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Effect on Lipid Profile Parameters by the Addition of Orange Juice in Diet of Hypercholesterolemic Patients

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Abstract

Proceedings of epidemiologic data revealed that a more consumption of vegetable and fruits allied with a less susceptibility of Acute Heart Disease. Orange juice is a good source of folate, Vitamin C and flavanones, but their effect on heart patients has not been studied scrupulously. Present research focused on fifty subjects having elevated total cholesterol and LDL cholesterol. Participants consumed 2 cups (5000 mL) of orange juice daily into their diets, each dose over a period of 4 week followed by a 5-week washout period. Plasma lipid, folate and vitamin C (a compliance marker) concentrations measured at baseline, after each treatment, and after the washout period. The consumption of calcium, fatty acids, protein total energy and cholesterol did not significantly varied in each of the period.

The dietary interference had no substantial effect on body weight body mass index or plasma lipid concentration. Though total plasma triglycerol and HDL-cholesterol concentration increased 17% and 24% however LDL-HDL cholesterol ratio declined during the study time period.

Keywords: Lipid; Parameters; Hypercholesterolemic; Orange juice

Introduction

Earlier epidemiologic data revealed that more consumption of vegetable and fruits IS directly proportional to less susceptibility of acute Heart Disease [1]. The advantageous outcome may be allied with inconsequential moieties, particularly flavanones, anticipated to apply their deed by restrain oxidation of LDL and thrombosis [2] and vitamins E, C and carotene which act chiefly as antioxidants [3]. Reduction in homo cysteine was reported through Folic acid found in high concentration in green food stuff and in citrus fruit a transitional component in methionine utilization, which is concerned as a threat for heart disease [4]. Orange and grapefruit juice are good source of folate, Vitamin C and flavanones, but their effect on heart patients has not been studied scrupulously. The foremost element citrus hesperidin, flavonoids naringenin from orange and grapefruits are somewhat analogous to genistein, from soybean thought to be hypocholesterolemic. Thus, juice of fruits containing vitamin E and flavonoids may have positive effect on hyper cholesterolemic patients [5].

Primary flavonoids contents of citrus juice were evaluated for cholesterol breakdown in rodents and HepG2 cell line. In rabbit, 43% and 32% reduction in cholesterol level was noted with orange and grape fruit juice in which experimental cholesterol level was induced by a casein-based and the semi cleaned food with citrus juices (reformed from freeze nectar at 2 times the standard potency. Additionally, esters of liver cholesterol lowered down by 42% but fecal cholesterol elimination or bile acids was not elevated, revealing that juice contents like flavanones, may affect beneficially cholesterol metabolism mainly in the hepatic cells. Parallel to this proposition, decreased level of serum cholesterol was detected due to the sterol *O*-acyltransferase-2 and hydroxylmethylglutaryl-CoA reductase, chief catalyst in metabolism of cholesterol-were repressed in cholesterol-fed mice including blend of primarily citrus flavonoids [6]. Additionally by blocking the synthesis of esters of cholesterol, reduced discharge of polyprotein was observed in incubate HepG2 cells with hesperidin and naringenin simultaneously [7]. Citrus juice contain large amount of flavonoids as well as lemonades which also have apoB-lowering effect in HepG2 [8].

Chances of coronary arteries ailment decreases with the Intake of green food stuff and fruits link and this outcome is accredited primarily to the efficient and preventive activity of phytochemicals and vitamins such as flavonoids and phenols [9]. Diet such as grape, tea and wine supply large amount of flavonoids afterward orange and citrus juice.

Substantial amount of flavonoids and its components are present in Orange juice and its quality depends upon processing conditions of juice. Investigational data revealed that anti-inflammatory, hypolipidemic, anti-carcinogenic and antiallergic characteristics are present in hesperidin and these components aid to minimize the LDL cholesterol concentration in serum [10]. Orange juice along with aerobic exercise helps in lowering the risk of heart and coronary disease by lowering triglyceride and uplifting HDL-cholesterol in obese middle aged women [11]. Vitamin C had vasoprotective effect moreover minor concentration of folate have been linked with reduced concentration of hemocysteine and minimal chances of thromboembolic events.

