Solanum Fruit Juice as a Natural and Sustainable Source of Antioxidants for Patients with Chronic Hepatitis C under Antiviral Therapy

Antiviral Tedavi Altındaki Kronik Hepatit C Hastalarında Doğal ve Sürdürülebilir Bir Antioksidan Kaynağı Olarak *Solanum* Meyve Suyu

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ABSTRACT

Variations in liver metabolism as a result of hepatitis C virus have been established by numerous clinical trials. The use of antioxidants supplements has been reported to minimize the implication of this disease. In this regard, we examined the suitability of *Solanum* fruit juice, a natural source of vitamin C and citrus flavoniod as a precursor for the treatment of patients with chronic hepatitis C. Forty adult patients who were diagnosed with chronic hepatitis C and were under antiviral therapy were divided into two equal groups. Group 1 patients received their antiviral therapy with normal food and water and served as the control group while patients in group 2 were supplemented with *Solanum* fruit juice for eight consecutive weeks. Measurements for Anthropometric data, C reactive protein (CRP), atherogenic indices, biochemical parameters and activities of liver marker enzymes were recorded before and after eight weeks. No alterations were found in waist circumference, body mass and body fat following regular use of *Solanum* fruit juice. The serum levels of oxidative stress markers, LDL-cholesterol, total cholesterol, CRP and atherogenic indices decreased in the *Solanum* fruit juice group when compared to the control group. Moreover, the activities of the liver marker enzyme AST decreased in those who had high levels before the intervention. These results underscore the benefits of *Solanum* fruit juice in the diet of patients with HCV as a result of decreased cholesterol in blood serum, decreased inflammation, and increase in antioxidant capacity as well as maintaining body mass index. This clinical trial is registered at Pan African Clinical Trial Registry (www.pactr.org) with unique identification number PACTR201802003092138.

Keywords: *Solanum* fruit juice, biochemical markers; Chronic hepatitis C; Oxidative stress; Antioxidants

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ÖZET

Hepatit C virüsünün bir sonucu olarak karaciğer metabolizmasındaki varyasyonlar, çok sayıda klinik çalışma ile tespit edilmiştir. Antioksidan takviyelerinin kullanımının bu hastalığın etkisini en aza indirdiği bildirilmiştir. Bu bağlamda, kronik hepatit C'li hastaların tedavisinde öncül olarak doğal bir C vitamini kaynağı olan Solanum meyve suyunun ve narenciye flavoniyodunun uygunluğunu inceledik. Kronik hepatit C tanısı alan ve antiviral tedavi gören 40 yetişkin hasta iki eşit gruba ayrıldı. Grup 1 hastalarına normal gıda ve su ile antiviral tedavi uygulandı ve kontrol grubu olarak görev yaparken, grup 2'deki hastalara sekiz hafta boyunca Solanum meyve suyu takviyesi yapıldı. Antropometrik veriler, C reaktif protein (CRP), aterojenik indeksler, biyokimyasal parametreler ve karaciğer marker enzimlerinin aktiviteleri için ölçümler sekiz hafta önce ve sonra kaydedildi. Solanum meyve suyunun düzenli kullanımının ardından bel çevresi, vücut kütlesi ve vücut yağında herhangi bir değişiklik görülmedi. Solanum meyve suyu grubunda oksidatif stres belirteçleri, LDL-kolesterol, toplam kolesterol, CRP ve aterojenik indekslerin serum seviyeleri kontrol grubuna göre azaldı. Ayrıca, müdahale öncesi yüksek seviyelere sahip olanlarda karaciğer belirteç enzimi AST'nin aktiviteleri azalmıştır. Bu sonuçlar, kan serumunda kolesterolün azalması, azalmış iltihaplanma ve antioksidan kapasitenin artması ve vücut kitle indeksinin korunmasının bir sonucu olarak HCV'li hastaların diyetinde Solanum meyve suyunun faydalarının altını çizmektedir. Bu klinik araştırma, Pan African Clinical Trial Registry'de (www.pactr.org) benzersiz kimlik numarası PACTR201802003092138 ile kayıtlıdır.

