterial pressure induced by *H. sabdariffa* tea or placebo beverage. MAP= Mean arterial pressure; HST = *H. sabdariffa* tea, PL = placebo, ***=** P < 0.0001 HST vs PL

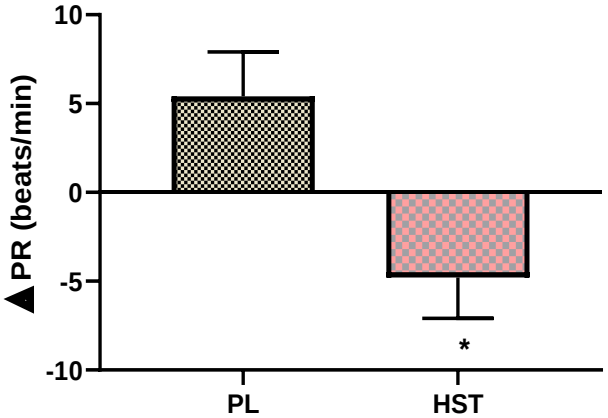

Figure 4: Comparison of changes (Δ) in pulse rate induced by *H. sabdariffa* tea or placebo beverage**.** PR=Pulse rate HST = *H. sabdariffa* tea, PL = Placebo, ***=** P < 0.0001 HST vs PL

Discussion

In this study, the most significant finding was that *Hibiscus sabdariffa* tea (HST) decreased systolic blood pressure acutely compared to the basal value. Another important finding was that HST significantly decreased the Changes (Δ) in blood pressure parameters (SBP, DBP, MAP) and pulse rate (PR) acutely compared to the placebo.

The physical characteristics of the study participants of both groups (HS tea and placebo) did not differ significantly, suggesting that they were properly matched. Thus, the changes in BP and PR parameters seen in this study are likely to be due to the administration of *H. sabdariffa* tea and not to differences in physical characteristics of the participants.

Acute administration of HST led to a fall in only systolic blood pressure while the other blood pressure parameters and pulse rate remained unchanged compared to their basal values. This suggests that acute HST administration lowers the contractile force of the heart that is responsible for the systolic blood pressure. An earlier study showed that the extract of the calyx of *Hibiscus sabdariffa* reduces the cardiac workload and cardiac oxygen consumption in man (Aliyu *et al.*, 2021). It is conceivable that these two observations may be related, that is, the reduction in systolic blood pressure and the reduction in cardiac workload and cardiac oxygen consumption by *Hibiscus sabdariffa.* Since the diastolic blood pressure and mean arterial pressure remained unchanged compared to basal values, it also suggests that acute HST administration may not be affecting the peripheral resistance. It is not clear why acute HST administration lowered only the systolic blood pressure significantly and left the diastolic blood pressure, mean arterial pressure and pulse rate unchanged. This could be due to the short period of administration (1 hour) used in this study. Earlier studies in which *Hibiscus sabdariffa* calyx extract was administered for longer periods of 15 days (Faraji and Tarkhani, 1999) in hypertensive patients and 4 weeks (Mozaffari-Khosravi *et al.*, 2013) in mildly hypertensive diabetic patients showed reductions in both systolic and diastolic blood pressures suggesting that its effect may be time dependent. However, in contradiction to this notion and in agreement with the findings of the present study, administration of *Hibiscus sabdariffa* calyx extract for 1 month or more led to only a significant fall in systolic blood pressure in diabetic (Mozaffari-Khosravi *et al.*, 2008) and hypertensive patients (Herrera-Arellano *et al.*, 2004; McKay *et al*., 2010) leaving the diastolic blood pressure unchanged. These differences probably suggest that larger studies need to be done on the acute and chronic effects of *Hibiscus sabdariffa* calyx extract on blood pressure in health and in disease. Results of the present study indicating that the diastolic, mean arterial pressure and pulse rate fell in the presence of HST but the fall was not significant suggest that the fall might become significant in larger studies using a higher number of subjects.

Despite the above dissonance, the calyx extract of *Hibiscus sabdariffa* has consistently shown its efficacy in acutely inhibiting the discharge of the sympathetic nervous system (Aliyu *et al*., 2014; Aliyu *et al*., 2021) as well as inhibiting parasympathetic withdrawal (Aliyu *et al*., 2021). These actions may also account for the mechanism by which it lowered the systolic blood pressure as observed in this study. It is also noteworthy that when the changes in these parameters in the presence of HST were compared with the changes in the presence of placebo, the differences for all the parameters became highly significant suggesting that HST exerted a hypotensive effect compared to the placebo. As mentioned above, the change was obtained by subtracting the basal value of each parameter from the value obtained in the presence of HST or placebo.