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Material and Methods

Fifty subjects having elevated total cholesterol and LDL cholesterol included in the study. Participants incorporated 2 cups (5000 mL) of orange juice daily into their diets, each dose over a period of 4 week followed by a 5-week washout period. Plasma lipid, folate and vitamin C (a compliance marker) concentrations were measured at baseline, after each treatment, and after the washout period. At the end of this study the variation in lipid profile parameters by orange juice consumption were observed to evaluate the effect of orange juice on lipid profile in hypercholesterolemic subjects.

First IV blood was collected for baseline testing of lipid profile. Then 2nd IV blood collected after consuming the orange juice for 4 weeks and again 3rd blood sample collected after washout period of five weeks for lipid profile (like Triglycerides, HDL, LDL and VLDL), folate level and vitamin C. Estimation of blood cholesterol level was determined by Chylomicrons, VLDL and LDL were precipitated by adding phosphotungstic acid and magnesium ions to the sample. Centrifugation left only the HDL in the supernatant. Their cholesterol content was determined enzymatically using Cholesterol determination kit.

Statstical Analysis

Obtained results were analyzed by using statistical tool ANOVA (two-way) on Statistical Package for the Social Sciences (SPSS) software version 21.0.0. At the end of this study we were able to conclude the variation in lipid profile parameters by orange juice consumption.

Results

Effect of orange juice consumption on lipid profile

Orange juice and energy intake at initial, after each nutritional level and after the washout period are given in Table 3.1. These data was derived from the diet records of the enrolled subjects. The consumption of calcium, fatty acids, protein total energy and cholesterol were not significantly variant in each of the period. Fiber consumption declined gradually during the study and significantly lowers that initial baseline after the washout period. The propensity for the consumption of dietary decreased during the 1, 2, 3 and washout period may be due to successive increase in orange juice intake through study period.

The dietary interference had no substantial effect on body weight body mass index or plasma lipid concentration. Though total plasma triglycerol and HDL-cholesterol concentration increased 17% and 24% however LDL-HDL cholesterol ratio declined during the study time period. Data shows that pair wise comparison showed that percentage variation in HDL cholesterol and LD-HDL ratio was significantly different in the 3rd from that of 1st and 2nd period the variation in initial plasma concentration observed in 3rd period were significantly associated with the VLDL variation.

At the end of the study period the significantly increased HDL-cholesterol and decreased LDL-HDL cholesterol ratio had not reverted to initial values. In fact, the decrease in the LD-HDL cholesterol ratio and increased HDL-cholesterol at the time of washout tend to be higher as compared to the detected in 3rd period. Moreover it was also observed that subjects consuming processed orange juice showed different results of HDL and LDL-HDL cholesterol concentration, subjects consuming processed orange juice had elevated level of HDL and decreased ratio of LDL-HDL ratio as compared to those who intake fresh orange juice.

Concentration of the HDL cholesterol at the time of wash out and changes at the initial baseline in HDL cholesterol after the wash out were not positively correlated. More-over HDL-concentration did not tend to be elevated in subjects who consumed large amounts of orange juice during the washout period. Plasma triacylglycerols and other indexes [BMI, total VLDL, and LDL cholesterol] which were not influenced by the intake of orange juice were not significantly different from baseline during the washout period.

The results showed, changes in the LDL-HDL cholesterol ratio induced by the intake of 500 mL orange juice/d were significantly inversely related to the initial LDL-HDL cholesterol. Similarly, changes in HDL-cholesterol concentrations tended to be inversely correlated with baseline HDL cholesterol but the association was not significant. Changes in plasma triacylglycerols and plasma folate concentrations induced by the highest dose of orange juice were not significantly correlated with the initial LDL-HDL cholesterol ratio or with the initial HDL cholesterol concentration.

Because many previous studies showed that diet-induced increases in serum folate are associated with decreases in plasma homocysteine concentrations, Concentrations during treatment with the highest dose of orange juice. The results showed no significant relation between the 2 indexes when all data were included. A slight significant inverse correlation was shown after exclusion from the analysis of 2 subjects with unusually high plasma concentrations however, values after the exclusion were still insignificant.