Anahtar Sözcükler: *Solanum* meyve suyu, biyokimyasal belirteçler; Kronik hepatit C; Oksidatif stres; Antioksidanlar

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INTRODUCTION

Liver inflammation as a result of infection by hepatitis C virus is one of the characteristics of Hepatitis C; which can result to complications like hepatocellular carcinoma, cirrhosis and fibrosis. The presence of hepatitis C virus (HCV) in the cells leads to oxidative stress as a result of increased cell metabolism, with concomitant increase of liver enzymes aspartate transaminase (AST) and alanine transaminase (ALT) and diminution of antioxidant enzymes (1). Patients with HCV exhibit high concentrations of C reactive protein - which is a marker of inflammation and proinflammatory cytokines like tumor necrosis factor alpha (TNF-α) - which increases the risk of hepatic steatosis and insulin resistance by inhibiting insulin signaling in the liver (2).

According to Huang *et al*. (3), there is an existing relationship between lipid metabolism disorders and elevated levels of C reactive protein. Numerous clinical trials have demonstrated that HCV trails the lipoprotein pathway inside the liver to increase its viral load and survive in the host. The virus replicates in the endoplasmic reticulum upon entry into the cells where it is secreted into the bloodstream bound to very low density lipoprotein (VLDL) (4). HCV displays a high level of infection under this condition; on the contrary the free virus infection efficiency is relatively low (5).

Hepatitis C virus, domicile in the Flaviviridae family, is a single stranded ribonucleic acid (RNA) virus that possesses 9600 nucleotides. It is classified based on six different genotypes as a result of its variability. The preference and monitoring of antiviral therapy is based on Hepatitis C virus genotyping test of the patients (6) which aims to delay disease progression and decrease viral replication (7). Conversely, this adopted therapy is connected with numerous side effects like anemia and nausea, which increases oxidative stress and provokes nutritional depletion. Several studies have over and over again proposed that antioxidant supplements may reduce the venomous effects of both the virus and pharmacological treatment (8).

Solanum fruits commonly called eggplants are one of the biggest genera of plants with over 9000 species of shrubs and climbing herbs in the tropical and temperate regions all over the world and they are extensively consumed in Nigeria. *Solanum* fruit juice contains nutrients and bioactive components of therapeutic appraisal like steriod alkaloid flavonoid and vitamin C (9). They hypolipidemic potentials of *Solanum* fruit juice have also been reported by Onyeike *et al*. (9).

Even though earlier studies propose that antioxidants might improve oxidative imbalance provoked by Hepatitis C virus infection (10, 11), there are no clinical trials on the influence of *Solanum* fruit juice on Hepatitis C virus infected patients.

Consequently, the aim of this study was to discover if *Solanum* juice can play a supporting role in the management and treatment of chronic hepatitis C infection. On this premise, the consumption of *Solanum* juice was investigated in selected patients with chronic HCV and treated with conventional antiviral therapy. The nutritional status as well as the oxidative stress and biochemical parameters was assessed.

MATERIAL and METHODS

Sample collection and preparation

Fresh *Solanum* fruits used in this study were purchased from a local market at Mile 1, Port Harcourt, Rivers State, Nigeria. The botanical identification of the samples was done at the Plant Science and Biotechnology department, University of Port Harcourt.

The samples were sorted to remove dirt and spoilt fruits. The good samples were washed thoroughly using distilled water and homogenized using an electric grinding machine. The juice extract was obtained by pressing the homogenate through a sieve and then filtered with Whatman No. 1 filter paper (9). The juices were stored in air tight plastic containers at 50ºC in the refrigerator until required for use.

Subjects

The Institutional Review Board of the Department of Biochemistry, University of Port Harcourt, Nigeria, granted approval for this study and all volunteers involved signed an informed consent form. This study was in harmony with the ethical principles that have their origins in the Declaration of Helsinki.

The screening and selection of volunteers was performed at University of Port Harcourt Teaching Hospital (UPTH).

Co-authors PU Amadi, IE Archibong, and OT Adeoti assisted the screening of patients with chronic hepatitis C, and CU Iheka and JO Osuoha performed the interviews. During the interview, HCV patients were interrogated with regard to smoking, use of drugs, comorbidities, and consumption of alcohol.

All volunteer patients receiving medical healthcare at the University of Port Harcourt Teaching Hospital were invited to partake in the study: patients registered at UPTH aged > 20 years, positive HCV-RNA serum and under hepatitis C treatment. Prior to the initiation of the study, the volunteers were screened for viral co-infection using immunological tests approved by the Nigerian Ministry of Health: anti-HAV total and IgG for hepatitis A; HBsAg, total anti-HBc for hepatitis B; anti-HIV 1/ 2 for HIV. The detection of negative HBV surface antigen or antibodies to HIV and circulating HCV RNA by polymerase chain reaction using the COBAS AMPLICORTM HCV 2.0 assay were the inclusion criteria of the patients screened as reported by (6). Viral genotyping was performed using the HCV Genotype 2.0 Assay-LiPA.

