Original Research Article

Role of serum Vitamin-C and lipid profile in cardiovascular diseases patients of Southern Rajasthan

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ABSTRACT

Background: Vitamin C's functions as an antioxidant and as an enzyme cofactor are well established, but the ways in which these functions may modify chronic disease risk are less well established. The belief that vitamin C may benefit heart health has stemmed from multiple pieces of evidence and lines of reasoning. Thus, we took the objective of this study to evaluate the correlations of serum concentrations of trace elements including vitamin-C with lipid profile parameters of adult men in Southern Rajasthan.

Methods: Present study was conducted in Cardiology and Biochemistry Department in GMCH, Udaipur during the year 2019. A total of 150 MI cases/controls were selected. Blood sample was collected by vein puncture using an aseptic technique. The blood was allowed to settle at 37°C and then centrifuged at 3000 rpm for 15 min. Serum was then separated and stored at -20°C until analysis. Vitamin C by NATELSON method on colorimeter.

Results: The study cases had significantly higher lipid values than those of controls (p<0.001). In cases the vitamin C levels were significantly low than the controls. There was significant difference in lipid levels among smokers and nonsmokers. When vitamin C levels were compared there was no statistically significant difference in their levels. There was no significant correlation between serum vitamin C levels and Lipids.

Conclusions: The physical characteristics such as diet, residence, smokers, non-smokers, showed significant difference in the controls and cases. Women seemed to have higher proportion of lipid abnormality than men. It is concluded from this study that lower levels of ascorbic acid definitely increase the risk of cardiovascular disease and inclusion of ascorbic acid in the normal diet will control the level of lipids.

Keywords: Ascorbic acid, Lipid profile, Lipoproteins, Trace elements

INTRODUCTION

Despite all the methods of diagnosis and the new strategies in the treatment of patients, cardiovascular diseases especially coronary heart disease (CHD) are considered as the most important cause of mortality in India over the last decade. Hyperlipidemia is one of the major risk factors of cardiovascular diseases (CVD), which can be modified either by proper lifestyle changes, medical management or by the combination of both. Therefore, study of lipid profile in the general population is important in society. In all the Asian countries, there is a concomitant rise in the level of serum total cholesterol (TC), and with it a rise in CVD. Serum TC levels are also higher in the urban compared with the rural population. A number of known risk factors have been recognised for this disease, but less known factors such as trace elements may have a role in the progress of atherosclerosis. The serum level of copper (Cu) and oxidants like free radicals have also been considered in coronary artery disease (CAD).
The studies on experimental animals (guinea pigs, monkeys, fish) have confirmed the important role of ascorbic acid deficiency in the development of hypercholesterolemia and atherosclerosis, but the clinical experience is not quite uniform. Meta-analyses of randomized controlled trials performed on subjects without established vitamin C-deficiency concluded that the evidence of the presence or absence of benefits derived from the ability of ascorbic acid to prevent cardiovascular diseases is not sufficient. Therefore the proof of atherogenic effect of chronic vitamin C deficiency is limited to indirect evidence only.3

Of particular interest is the potential role of vitamin C in heart health. Vitamin C’s functions as an antioxidant and as an enzyme cofactor are well established, but the ways in which these functions may modify chronic disease risk are less well established. The belief that vitamin C may benefit heart health has stemmed from multiple pieces of evidence and lines of reasoning. Many works has documented the beneficial effects of fruit and vegetable consumption on heart health, which has led to the hypothesis that, among other nutrients, vitamin C may be partially responsible for this relationship. Importantly, the link between fruit and vegetable consumption and improved cardiovascular outcomes has primarily been established through cohort studies, as randomized controlled trials in this area are scarce. Of note, green leafy vegetables and vitamin C-rich fruits contributed most to the apparent protective effect of total fruit and vegetable intake.8

Decade earlier in the developing country like India as compared to same age group of patients in the developed countries. Vitamin C and copper play an important role in cardiovascular diseases. Till date we have found very few studies with serum lipid profile and vitamin C in association with cardiac in southern part of Rajasthan.

Authors conducted this study to determine levels of ascorbic acid and lipid profile in cardiovascular disease patients.

METHODS

The present study was conducted in Cardiology Department and Biochemistry Department in Geetanjali Medical College, Udaipur during the period Jan 2019 to Aug 2019 fulfilling inclusion criteria till sample size of 75 is achieved.

Inclusion criteria

A total of 150 MI cases/controls were selected attending cardiac department of Geetanjali Medical College and Hospital, Udaipur, Rajasthan.

