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High eggplant (*Solanum aethiopicum* L.) fruit intake for short duration enhances the antioxidant activity and biochemical parameters in rats

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ABSTRACT

Solanum aethiopicum is consumed in large quantities by various ethnic groups within Nigeria. This study evaluated the effect of *S. aethiopicum* fruit ethanol extract (SAFE) short-term intake at high doses on antioxidant and biochemical properties in rats. Thirty-six male Wistar rats were divided into six experimental groups. Two groups served as controls, while others were treated daily with graded doses of SAFE (500, 1000, 1500 and 2000 mg/kg b.w.) for fourteen days. Liver function, lipid profile, oxidative stress biomarkers, and liver pathology were assayed. Oral administration of SAFE significantly ($p<0.05$) reduced lipid peroxidation while increasing superoxide dismutase activity and glutathione concentration. Biomarkers of lipid profile and liver function were significantly ($p<0.05$) improved in SAFE-administered groups without altering the liver microarchitecture. The results suggest that short-term administration of *S. aethiopicum* fruit at high concentrations improves antioxidants and possesses hypocholesterolaemic properties without inducing hepatic alteration associated with its principles.

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INTRODUCTION

Fruits and vegetables have immense nutritional benefits required for optimal body function; thus, their importance cannot be overemphasised with poor diets are among the leading contributors to the global disease burden (Camillo *et al.*, 2023; Charlebois *et al.*, 2023). In addition to these benefits, they play important roles in the regulation of biochemical parameters, combat and remedy various disease conditions as a result of their chemical phytoconstituents, which are pharmacologically active (Ding *et al.*, 2023). Fruits and vegetables are crucial in maintaining a healthy lifestyle, due to a myriad of mineral, vitamin, fibre, and phytochemical constituents that address malnutrition, boost immune system, and tackle disease pathogenesis across all age groups (Le Turc *et al.*, 2024). Natural products obtained from medicinal plants, fruits and vegetables have been utilised in managing and treating various ailments. These natural products are not only pharmacologically active but cheap with minimal side effects compared to synthetic therapeutic agents that have underlying side effects associated with constant use (Nwanna *et al.*, 2014, 2019b). Phytocompounds such as carotenoids, flavonoids, anthocyanins, other polyphenolics, and vitamins have been reported to reduce disease risk factors and prevent illnesses by eliciting antioxidant, antidiabetic, hypolipidaemic, membrane protective and other protective properties (Srivastav *et al.*, 2024). Other numerous health benefits of phytochemicals, not limited to lycopene in tomatoes, curcumin in turmeric, gingerol in ginger, organosulfur compounds in allium species, and omega-3 fatty acids in the seeds of cucurbitaceous vegetables (Ajanaku *et al.*, 2022; Ademosun *et al.*, 2024).

Solanum species, also known as garden eggs or eggplants in West Africa, are a good source of plant food cultivated for medicinal, therapeutic and ornamental purposes (Nwanna *et al.*, 2019a; Silva *et al.*, 2020). *Solanum aethiopicum*, *Solanum kumba*, *Solanum gilo* and *Solanum macrocarpum*, to mention a few are common species found and sold all year round in different parts of Nigeria (Onyenibe *et al.*, 2022). *Solanum aethiopicum* L., commonly known as scarlet eggplant, is a vegetable specie in the Solanaceae family and is one of the primary vegetables consumed in tropical Africa. Alongside onion, tomato, okra, and pepper, it ranks among the top five most widely cultivated crops in West Africa (Ponticelli *et al.*, 2023). *S. aethiopicum* is a seasonal plant grown in all parts of Nigeria with the fruits are edible in fresh, dried, cooked or processed form (Edeke *et al.*, 2021). *S. aethiopicum* fruit has many nomenclatures ascribed to it by various Nigerian ethnic groups. The Igbos call it "Aṅara" "Afufa" or "Mkpuruofe", the Yorubas call it "Igbagba" while the Hausas call it "Dauta" (Edeke *et al.*, 2021). In ethnomedicine, *S. aethiopicum* is used in numerous applications but not limited to allergic diseases, overweight, asthma, constipation, dyspepsia and gastro-oesophageal reflux disease (Tunwagun *et al.*, 2020). Different plant parts, ash and powder, are traditionally used to treat ailments such as diabetes, cholera, otitis, bronchitis, toothache, dysuria, haemorrhoids, dysentery, asthenia, and skin infections through decoction (Faraone *et al.*, 2022). *S. aethiopicum* contains high levels of alkaloids, phenolic acids, quercetin, kaempferol, rutin, chlorogenic acid and its isomers compared to other Solanaceae members with lower carotenoid, lutein, and β-carotene content than vegetables like tomatoes, carrots and tomatoes (Gürbüz *et al.*, 2018). Purgative, antioxidant, anti-diabetic, anti-inflammatory, anti-obesity, hypolipidaemic, anti-atherosclerotic, and anti-ulcerogenic activities are some of the reported pharmacological properties (Ekweogu *et al.*, 2020; Han *et al.*, 2021). These numerous beneficial activities are associated with the rich in phenolic and alkaloid phytochemical profiles of *S. aethiopicum* (Mbondo *et al.*, 2018).