Effect of orange juice consumption on cholesterol level

A sum of 50 hypercholesterolemic subjects was enrolled in this setup in order to analyze the effect of orange juice consumption on cholesterol level with a mean of 250.22 ± 18.5. All of the enrolled patients used to intake 500 ml orange juices daily. Their total cholesterol level was checked after each week and at the time of wash out (Table 1). It was observed from the obtained data that level of total cholesterol increased successively in each period (257.23 ± 17.9 for 1st, 259.23 ± 17.9 for 2nd and 263.07 ± 17.31) while at the time of washout period level of total cholesterol find to be same as in baseline (Figure 1).

Variables	Initial	1	2	3	Washout
Cholesterol	250.22 ± 18.5	257.23 ± 17.9	259.23 ± 17.9	263.07 ± 17.31	251.02 ± 18.4
Tri-Glycerides	228.26 ± 17.5	228.16 ± 17.5	240.3 ± 17.7	234.5 ± 17.8	262.8 ± 18.3
HDL	41.08 ± 10.08	41.2 ± 10.08	44.6 ± 10.08	47.9 ± 11.3	51.3 ± 12.6
LDL	158.42 ± 13.04	164.2 ± 13.11	159.11 ± 13.21	151.21 ± 13.24	148.22 ± 14.2
VLDL	41.82 ± 9.7	48.06 ± 10.7	41.78 ± 9.7	45.05 ± 9.87	41.91 ± 9.7
Changes in HDL %	----	5	8	19	24
Vitamin C µg/L	9.3 ± 3.4	21.3 ± 4.93	29.3 ± 12.2	37.8 ± 17.3	17.8 ± 7.5
Folate µg/L	35.7 ± 10.2	37.9 ± 12.7	41.6 ± 13.5	45.4 ± 16.7	39.7 ± 10.8

Table 1: Effect of Orange Juice consumption on lipid profile parameter of hypercholesterolemic subjects.

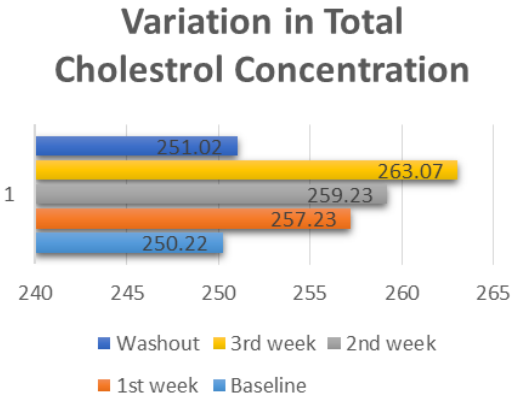

Figure 1: Effect of orange juice consumption on cholesterol level.

Effect of orange juice consumption on tri glyceride concentration level

A sum of total 50 subjects with elevated concentration of total plasma cholesterol concentration were enrolled in this setup in order to analyze the effect of orange juice consumption on Triglyceride Concentration with a mean of 228.26 ± 17.5. All of the enrolled patients used to consume 500 ml orange juice daily. Their Triglyceride level was checked after each week and at the time of wash out (Table 1). It was observed from the obtained data that level of Triglyceride concentration increased successively in each period (228.16 ± 17.5 for 1st, 240.3 ± 17.7 for 2nd and 234.5 ± 17.8) it was also observed at the time of washout period level of Triglyceride was elevated (262.08 ±18.3) as compares to initial baseline level (Figure 2).

Effect of orange juice consumption on HDL concentration

A sum of total 50 subjects with elevated concentration of total plasma cholesterol concentration were enrolled in this setup in order to analyze the effect of orange juice consumption on HDL Concentration with a mean of 41.08 ± 10.08. All of the enrolled patients used to consume 500 ml orange juice daily. Their HDL level was checked after each week and at the time of wash out (Table 1). It was observed from the obtained data that level of HDL concentration increased successively in each period (41.2 ± 10.08 for 1st, 44.6 ± 10.08 for 2nd and 47.9 ± 11.3) while at the time of washout period level of HDL was elevated (51.3 ± 12.6) as compares to initial baseline level (Figure 3).