Another interesting observation in this study is that the blood pressure parameters and pulse rate tended to go up in the presence of the food colourant we used as placebo although the rise was not significant. This raises the question as to whether the food colourant (Preema pink food colouring) is an inert placebo or not. Since the rise in the measured parameters was not significant, this suggests that it is an inert placebo. In addition, there is no evidence in the literature suggesting that the food colourant affects blood pressure or other cardiovascular parameters. However, as already suggested above, larger studies may help throw more light on this notion and help answer these questions. The use of the food colourant as a placebo in this study was based on the assumption that it would not affect the blood pressure. Secondly, it produced a colour similar to HST and so produced adequate blinding to both the subjects and the investigators. However, to make assurance doubly sure, the study should have contained another group that was administered the vehicle (water) only in order to rule out any effect the food colourant might have on blood pressure. This was not done. We accept this as one of the shortcomings of this study.

Some earlier studies suggest that acute (1 hour) oral administration of 15mg/kg of *Hibiscus sabdariffa* tablets inhibited sympathetic nervous system activity or parasympathetic withdrawal using maneuvers such as the cold pressor test, hand grip exercise and Harvard step test (Aliyu *et al.*, 2014; Aliyu *et al.*, 2021). The findings of the present study suggest that acute ingestion of 200mg/kg HST for the same duration lowers the blood pressure compared to placebo. Put together, these observations suggest that the former dose (15mg/kg) may be a dose that inhibits sympathetic nervous system activity and parasympathetic withdrawal but leaves the blood pressure unchanged while

the latter (200mg/kg) may be regarded as a dose that lowers it. However, more studies are required to confirm this notion. In summary, acute administration of *Hibiscus sabdariffa* tea significantly lowered systolic blood pressure and significantly lowered the changes in blood pressure parameters and pulse rate compared to changes in these parameters in the presence of placebo. Thus acute consumption of *Hibiscus sabdariffa* tea lowers blood pressure in normotensive humans.

REFERENCES

Adegunloye BJ, Omoniyi JO, Owolabi OA, Ajagbonna OP, Sofola OA, Coker HA (1996). Mechanisms of blood pressure lowering effects of the calyx extract of Hibiscus sabdariffa in rats. Afr J Med Med Sci. 25:235-238

Ajay M, Chai HJ, Mustafa AM, Gilani AH, Mustafa MR (2007). Mechanism of antihypertensive effect of Hypotensive effect of HS and associated autonomic mechanisms 208 Hibiscus sabdariffa L. calyces. J Ethnopharmacol. 109:338-393

Aliyu B, Mojiminiyi FBO, Oyeniran OI, Isu AU (2021). The hypotensive effect of the aqueous calyx extract of *Hibiscus sabdariffa* may occur through the attenuation of Autonomic nervous system activity. Niger J Physiol Sci. 36:203-209

Aliyu B, Oyeniyi YJ, Mojiminiyi FBO, Isezuo SA, Alada ARA (2014). The aqueous calyx extract of *Hibiscus sabdariffa* lowers blood pressure and heart rate via sympathetic nervous system-dependent mechanisms. Niger J Physiol Sci. 29:131-136.

Alsayed AMA, Zhang BL, Bredeloux P, Boudesocque-Delaye L, Yu A, Peineau N, Enguehard-Gueier C, Ahmed EM, Pasqualin C, Maupoil V (2020). Aqueous Fraction from *Hibiscus sabdariffa* Relaxes Mesenteric Arteries of Normotensive and Hypertensive Rats through Calcium Current Reduction and Possibly Potassium Channels Modulation. Nutrients. 12:1782

Amin A, Hamza A (2005). Hepatoprotective effect of *Hibiscus sabdariffa* on azathioprine-induced toxicity in rats. *Life Sciences*. 77:266-278

Carvajal-Zarrabal O, Waliszewski SM, Barradas-Dermitz DM, Orta-Flores Z, Hayward-Jones PM, Nolasco-Hipólito C, Angulo-Guerrero O, Sánchez-Ricaño R, Infanzón RM, Trujillo PR (2005). The consumption of *Hibiscus sabdariffa* dried calyx ethanolic extract reduced lipid profile in rats. Plant Foods Hum Nutr. 60(4):153-159

Chang YC, Huang HP, Hsu JD, Yang SF, Wang CJ (2005). *Hibiscus* anthocyanins rich extract-induced apoptotic cell death in human promyelocytic leukemia cells. Toxicol Appl Pharmacol. 205:201-212

Da-Costa-Rocha I, Bonnlaender B, Sievers H, Pischel I, Heinrich M (2014). *Hibiscus sabdariffa* L.–A phytochemical and pharmacological review. Food chemistry. 165: 424-443

Dalziel JM (1973). The use of plants of West Tropical Africa, The crown agents, London, pp.314-315

Faraji MH, Tarkhani AH, (1999). The effect of sour tea (Hibiscus sabdariffa) on essential hypertension. J Ethnopharmacol, 65(3): 231-236.