S. aethiopicum fruit is one of the few *Solanum* species that have a high amount of calcium, glycoalkaloids, cardiac glycosides and calcinogenic glycosides. These compounds have been associated with food poisoning by *Solanum* species, calcium deposition in tissues and incidence of diarrhoea (Chinedu *et al.*, 2011). Caution has been advised when using or consuming the fruit, with researchers recommending that it should be consumed in small quantities (Han *et al.*, 2021). Nonetheless, it is consumed within Nigeria at a very high rate in large amounts either at home or in ceremonies. These reports prompted the present study to investigate the short-term effect of *S. aethiopicum* ethanol fruit extract intake at high concentrations on antioxidant activity, biochemical parameters and liver histology in male Wistar rats.

MATERIAL AND METHODS

Sample Collection and Preparation

Matured fruits of *Solanum aethiopicum* were bought from the Oja-Ota market in Ota, Ogun State, Nigeria. The fruits were identified and deposited in the Herbarium with Voucher No. SA/CUBio/A021. Fruits were prepared into crude extract using the method described by Iheagwam *et al.* (2020). The fruits were dried under shade, pulverised to a smooth blend, macerated in ethanol (80%) for 72 h, concentrated in a rotary evaporator and stored in a -20 °C until it was ready for use.

Experimental Animals

Thirty-six matured male Wistar rats of about six weeks old, weighing between 120 g to 150 g, were accommodated in the animal house of the Department of Biochemistry under standard laboratory conditions and maintained in plastic cages with standard rat chow and water *ad libitum*. Prior to the experiments, they were first acclimatised for 14 days and fasted overnight with free access to water. The animals were conducted in compliance with the Biological Sciences Research Ethics Committee guide for the care and use of laboratory animals (CU/BIOSCRECU/BIO/2015/007) in accordance with ARRIVE and National Institutes of Health (NIH) guidelines.

Animal Grouping

The rats were divided into six groups (n=6) randomly. Group 1 was administered 1 mL/kg body weight (bw) of water as normal control. Group 2 was administered 10 mg/kg bw of vitamin C, serving as the positive control, while the remaining four groups (SAFE500, 1000, 1500 and 2000) were administered orally with 500, 1000, 1500 and 2000 mg/kg bw of SAFE, respectively. All administration was done by oral gavage at 2 pm WAT daily for 14 days (Iheagwam & Chinedu, 2022).

Preparation of Serum and Organ Homogenate

After the experimental duration, the animals were fasted overnight for 16 hr and sacrificed by cardiac puncture under mild euthanasia using a ketamine/xylazine mixture (10:1 v/v). Fresh blood was collected in heparin tubes while liver tissue was excised, cleaned and weighed. Plasma and liver homogenates were prepared for various biochemical assays while relative organ weight was calculated as outlined by Iheagwam *et al.* (2022). A portion of the liver was submerged in formalin buffer for histological assessment (Iheagwam *et al.*, 2023).