Effect of orange juice consumption on LDL Concentration

A sum of total 50 subjects with elevated concentration of total plasma cholesterol concentration were enrolled in this setup in order to analyze the effect of orange juice consumption on LDL Concentration with a mean of 158.42 ± 13.04. All of the enrolled patients used to consume 500ml orange juice daily. Their LDL level was checked after each week and at the time of wash out (Table 1). It was observed from the obtained data that level of LDL concentration decreased successively in each period (164.2 ± 13.11 for 1st, 159.11 ± 13.21 for 2nd and 151.21 ± 13.24) while at the time of washout period level of LDL was decreased (148.22 ±14.2) as compares to initial baseline level (Figure 4).

Effect of orange juice consumption on VLDL concentration

A sum of total 50 subjects with elevated concentration of total plasma cholesterol concentration were enrolled in this setup in order to analyze the effect of orange juice consumption on VLDL Concentration with a mean of 41.82 ± 9.7. All of the enrolled patients used to consume 500ml orange juice daily. Their VLDL level was checked after each week and at the time of wash out (Table 1). It was observed from the obtained data that level of LDL concentration decreased successively in each period (48.06 ± 10.7 for 1st, 41.78 ± 9.7 for 2nd and 45.05 ± 9.87)

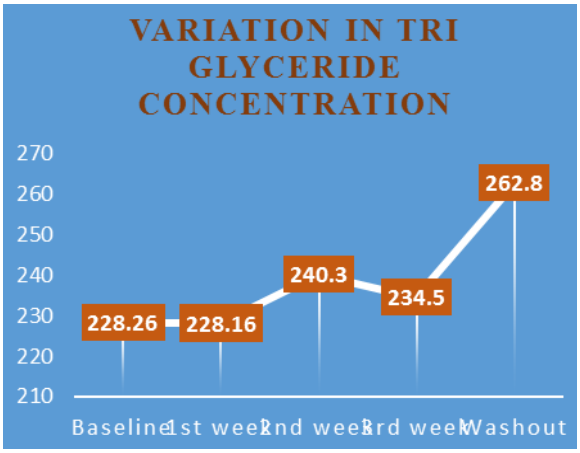

Figure 2: Effect of orange juice consumption on Triglyceride concentration level.

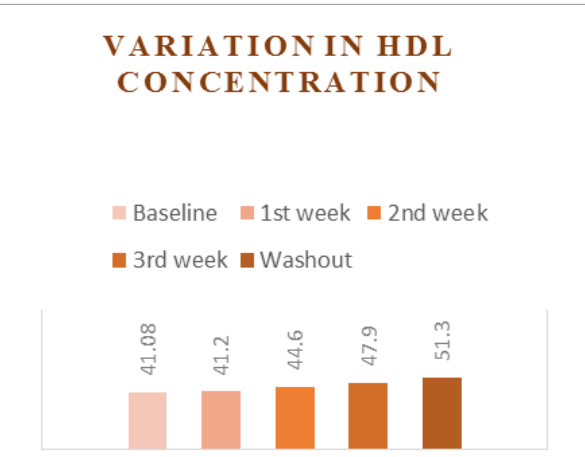

Figure 3: Effect of orange juice consumption on HDL Concentration.

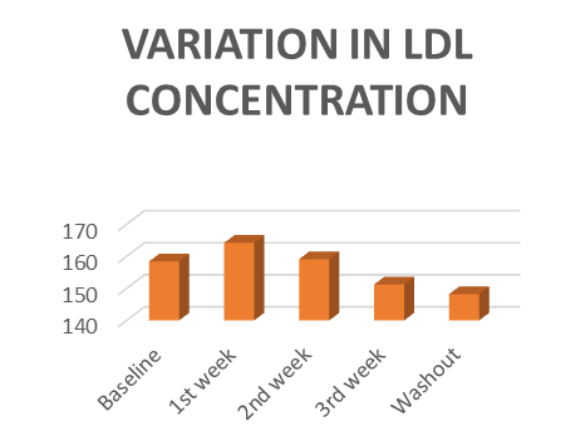

Figure 4: Effect of orange juice consumption on LDL Concentration.

while at the time of washout period level of VLDL was almost same (41.91 ± 9.7) as detected in initial baseline level (Figure 5).