Forty patients with chronic hepatitis C, who had undergone liver biopsy before treatment with drugs were enrolled. The histological classification of liver biopsy was according to the method of Gonçalves *et al*. (6). Uninfected patients were not included in this proposal because the effect of an antioxidant supplement would not be perceived in such condition.

Experimental protocol

The forty subjects were divided into two equal groups:

Group 1: received only the antiviral therapy and served as the control group

Group 2: received antiviral therapy supplemented with *Solanum* juice (500 ml) daily for eight weeks and served as the test group.

Each participant of the second group was given 100% of *Solanum* juice. They were instructed to drink the juice in two daily portions over the eight consecutive weeks of the experiment. The patients in both groups were asked to maintain their usual diet, lifestyle, and physical activity, and they were monitored by the researchers' team weekly. Evaluation of anthropometric data and collection of blood samples for biochemical analysis were performed in all patients on the first and the last day of the study.

Assessment of anthropometric data

The waist circumference was measured midway between the iliacrest and costal margin. Using a digital scale, the body mass was obtained by trained personnel, and the various heights were recorded using a standing stadiometer. A Lange Skinfold Caliper was used to measure Subscapular, suprailiac skinfolds, biceps, and triceps of the subjects. All measurements were recorded in triplicates.

Biochemical assessment

Following a 12 h fast at the beginning and the end of the experimental period, blood samples (30 ml) were collected for biochemical estimation. The blood serum was separated by centrifugation at 3000 g for 10 min and stored at 80°C until required for analyses. The tests were performed at University of Port Harcourt Teaching Hospital Laboratory. The tissue activities of ALT, AST, ALP, CRP, and gamma-glutamyl transpeptidase (γ-GT) and serum concentrations of glucose triglycerides, total cholesterol, HDL-cholesterol were analyzed using Randox Kits in a spectrophotometer. Insulin was estimated by electrochemiluminescence, and the values for LDL-cholesterol were calculated according to Friedewald *et al*. (12). Insulin resistance was calculated using the homeostasis model assessment index (HOMA-IR) (13) and >2.71 was used as the cutoff point as reported by Gonçalves *et al*. (6).

Oxidative stress assessment

In order to evaluate the total antioxidant capability of the subjects; the 2, 2´-azinobis (3-ethylbenzthiazoline sulfonate) (ABTS) assay was employed (14). In this technique, an $ABTS^{+}$ radical is generated in the assay and the antioxidant activity of the sample against the radical is measured according to the reduction of the $ABTS^{+}$ by the hydrogen-donating antioxidant present on the sample. Trolox was used as standard. Five µl of a 7 mM solution of ABTS was added along with 88 ml of a 140 mM potassium persulfate solution and the mixture left at room temperature, in the dark for 16 h. Before use, the solution was diluted (1:88) with a 10 mM sodium phosphate buffer, pH 7.4 (initial absorbance at 734 nm of 0.7). Five µl of Trolox standard (0, 0.50, 0.75, 1.00, 1.25, 1.50, 1.75, 2.00 mM) and 5 µl of each serum samples were mixed with 300 µl of ABTS+ solution.

After 6 min, absorbance at 734 nm was measured in a microplate reader. Total antioxidant capacity was based on the molar extinction coefficient of Trolox obtained by an analytical curve.

The thiobarbituric acid-reactive substances (TBARS) assay was used as an indicator of lipid peroxidation in subject serum (15). Although not specific for lipid peroxides, because thiobarbituric acid (TBA) also reacts with protein, sucrose and metabolites in the plasma, it is a usual method for this purpose. For the assay performance the 1,1,3,3-tetraethoxypropane (TEP) was used as standard for malondialdehyde (MDA) equivalents (1 mol TEP = 1 mol MDA in reacting with thiobarbituric acid – TBA). Two hundred µl of MDA standard (0, 1.25, 1.88, 2.50, 3.13, 3.75, 6.25, and 12.50 µM) and 200 µl of each serum sample were mixed with 200 µl of sodium dodecyl sulfate (SDS) and then 500 µl of staining reagent (5.3 mg ml–1 of TBA diluted in 20% acetic acid, pH 3.5) were vortexed, incubated at 100°C for 60 min, and cooled on ice for 10 min. The standards and samples were centrifuged at 8000 × g for 10 min, and the absorbance of the supernatant was determined at 532 nm in a microplate reader. TBARS concentration was based on the molar extinction coefficient of MDA obtained by an analytical curve.