Antioxidant and Biochemical Assessment

Assessment of plasma and hepatic superoxide dismutase (SOD) activity, reduced glutathione (GSH) concentration and malondialdehyde (MDA) concentration were determined according to the methods of Misra and Fridovich (1972), Sedlak and Lindsay (1968) and Buege and Aust (1978), respectively. Total cholesterol, HDL-cholesterol (HDL), LDL-cholesterol (LDL), triglyceride (Trig), alanine transaminase (ALT), aspartate transaminase (AST) and bilirubin (BIL) plasma concentrations were measured using Randox diagnostics kits (UK) according to manufacturer's instruction.

Histopathology Assessment

Liver histology was carried out using the haematoxylin and eosin stain procedure as described by Iheagwam *et al.* (2022).

Statistical Analysis

Statistical analysis of variance between group means was carried out by one-way analysis of variance (ANOVA) and Duncan post hoc test on IBM SPSS v23 (USA). Results were considered statistically significant at $p<0.05$ and expressed as mean±standard error of the mean.

RESULTS AND DISCUSSION

Table 1 shows the effect of *S. aethiopicum* ethanol fruit extract (SAFE) on the animal and organ weight. There was a significant ($p<0.05$) reduction in the body weight gain of SAFE-administered groups when compared with both the normal and positive control, while there was no significant ($p>0.05$) difference in the relative liver and kidney weights. This observable increase in animal weight indicates the good health status of the animals (Ekweogu *et al.*, 2020). The weight gained in the experimental groups was lower probably due to the high crude fibre, low fat and low dry matter present in SAFE corroborated by studies on *Solanum* sp. and other plants (Chinedu *et al.*, 2011, 2017; Eze & Kanu, 2014). The organ weight results suggest there was no organ toxicity during the experimental time frame. This parameter has been reported to be a sensitive indicator of general toxicity (Ekweogu *et al.*, 2020).

There was a significant ($p<0.05$) decrease in the plasma activity of SOD in vitamin C and SAFE-treated animals compared to the untreated rats, while the hepatic SOD activity was not altered in the experimentally treated rats ($p>0.05$) compared with the normal and vitamin C-treated rats. However, the SAFE2000 group exhibited significantly ($p<0.05$) higher plasma SOD activity compared to the control groups (Figure 1). In Figure 2, SAFE and vitamin C oral treatment significantly ($p<0.05$) increased the plasma and liver GSH concentration when compared to the normal group. Nonetheless, the increased plasma GSH concentration in the SAFE500-administered group was significantly lower than ($p<0.05$) in the vitamin C-administered group. SAFE administration did not alter ($p>0.05$) the MDA concentration in the plasma; however, it significantly ($p<0.05$) reduced the liver MDA concentration compared to the normal group (Figure 3). The reduction in the plasma and hepatic SOD activity may be due to the inhibition of their synthesis by calcium and a lack of systemic oxidative stress. Calcium in *Solanum* sp. has been reported to concentrate mainly in the liver and takes over 24 h to be excreted with extended accumulation leading to liver necrosis (Serras *et al.*, 2021; Venkatesh *et al.*, 2014). This finding was further corroborated by studies reporting reduced SOD activity, distorted redox status and enhanced production of oxidative stress by intracellular deposition of calcium in tissues (Vairetti *et al.*, 2021; Valduga *et al.*, 2023). Studies have shown flavonoids and other phenolic compounds are copiously present in *S. aethiopicum* fruit compared to other *Solanum* species, thus, these phytochemicals boost the enzymatic antioxidant defence system by protecting the tissues from oxidative damage (Moraes *et al.*, 2023; Nwanna *et al.*, 2019a). The increase in the GSH levels may be attributed to the high content of flavonoids and polyphenols which induce the expression of nuclear factor erythroid 2-related factor 2 and glutamate-cysteine ligase, an important rate-limiting enzyme in the synthesis of glutathione, concomitantly increasing glutathione reductase activity (Faraone *et al.*, 2022). Glycine, glutamate and cysteine concentrations in *S. aethiopicum* have been reported to be higher than other vegetables and medicinal plants such as *Telfairia occidentalis*, *Calotropis procera* and *Amaranthus hybridus*. Hence, their presence may also be responsible for the increased level of glutathione since they are primary constituents (Aja *et al.*, 2021; Aremu *et al.*, 2023). The phytochemicals and vitamin C present in the fruit might be responsible for the decrease in the hepatic MDA concentration correlating with other *Solanum sp.* studies (Adelakun *et al.*, 2020; Okesola *et al.*, 2020).