Orange juice: An enhancer of HDL

A sum of total 50 subjects with elevated concentration of total plasma cholesterol concentration were enrolled in this setup in order to analyze the effect of orange juice consumption on HDL Concentration. All of the enrolled patients used to consume 500ml orange juice daily. Their HDL level was checked after each week and at the time of wash out (Table 1). It was observed from the obtained data that HDL concentration increased successively in each period (5% in 1st, 8% for 2nd and 19%) while at the time of washout period level of HDL was significantly elevated (24%) as compared to initial baseline value (Figure 6).

Effect of orange juice consumption on Vitamin C concentration

A sum of total 50 subjects with elevated concentration of total plasma cholesterol concentration were enrolled in this setup in order to analyze the effect of orange juice consumption on vitamin-C Concentration. All of the enrolled patients used to consume 500 ml orange juice daily. Their vitamin-C level with a mean of 9.3 ± 3.4 was checked after each week and at the time of wash out (Table 1). It was observed from the obtained data that vitamin-C concentration increased successively in each period (21.3 ± 4.93 in 1st, 29.3 ± 12.2 for 2nd and 37.8 ± 17.3) while at the time of washout period level of vitamin-C was significantly elevated (17.8 ± 7.5) as compared to initial baseline value (Figure 7).

Effect of orange juice consumption on Folate Concentration

A sum of total 50 subjects with elevated concentration of total plasma cholesterol concentration was enrolled in this setup in order to analyze the effect of orange juice consumption on folate Concentration. All of the enrolled patients used to consume 500 ml orange juice daily. Their folate level with a mean of 35.7± 10.2 was checked after each week and at the time of wash out (Table 1). It was observed from the obtained data that folate concentration increased successively in each period (37.9 ± 12.7 in 1st, 41.6 ± 13.5for 2nd and 45.4 ± 16.7) while at the time of washout period level of folate was significantly elevated (39.7 ± 10.8) as compared to initial baseline value (Figure 8).

Discussion

This study showed, in a group of subjects consisting mainly of individuals with mild-to-moderate hypercholesterolemia, that intake of 500 mL (2 cups) but not of 750 or 250 Ml orange juice/d for 4 week improved the plasma lipoprotein profile by significantly increasing HDL-cholesterol concentrations and by reducing the LDL-HDL cholesterol ratio. The reduction in the LDL-HDL cholesterol ratio observed during treatment with the highest dose of orange juice was entirely due to changes in HDL cholesterol concentrations. This

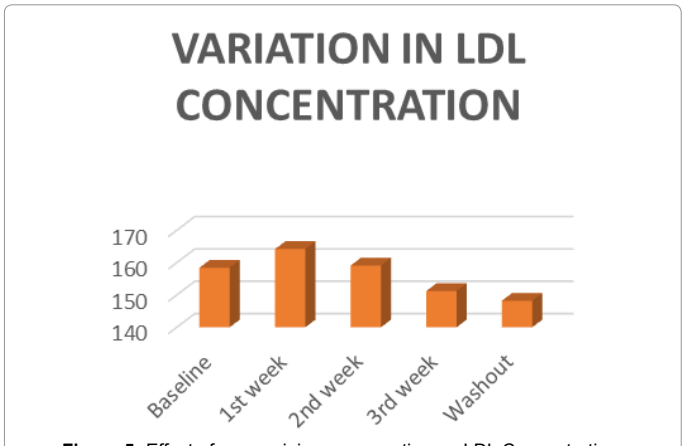

Figure 5: Effect of orange juice consumption on LDL Concentration.

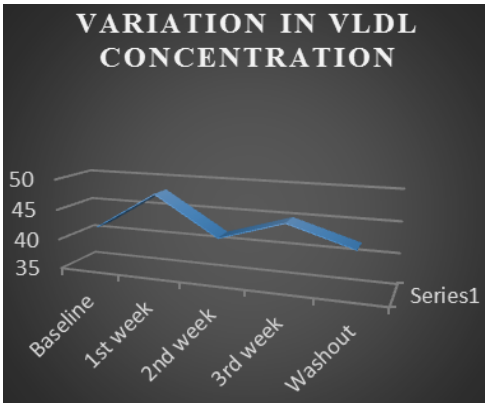

Figure 6: Effect of orange juice consumption on VLDL Concentration.