Determination of Atherogenic indices

Atherogenic indices were evaluated using the formula reported by Ikewuchi and Ikewuchi (16).

$$\text{Atherogenic Coefficient} = \frac{[\text{Total cholesterol}] - (\text{HDL cholesterol})}{(\text{HDL cholesterol})}$$

$$\text{Cardiac Risk Ratio} = \frac{(\text{Total cholesterol})}{(\text{HDL cholesterol})}$$

$$\text{Atherogenic index of plasma} = \log\frac{(\text{Triglyceride})}{(\text{HDL cholesterol})}$$

Statistical analysis

SPSS Software 20 was used for statistical analysis of the results. We employed the Linear Model of Regression Analysis followed by one way ANOVA to determine significant differences.

RESULTS

Totally 40 patients from initial 60 patients were randomly divided into the *Solanum* juice group (n = 20) and control group (n = 20). The 20 patients that were excluded from the experiment did not satisfactorily meet the study's criteria. No patient was on any form of lipid-lowering or any other interfering medication which could produce a cofounder effect as such medications are known to block the replication and release of the hepatitis virus. The patients who were in the *Solanum* juice group complied effectively with the juice supplementation. Those that were in the control group adhered judiciously to their antiviral medication. Clinical records of the patients used in this study revealed very high occurrence of chronic HCV and moderate inflammatory activity of the liver as shown by a liver biopsy.

The anthropometrical measurements of hepatitis C patients before and after supplementation with *Solanum* juice is shown in Figure 1, no changes were observed in the body weight, body fat, BMI, and waist circumference of both the *Solanum* juice and control groups patients in the beginning and after the study.

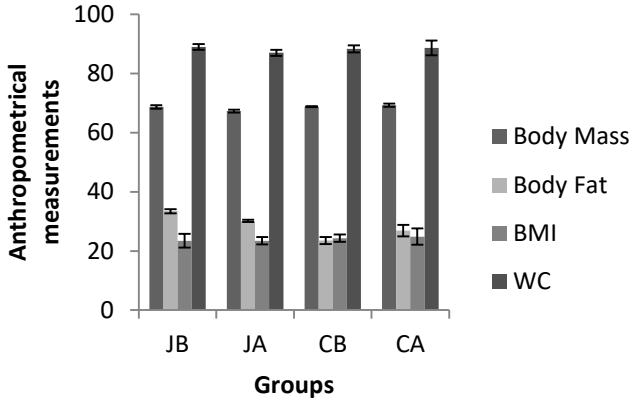

Figure 1: Anthropometrical measurements of hepatitis C patients before and after supplementation with *Solanum* juice.
Mean values ± SD before and after ingestion of 500 ml day–1 orange juice for eight weeks, n=40.
General linear model of repeated measures analysis followed by one-way and post-hoc LSD test, p ≤ 0.05. Bars with similar superscript letters in are significantly different, while those with dissimilar superscript letters are not statistically significant.
BMI=Body Mass Index, WC=Waist circumference.
JB, Juice Baseline; JA, Juice After; CB, Control Baseline; CA, Control After.

Figure 2 shows the antioxidants, CRP and γ-GT measurements of hepatitis C patients before and after supplementation with *Solanum* juice.

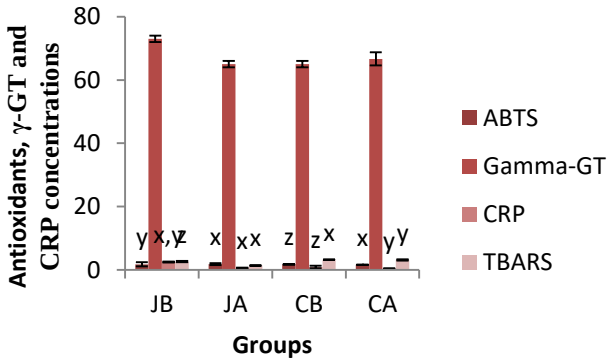

Figure 2: Antioxidants, CRP and γ-GT concentrations of hepatitis C patients before and after supplementation with *Solanum* juice.
Mean values ± SD before and after ingestion of 500 ml day–1 orange juice for eight weeks, n=40.
General linear model of repeated measures analysis followed by one-way and post-hoc LSD test, p ≤ 0.05. Bars with similar superscript letters in are significantly different, while those with dissimilar superscript letters are not statistically significant.
γ-GT= Gamma-glutamyl transpeptidase , CRP= C-reactive protein, ABTS=2,2′-azinobis(3 ethylbenzthiazoline sulfonate), TBARS= Thiobarbituric acid-reactive substances.
JB, Juice Baseline; JA, Juice After; CB, Control Baseline; CA, Control After.