Table 1: Effect of *S. aethiopicum* ethanol fruit extract on the animal and organ weight

Groups	WG (g)	RLW (%)	RKW (%)
Normal	39.25±2.12[b]	4.47±0.29	0.99±0.12
Vitamin C	34.99±5.13[a]	3.90±0.12	1.02±0.08
SAFE500	28.13±3.90[ab]	4.19±0.30	0.97±0.11
SAFE1000	26.04±3.33[ab]	4.47±0.13	1.03±0.04
SAFE1500	23.77±4.42[ab]	4.27±0.12	0.92±0.20
SAFE2000	24.27±3.01[ab]	4.20±0.13	0.91±0.05

Values represent the mean±SEM (N=6). [a]$p<0.05$ and [b]$p<0.05$ denote respective statistical difference to normal and vitamin C groups. WG: Weight gain, RLW: Relative liver weight, RKW: Relative kidney weight, SAFE: *S. aethiopicum* fruit extract.

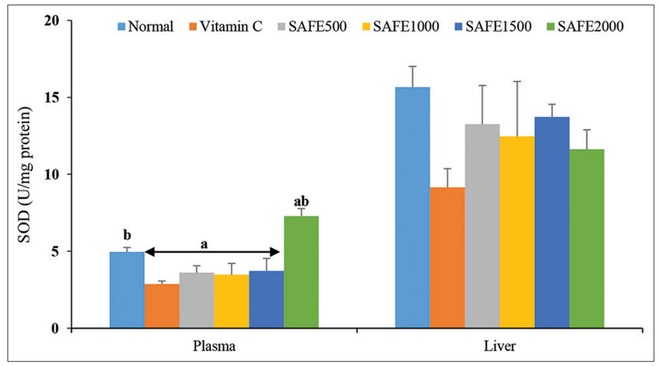

Figure 1: Effect of *S. aethiopicum* ethanol fruit extract on the plasma and liver SOD activity. Bars are expressed as mean±SEM (N=6). [a]$p<0.05$ and [b]$p<0.05$ denote respective statistical difference to normal and vitamin C groups. SAFE-*S. aethiopicum* fruit extract

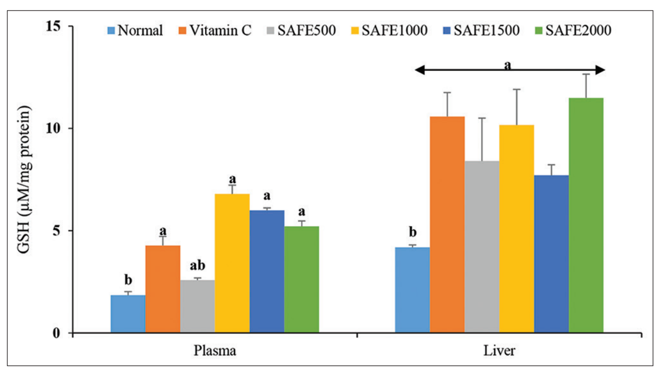

Figure 2: Effect of *S. aethiopicum* ethanol fruit extract on the plasma and liver GSH concentration. Bars are expressed as mean±SEM (N=6). [a]$p<0.05$ and [b]$p<0.05$ denote respective statistical difference to normal and vitamin C groups. SAFE-*S. aethiopicum* fruit extract.