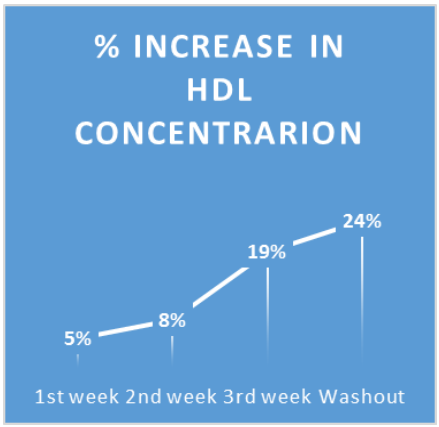

Figure 7: Increase in HDL Concentration.

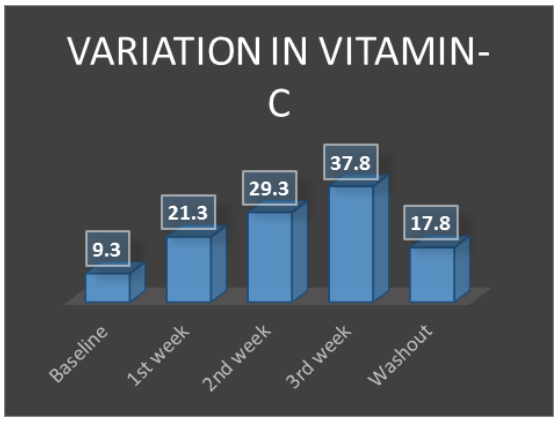

Figure 8: Effect of orange juice consumption on Vitamin C Concentration.

observation contrasts with a substantial reduction in LDL cholesterol induced in orange juice–fed, hypercholesterolemic rabbits [12], with a cholesterol lowering effect of citrus flavonoids observed in rats [6] and with the lack of changes in plasma lipids in normo-cholesterolemic, young men consuming unspecified doses of fresh orange juice for 2 months (Figure 9).

The disagreement between our data in humans and the results in animals could be due to the lower amount of the Juice or its minor components consumed by participants of the study than by rabbits or rats. Differences in responses between Human trials could be due to the fact that some of the participant shad hypercholesterolemia initially and some did not. The observed during the treatment with 250 mL orange juice/d suggest that the beneficial alterations occurred mostly in HDL, a subclass of HDL containing greater proportions of cholesterol but lower proportions of apo A-I than another major HDL subclass, HDL [13]. This response could provide an additional cardio protective effect because previous studies reported a reduction in HDL but no changes in HDL in individuals with coronary heart disease [14].

Significant elevations in plasma triacylglycerol produced in response to treatment with 250 mL orange juice did not exceed the normal range and may not be clinically significant or result in increased cardiovascular risk. Similar changes were also found in hypercholesterolemic rabbits [12] but not in normocholesterolemic men given orange juice. The effect was most likely not due to fructose and sucrose, which are abundant in orange juice, because increases in plasma triacylglycerol induced by these sugars in human trials were associated with a decline rather than an increase in HDL cholesterol [13] and because plasma triacylglycerols were not elevated in rabbits fed grapefruit juice, which consumed only 23% less of both sugars than animals given orange juice [12]. Likewise, previous studies do not suggest that increases in plasma triacylglycerols are due to intake of hesperidin. In rats, serum triacylglycerols were not altered by consumption of a 0.1%-hesperidin diet [6] and actually decreased after consumption of a 10%-hesperidin diet, presumably because of the inhibition of lipase [14]. The 18% increase in plasma folate concentration induced by treatment with the highest dose of orange juice (providing 188.4 g folate/d) was relatively moderate. After similar 4-5 wk periods, more pronounced increases in plasma folate were reported for individuals with coronary heart disease supplemented with 127 µg folic acid/d [15], for healthy subjects who consumed a vegetable- and citrus-rich diet enriched with 350 µg folate/d [16], and for women who consumed 250 µg folic acid/d [7].