There was a significant increase in ABTS levels in the *Solanum* juice group after supplementation when compared to the control group after antiviral therapy as revealed in Figure 2. A significant change in TBARS was observed in the *Solanum* juice group after supplementation when compared to the control group before administration of the antiviral therapy. Furthermore, there were no changes in the γ-GT levels in both the *Solanum* juice and control groups but there was however a significant decrease in the CRP levels in the *Solanum* juice group after supplementation when compared to before supplementation.

There was also a significant change in CRP levels in the control group after administration of the antiviral therapy when compared to the *Solanum* juice group before supplementation. Figure 3 shows the lipid Profile of hepatitis C patients before and after supplementation with *Solanum* juice.

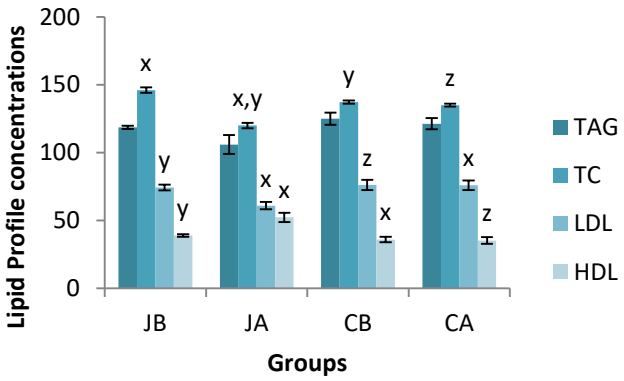

Figure 3: Lipid Profile of hepatitis C patients before and after supplementation with *Solanum* juice.
Mean values ± SD before and after ingestion of 500 ml day–1 orange juice for eight weeks, n=40.
General linear model of repeated measures analysis followed by one-way and post-hoc LSD test, p ≤ 0.05. Bars with similar superscript letters are significantly different, while those with dissimilar superscript letters are not statistically significant.
TAG=Triglycerides, TC=Total Cholesterol, LDL=Low-density lipoproteins, HDL= High-density lipoproteins.
JB, Juice Baseline; JA, Juice After; CB, Control Baseline; CA, Control After.

The lipid profile concentrations are shown in Figure 3. There were no changes in the TAG levels in both the *Solanum* juice and control groups. However, there was a significant decrease in the TC levels in the *Solanum* juice group after supplementation when compared to before supplementation. There was also a significant change in the *Solanum* juice group after supplementation when compared to the control group before administration of the antiviral therapy. Also, there was a significant decrease in the LDL levels of the *Solanum* juice group after supplementation when compared to the control group after antiviral therapy. HDL levels showed a significant increase in the *Solanum* juice level after supplementation when compared to the control group before antiviral therapy. Figure 4 shows the liver enzymes' activities of hepatitis C patients before and after supplementation with *Solanum* juice.

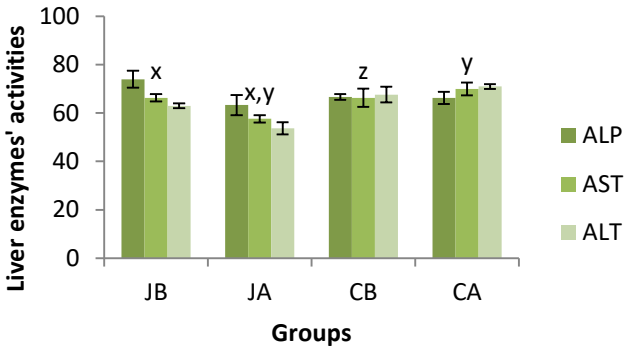

Figure 4: Liver enzymes' activities of hepatitis C patients before and after supplementation with *Solanum* juice.
Mean values ± SD before and after ingestion of 500 ml day–1 orange juice for eight weeks, n=40.
General linear model of repeated measures analysis followed by one-way and post-hoc LSD test, p ≤ 0.05. Bars with similar superscript letters are significantly different, while those with dissimilar superscript letters are not statistically significant.
ALP=Alkaline phosphatase, AST=Aspartate aminotransferase, ALT=Alanine aminotransferase.
JB, Juice Baseline; JA, Juice After; CB, Control Baseline; CA, Control After.