Vitamin C and SAFE administration significantly ($p<0.05$) decreased the plasma cholesterol and LDL levels without altering the triglyceride and HDL concentrations compared to the normal group. However, high SAFE doses (1500 and 2000 mg/kg) significantly ($p<0.05$) increased the HDL

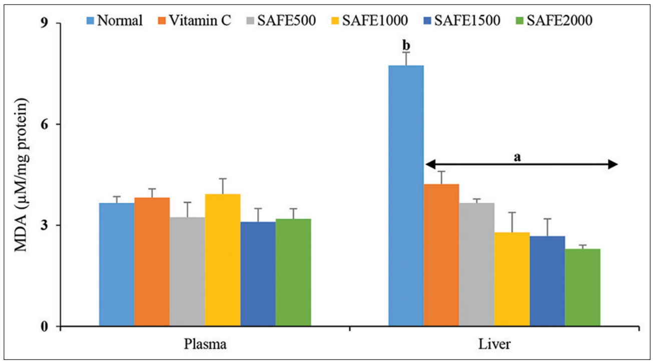

Figure 3: Effect of *S. aethiopicum* ethanol fruit extract on the plasma and liver MDA concentration. Bars are expressed as mean±SEM (N=6). $^{a}p<0.05$ and $^{b}p<0.05$ denote respective statistical difference to normal and vitamin C groups. SAFE-*S. aethiopicum* fruit extract

concentration compared with the control groups (Table 2). The hypolipidaemic effect observed in this study may be ascribed to the presence of saponins and crude fibre in the extract. Crude fibre binds to cholesterol and plays a vital role in gastric emptying, while saponins bind with cholesterol in the lumen, increasing its faecal excretion in the process (Nimenibo-Uadia & Omotayo, 2019). Increased bile acid excretion leads to the rapid synthesis of bile acid from cholesterol in the liver, consequently leading to a drop in the plasma cholesterol concentration (Xie *et al.*, 2024). Polyphenolic compounds such as flavonoids have been reported to reduce hyperlipidaemia by inhibiting hepatic HMG-CoA reductase activity and truncating cholesterol synthesis (Iheagwam *et al.*, 2023). Nwanna *et al.* (2019b) reported similar findings in their work, corroborating the results of this study. The observed increase in HDL cholesterol by *S. aethiopicum* may be a result of HDL cholesterol synthesis induction by flavonoids and phenols (Akinwunmi & Ajibola, 2018; Lela *et al.*, 2023; Ogunka-Nnoka *et al.*, 2018).

Table 3 shows the effect of SAFE on liver function parameters. No significant ($p>0.05$) difference was observed in the plasma ALT activity, AST activity, TBIL and DBIL concentrations between the control and experimental groups. Notwithstanding, 1000 and 2000 mg/kg administration of SAFE was able to reduce the plasma AST activity significantly ($p<0.05$). Figure 4 shows the representative histopathological sections of the liver tissue of normal control, positive control and SAFE-administered groups where a normal cellular architecture was observed in all the hepatic histopathological sections. AST is a sensitive biomarker employed in the diagnosis of hepatic damage. During cellular damage, they are released into circulation from their cytoplasmic location. The extent of tissue damage is proportional to the serum hepatic enzyme activity (Iheagwam *et al.*, 2023). During normal function, the liver removes bilirubin from the blood and excretes it through bile and urine (Gaspari *et al.*, 2024). Results from this study indicate that the structural integrity of the liver was maintained, and the synthetic function of the liver in producing and secreting bile was not affected despite the increased removal of cholesterol. Histopathological results showed no degenerative condition and pathologic abnormality in the liver tissue, correlating the biochemical observations (Iheagwam *et al.*, 2022).