However, other studies showed that in young women, a dose of

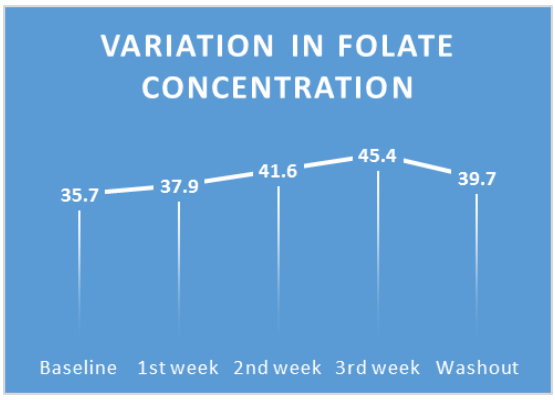

Figure 9: Effect of orange juice consumption on Folate Concentration.

400 µg folic acid/d increased plasma folate concentrations only after 8 week of consumption and not after 4 week [17]. Treatment with 500 mL orange juice/d did not decrease plasma homocysteine, although its lowest concentrations were generally found in subjects with the highest plasma folate concentrations. The lack of association may have been due to insufficient increases in plasma folate or to a relatively low dose of folate in the juice. A suggested, folate intakes need to be=200 µg/d to produce decreases in plasma homocysteine [18].

The residual effect of the orange juice intervention on plasma HDL cholesterol and on the LDL-HDL cholesterol ratio observed during the washout period was unlikely due to continued consumption of large amounts of juice by some subjects, as confirmed by a lack of correlation between plasma vitamin C and HDL-cholesterol concentrations during the washout. The tendency toward more pronounced responses observed for plasma HDL cholesterol and for the LDL-HDL cholesterol ratio during the washout period (24% and-17%,respectively) than during period 3 (21% and-16%, respectively) suggests a long-term effect of the juice components, possibly flavonoids, on hepatic lipoprotein metabolism. This could not be verified *in vivo* because after oral administration of orange juice, plasma hesperidin concentrations are below accurate detection limits [19]. However, in our previous study in HepG2 cells, the apo B-lowering effect of citrus flavonoids was partly reversible, implying a long-term effect of these compounds or their metabolites on lipoprotein metabolism in the liver [20].

A significant negative correlation between the baseline LDL HDL cholesterol ratio and the reduction in this ratio caused by the intake of 750 mL orange juice/d indicates that individuals with the highest initial LDL-HDL cholesterol ratio are most likely to experience a reduction in this ratio by consuming orange juice. A similar association was found in hypercholesterolemic subjects after consumption of a diet enriched with soybean products [21]. Our results do not imply that consumption of large quantities of orange juice (20% of daily energy) should be recommended to hypercholesterolemic individuals. According to the US food guide pyramid, a healthy diet includes =5-10 servings of fruit and vegetables daily as well as adequate quantities of fiber. Thus, cardioprotective nutrients in amounts equivalent to those in 500 mL orange juice should be provided from a combination of different foods [21-25].

Conclusion

Epidemiologic data from the present research revealed that a more consumption of vegetable and fruits allied with a less susceptibility of Acute Heart Disease. Orange and grapefruit juice are good source of folate, Vitamin C and flavanones, but their effect on heart patients has not been studied scrupulously. The advantageous outcome may be allied with inconsequential moieties, particularly flavanones, anticipated to apply their deed by restrain oxidation of LDL and thrombosis and vitamins E, C and carotene which act chiefly as antioxidants. The foremost element citrus hesperidin, flavonoids naringenin from orange and grapefruits are somewhat analogous to genistein, from soybean thought to be hypocholesterolemic [26].

Orange juice and energy intake at initial, after each nutritional level and after the washout period derived from the diet records of the enrolled subjects. The consumption of calcium, fatty acids, protein total energy and cholesterol were not significantly variant in each of the period. Fiber consumption declined gradually during the study and was significantly lower than initial baseline after the washout period. The propensity for the consumption of dietary decreased during the 1,

2, 3 and washout period may be due to successive increase in orange juice intake throughout study period.

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