Figure 4 represents the hepatic enzymes' activities in both groups. There were no changes in ALP and ALT activities in both the *Solanum* juice and control groups. However, there was a significant decrease in the AST activities in the *Solanum* juice group after supplementation when compared to before supplementation. A significant decrease in AST activities was also seen in the *Solanum* juice group after supplementation when compared to the control group after administration of the antiviral therapy.

Figure 5 shows the HOMA-IR and insulin measurements of hepatitis C patients before and after supplementation with *Solanum* juice.

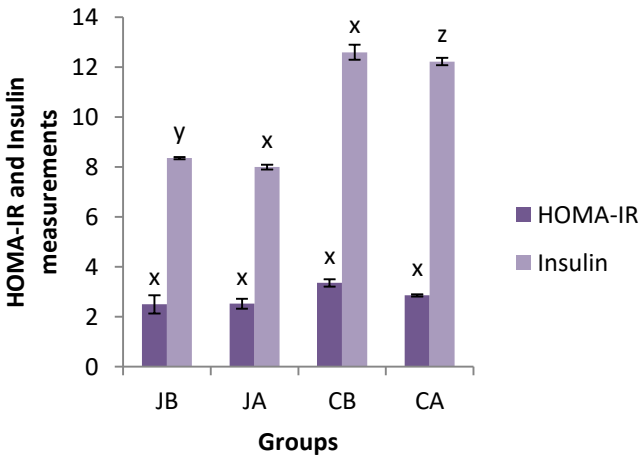

Figure 5: HOMA-IR and insulin measurements of hepatitis C patients before and after supplementation with *Solanum* juice.
Mean values ± SD before and after ingestion of 500 ml day–1 orange juice for eight weeks, n=40.
General linear model of repeated measures analysis followed by one-way and post-hoc LSD test, p ≤ 0.05. Bars with similar superscript letters are significantly different, while those with dissimilar superscript letters are not statistically significant.
HOMA-IR= Homeostasis Model Assessment Index of Insulin Resistance
JB, Juice Baseline; JA, Juice After; CB, Control Baseline; CA, Control After.

The HOMA-IR and Insulin measurements are shown in Figure 5. After the study period, a significant decrease in the insulin concentration in the juice group was observed when compared to the control group. There was also a significant change in the marker of insulin resistance (HOMA-IR) between both groups. The threshold of HOMA-IR in the control group was higher than 2.71 which infers a risk for development of resistance to insulin.

Figure 6 shows the atherogenic indices of hepatitis C patients before and after supplementation with *Solanum* juice.

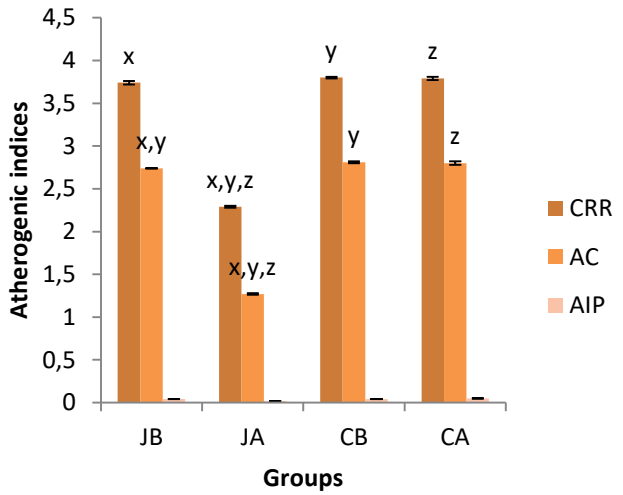

Figure 6: Atherogenic Indices of hepatitis C patients before and after supplementation with *Solanum* juice.
Mean values ± SD before and after ingestion of 500 ml day–1 orange juice for eight weeks, n=40.
General linear model of repeated measures analysis followed by one-way and post-hoc LSD test, $p \leq 0.05$. Bars with similar superscript letters are significantly different, while those with dissimilar superscript letters are not statistically significant.
CRR=Cardiac Risk Ratio, AC=Atherogenic coefficient, AIP=Atherogenic Index of Plasma
JB, Juice Baseline; JA, Juice After; CB, Control Baseline; CA, Control After.

The atherogenic indices are revealed in Figure 6. *Solanum* juice induced significant decreases in the Cardiac Risk Ratio (CRR) and Atherogenic coefficient (AC) when compared to the control group. There were no changes in the Atherogenic index of plasma.