Guyenet PG (2006). The sympathetic control of blood pressure. Nat Rev Neurosci. 7:335-346

Herrera-Arellano A, Flores-Romero S, Chavez-Soto MA, Tortoriello J (2004). Effectiveness and tolerability of a standardized extract from Hibiscus sabdariffa in patients with mild to moderate hypertension: a controlled and randomized clinical trial. Phytomedicine. 11(5):375-382.

Hopkins LA, Marnie GL, Janet F, Cheryl R (2013). *Hibiscus sabdariffa* L. in the treatment of hypertension and hyperlipidemia: a comprehensive review of animal and human studies. Fitoterapia. 85:84-94

Jaja SI, Kehinde MO, Gbenebitse S, Mojiminiyi FBO, Ogungbemi AI. (2000). Effect of Vitamin C on Arterial Blood Pressure, Irreversibly Sickled Cells, and Osmotic Fragility in Sickle Cell Anemia Subjects. Niger J Physiol Sci. 16 (1-2):14-18

Jonadet M, Bastide J, Bastide P, Boyer B, Carnat AP, Lamaison JL (1990). *In vitro* enzyme inhibitory and *in vivo* cardioprotective activities of *Hibiscus* (*Hibiscus sabdariffa*). JPharmacol Belgium. 45(2): 120-124

McKay DL, Chen CO, Saltzman E, Blumberg JB (2010). *Hibiscus sabdariffa* L. tea (tisane) lowers blood pressure in prehypertensive and mildly hypertensive adults. J Nutr Dis. 140(2): 298-303.

Mojiminiyi FBO, Adegunloye BJ, Egbeniyi YA, Okolo RU (2000). An investigation of the diuretic effect of an aqueous extract of the petals of *Hibiscus sabdariffa*. J Med Sci. 2: 77-80

Mojiminiyi FBO, Audu Z, Etuk EU, Ajagbonna OP (2012). Attenuation of salt-induced hypertension by aqueous calyx extract of Hibiscus Sabdariffa. Niger J Physiol Sci. 27:195-200.

Mojiminiyi FBO, Dikko M, Muhammad BY, Ojobor PD, Ajagbonna OP, Okolo RU, Igbokwe UV, Mojiminiyi UE, Fagbemi MA, Bello SO, Anga TJ (2007). Antihypertensive effect of an aqueous

extract of the calyx of *Hibiscus sabdariffa*. Fitoterapia. 78(4):292-297

Mozaffari-Khosravi H, Ahadi Z, Barzegar K (2013). The effect of green tea and sour tea on blood pressure of patients with type 2 diabetes: a randomized clinical trial. J Diet Suppl. 10:105-115

Mozaffari-Khosravi H, Jalali-Khanabadi BA, Afkhami-Ardekani M, Fatehi F, Noori-Shadkam M (2009). The effects of sour tea (*Hibiscus sabdariffa*) on hypertension in patients with type II diabetes. J Hum Hypertens. 23:48-54

Ochani PC, D'Mello P (2009). Antioxidant and antihyperlipidemic activity of *Hibiscus sabdariffa* Linn. leaves and calyces extract in rats. J Exp Biol. 47:276-282.

Odigie IP, Ettarh RR, Adigun SA (2003). Chronic administration of aqueous extract of *Hibiscus sabdariffa* attenuates hypertension and reverses cardiac hypertrophy in 2K–1C hypertensive rats. J Ethnopharmacol. 86:181-185.

Ojeda D, Jimenez-Ferrer E, Zamilpa A, Herrera-Arellano A, Tortoriello J, Alvarez L (2010). Inhibition of angiotensin-converting enzyme activity by the anthocyanins delphinidin and cyanidin-sambubiosides from *Hibiscus sabdariffa*. J Ethnopharmacol. 12(1):7-10

Vadivel V, Sriram S, Brindha P (2016). Distribution of flavonoids among Malvaceae family members–A review. Int J Green Pharmacy. 10(1): S33-S45.

Wood DL, Sheps SG, Elveback LR, Schirger A (1984). Cold pressor test as a predictor of hypertension. Hypertension. 6:301-306.