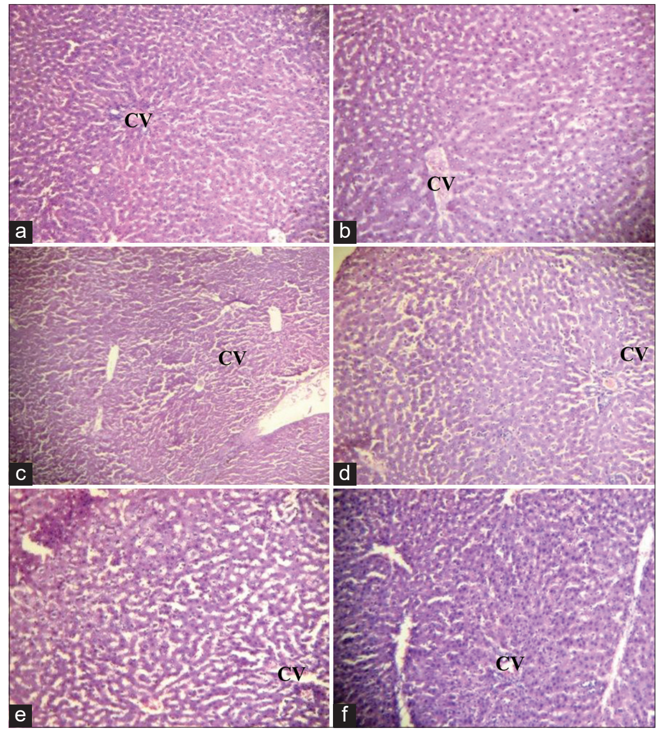

Figure 4: Liver histology sections of a) Normal, b) Vitamin C, c) SAFE500, d) SAFE1000, e) SAFE1500 and f) SAFE2000 showing normal cellular architecture (H&E×400). CV- central vein, SAFE-*S. aethiopicum* fruit extract

Table 2: Effect of *S. aethiopicum* ethanol fruit extract on the lipid profile (mmol/L)

Groups	Cholesterol	HDL	Triglycerides	LDL
Normal	11.38±2.10[b]	0.12±0.02	1.66±0.19	0.94±0.01[b]
Vitamin C	7.88±1.75[a]	0.16±0.01	1.61±0.16	0.29±0.18[a]
SAFE500	8.92±0.26[a]	0.16±0.01	1.68±0.18	0.54±0.25[b]
SAFE1000	5.00±0.38[ab]	0.18±0.02	1.50±0.15	0.80±0.12[b]
SAFE1500	8.35±0.90[a]	0.32±0.09[ab]	2.20±1.08	0.16±0.02[a]
SAFE2000	7.14±0.47[a]	0.23±0.03[ab]	1.61±0.21	0.12±0.01[a]

Values represent the mean±SEM (N=6). $^{a}p<0.05$ and $^{b}p<0.05$ denote respective statistical difference to normal and vitamin C groups. HDL-High-density lipoprotein cholesterol, LDL-Low-density lipoprotein cholesterol, SAFE-*S. aethiopicum* fruit extract

Table 3: Effect of *S. aethiopicum* ethanol fruit extract on the liver function parameters

Groups	ALT (U/I)	AST (U/I)	TBIL (µmol/L)	DBIL (µmol/L)
Normal	17.55±0.86	141.20±28.15[b]	28.67±6.80	50.96±5.79
Vitamin C	18.78±2.27	107.94±9.24[a]	19.88±1.42	40.63±4.08
SAFE500	19.85±2.02	132.43±11.94[b]	22.24±3.99	35.50±4.01
SAFE1000	18.10±0.99	111.05±10.82[a]	27.35±5.53	57.57±5.23
SAFE1500	21.56±1.03	138.52±37.23[b]	25.21±2.64	32.60±2.48
SAFE2000	15.48±1.03	111.85±15.34[a]	24.81±1.54	43.62±3.36

Values represent the mean±SEM (N=6). $^{a}p<0.05$ and $^{b}p<0.05$ denote respective statistical difference to normal and vitamin C groups. ALT-alanine transaminase, AST-aspartate transaminase, TBIL-total bilirubin, DBIL-direct bilirubin, SAFE-*S. aethiopicum* fruit extract

CONCLUSION

Conclusively, short-term administration of *S. aethiopicum* fruit at high concentrations elicits antioxidant and hypocholesterolaemic activities without inducing hepatic alteration associated with its principles. Thus, due to these beneficial effects, S. *aethiopicum* L. fruits may be taken as a food supplement.

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AUTHORS' CONTRIBUTION

Iheagwam FN: Conceived, designed methodology, carried out the experiments, analysed the data, wrote the manuscript's first draft and approved the final manuscript version; Chinedu SN: Conceived, supervised and approved the final manuscript version.

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