DISCUSSION

Lower quality of life, anorexia and weight loss are frequently exhibited by patients who undergo medication (17). Some authors have implied that due to sugar content, juice intake like that of oranges could result in weight gain (18). This present study after an eight-week treatment with *Solanum* juice did not affect body weight or body fat which could infer a positive outcome in maintaining the patients' nutritional statuses. In patients with chronic hepatitis C, BMI and elevated body fat were significantly associated with hepatic steatosis which may have contributed to the growth of steatosis in those patients as revealed in a study by (19). Apparently, *Solanum* juice did not contribute to those harmful effects as revealed by no changes in BMI, body fat and waist circumference portending a very minimal risk for syndrome development in metabolism (20).

Hepatitis C infection is prominently characterized clinically by oxidative stress (21). Hepatitis C viruses (HCVs) significantly promote the development of hepatic inflammation and the production of more reactive oxygen species (ROS). However, the key to its survival in its hosts is by adaptation to oxidative stress (22, 23). Oxidative stress has been known to be stimulated by antiviral therapy. This is revealed in increased lipid peroxidation and decreased levels of vitamins E, B and C as well as activities of hepatic antioxidant enzymes in patients undergoing anti-HCV therapy (23, 24).

Studies have however shown that antioxidants may influence replication of HCV, levels of hepatic enzymes, make interferon antiviral treatment more efficient and protect against hepatic cell damage (22). This present study revealed an enhanced total antioxidant capacity and changes in TBARS level in the serum by regular intake of *Solanum* juice. Hesperidin has been shown to elevate antioxidant liver enzymes (25) but decreases pro-oxidant enzymes (26). The elevation in the capacity of the antioxidant as shown by this present study could be attributed to vitamin C present in *Solanum* juice. Vitamin C is a natural water-soluble free radical scavenger. From its double bond of the 6-carbon, it has the ability to donate two electrons. This mechanism enables vitamin C in the oxidized state to generate dehydroascorbic acid (DHA), a stable intermediate product.

This product gets absorbed by erythrocytes and again reduced to vitamin C through an endogenous enzyme known as glutathione reductase (27, 28, 29). Vitamin C plays a profound role in recycling vitamin E (α-tocopherol) by oxidation thereby preserving it. Vitamin E plays a similar function to vitamin C, it becomes a less toxic product which is known as α-tocopheroxyl by donating two electrons. This less toxic product must be reduced so it could become an efficient antioxidant vitamin although this mechanism is slower than the recycling of ascorbate. It is therefore possible that vitamin E is recycled in the membrane of the cell through a process that involves enzymatic ascorbate recycling through vitamin E (30).

This present investigation showed normal CRP levels (<1.0 mg l–1) in the patients in the control groups even after the investigation. However, the patients in the *Solanum* juice group exhibited elevated CRP levels before supplementation but these levels were significantly decreased after intake of the *Solanum* juice. HCV infection results in the inflammation of the liver (31). Through stimulation by IL-6 and IL-1β in response to an acute inflammatory process, CRP which is a plasmatic protein is released by the liver. Increased plasma levels of CRP (between 3 and 10 mg l–1) have been observed in low and continuous chronic inflammation conditions (32). HCV infected individuals have a higher CRP when compared to uninfected individuals although the levels in those on antiviral therapy are lower when compared to those that are untreated (33). Devaraj *et al*. (34) have suggested incorporating orange juice as a daily part of a healthy diet as their study in healthy individuals revealed a reduction in CRP and serum cholesterol levels.

There were significant changes in the TC, LDL and HDL levels in the studied groups. There were increases in naringin, hesperidin and vitamin C on consumption of 500ml day-1 of orange juice according to Franke *et al*. (35). These compounds could have been the reason for the decreased levels of TC and LDL-cholesterol in this present study. The levels of the blood serum lipids were within recommended range before this present investigation. This agrees with studies that have shown appropriate levels of TC in patients with chronic hepatitis C (CHC) when compared with uninfected individuals (36, 37). A study by Onyeike *et al*. (9) showed significant decrease in the amount of total cholesterol in all the species and concentraions of *Solanum* juice at the 4th and 6th week. This suggested that these *Solanum* juice may have affected cholesterol biosynthesis which resulted to reduction in the level of cholesterol in the blood. The values for the test groups at the 4th and 6th weeks were significantly ($p>0.05$) different and lower from the negative and test control. These results corroborate the study by (38), who carried out work on the effect of eggplant juice on plasma lipid levels. There is yet no clarity on the mechanism of action of these *Solanum* species on the metabolism of cholesterol. Through a number of mechanisms, soluble fibers reduce blood cholesterol level by 5 to 15% in humans and experimental animals (39). The effect of fiber on enterohepatic circulation through sequestration and bile acids' binding could be one possible mechanism. The *Solanum* juice at all concentrations according to Onyeike *et al*. (9) increased plasma HDL-cholesterol level compared with the test control. Edijala *et al*. (40) who studied the comparative effect of garden egg fruit, oat and apple on serum lipid profile in rats fed a high cholesterol diet reported that garden egg fruit lowered low density lipoprotein.

There are low levels of blood cholesterol in patients with HCV because viral replication in the cells makes use of cholesterol (37). In the hepatocytes, the replication of HCV which produces virions demands TAG and cholesterol. Infection with HCV elicits the biosynthesis of fatty acids with consequent elevation of lipogenesis contributing to turmogenesis and steatosis (41). Hesperidin, naringin and citrus flavonoids which are bioactive constituents in orange juice could decrease the availability of cholesterol to hepatocytes.

This it does by blocking the ACAT2 enzyme, reducing the availability of triacylglycerols to very low density lipoproteins (VLDL) assembly by reducing the MTP's activity which then reduces the biosynthesis and secretion of these lipids of the liver. Decrease in serum LDL cholesterol is the consequence of the inhibition of this very vital step in metabolism in the synthesis of VLDL, stimulating receptor expression of low density lipoprotein (LDL) (42). On ingestion of orange juice or isolated flavones, elevated levels of naringin and hesperidin metabolites have been detected which has been linked to effects of hypolipidemia (43, 44, 45, 46). This therefore infers that flavanoids in citrus could positively interfere with the metabolism of lipid, reducing the synthesis and secretion of the HCV (47).

Many parameters are employed in the diagnosis of HCV. Liver function test in most of the cases is the basis of the prognosis of HCV. The prognosis of HCV infection could be as a result of the abnormal concentration of serum amino transaminases.

As a consequence of liver damage, indicating average liver status, ALT which is most intense in the liver is released into the bloodstream (48). The fact is that the non-A and non-B hepatitis have been based on records of consistently abnormal ALT levels before the molecular based assays for HCV RNA exploitation and HCV detection (49). Persistently normal ALT values are observed in almost 25% of patients with active infection with HCV (50). This present investigation revealed a decrease in AST level in the *Solanum* juice group after supplementation when compared to before the supplementation in the patients. Some investigations have reported that when hesperidin is administered, a significant reduction in serum ALP, AST and ALT levels were observed including a decrease in oxidative stress in lipopolysaccharide-induced toxicity (LPS-induced toxicity) which suggests that this flavanone constituent could protect against damage in the liver (51). Other studies have shown similar results after supplementation with just naringerin (52) or in combination with vitamins E and C in cadmium induced hepatotoxicity in rats (53).

Solanum juice induced significant differences in the HOMA-IR and insulin concentrations in this present study. This result did not corroborate the study by Bin *et al*. (54) that showed no significant effects of fruit juice on insulin concentrations. The study suggested that the possibility could have been because fruit juice has less fibre than whole fruit. The study further suggested that fruit juice intervention could have modestly increased the participants' dietary consumption of sugars and energy, which may influence the total effects of fruit juice on glucose control since most of the trials suggested that the participants maintained their usual diet during the intervention duration. Liu *et al*. (55) suggested that the consumption of fruit juice had no significant favourable outcome on lipid abnormalities which often clusters with insulin resistance.

Atherogenic indices are profound markers of risks related to heart disease; the higher the values, the higher it is likely that a person would develop coronary heart disease and so on (56, 57, 58). This present study revealed that *Solanum* juice could possess anti-atherogenic potentials as it induced reductions in cardiac risk ratio and atherogenic coefficient.

CONCLUSION

This study has revealed the profound role that *Solanum* juice plays in HCV patients who are on antiviral medication. This is because it induced significant changes in aspartate aminotransferase (AST), C-reactive proteins (CRP), serum lipids, oxidative stress, insulin, insulin resistance and atherogenic indices. These effects could have been as a result of the interplay between flavonoids and other vital constituents of *Solanum* juice. Therefore, *Solanum* juice might be a potential dietary supplement for HCV patients undergoing antiviral therapy.

Conflict of interest

No conflict of interest was declared by the authors.

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