Group I: It consisted of healthy adult of age 25-70 yrs (control subjects) n=75. By routine examination and tests, we ensured that all the subjects were healthy and there are no signs and symptoms or history of MI and other chronic diseases.

Group II: It consisted of MI patients of age 25-70 yrs subjects (n=75).

Patients in both groups with age, height, weight and body mass index were matched. Other factors affecting trace elements levels (eating disorders, trace elements medication, alcoholism, smoking, kidney disease, type 2 diabetes mellitus, liver disease, pancreas disease) were excluded by history.

Intervention / assessment

Informed consent was obtained from all subjects participating in the study. Blood sample was collected by vein puncture using an aseptic technique. Serum was separated from the sample and was analyzed for following biochemical parameters:

- Serum copper was estimated by BR-PAESA method on semi autoanalyzer.9
- Lipid profile on Cobas-6000 fully automate analyzer.10
- Vitamin C by NATELSON method on colorimeter.11

About 10 mL of blood was obtained from each patient in both groups. The blood was allowed to settle at 37°C and then centrifuged at 3000 rpm for 15 min. Serum was then separated and stored at -20°C until analysis.

Statistical analysis

The data of all the subjects (control and the subjects) were statistically calculated and compared and their level of significance and the correlation coefficient was calculated. Summary statistics was done by proportions, mean, median, and standard deviation. The inferential statistics were done by, Student’s ‘t’ test and Pearson correlation. All measurements were analysed using SPSS version 16.0. ‘p’ value <0.05 was considered as statistically significant.

RESULTS

There were no significant differences between the two groups for patients according to mean age, dietary habits, residence, smoking habit and socio-economic status (Table 1). Biochemical lipid profile characteristics (TC, TG, LDL-C, HDL-C and LDL-C/HDL-C ratio) and vitamin - C in study and control cases.

There is statistically highly significant difference between mean lipid profile characteristics of controls and cases. The study cases have significantly higher lipid values than those of controls. The vitamin C levels are significantly low in cases as compared to controls. This
shows reduced vitamin C levels in cases (Table 2). Females had significantly higher lipid values than males. Vitamin C levels were almost similar in males and females (p>0.05) (Table 3).

Table 1: Descriptive physical characteristics of controls and cases.

| Physical characteristics | Cases (Mean±SD yrs) | Controls | p value |
|--------------------------|---------------------|----------|---------|
| Age                      | 54.15 ±13.35        | 50.29 ±16.10 | 0.11 (NS) |
| Diet                     | Non vegetarian 42.00 | 38.00 | 0.51 (NS) |
|                          | Vegetarian 33.00    | 37.00 | 0.78 (NS) |
| According to area         | Rural 28.00         | 30.00 | 0.61 (NS) |
|                          | Urban 47.00         | 45.00 | 0.04* |
| Smoking habit             | Non smokers 49.00   | 46.00 | 0.04* |
|                          | Smokers 26.00       | 29.00 | 0.04* |
| Socio Economic Status (SES) | High 26.00       | 38.00 | 0.04* |
|                          | Middle 49.00        | 37.00 | 0.04* |

*Significant; NS- Non significant

Table 2: Descriptive biochemical characteristics in controls and cases.

| Biochemical characteristics | Cases (n=75) | Controls (n=75) | P value |
|-----------------------------|-------------|----------------|---------|
| Cholesterol (mg/dl)         | 251.50      | 169.21         | <0.001** |
| Triglycerides (mg/dl)       | 213.32      | 138.27         | <0.001** |
| HDL (mg/dl)                 | 61.19       | 46.03          | <0.001** |
| LDL (mg/dl)                 | 137.54      | 92.31          | <0.001** |
| VLDL (mg/dl)                | 60.40       | 28.09          | <0.001** |
| LDL-C/HDL-C (mg/dl)         | 2.78        | 3.87           | <0.001** |
| Vitamin-C (mg/dl)           | 0.53        | 1.20           | <0.001** |

**Highly Significant

Table 3: Descriptive biochemical characteristics across genders in cases.

| Biochemical characteristics | Males (n=49) | Females (n=26) | P value |
|-----------------------------|-------------|----------------|---------|
| Cholesterol (mg/dl)         | 236.05      | 274.44         | <0.001* |
| Triglycerides (mg/dl)       | 211.16      | 217.39         | 0.05*   |
| HDL (mg/dl)                 | 52.62       | 68.16          | <0.001* |
| LDL (mg/dl)                 | 135.28      | 146.78         | 0.036*  |
| VLDL (mg/dl)                | 57.28       | 66.26          | 0.121   |
| LDL-C/HDL-C (mg/dl)         | 2.57        | 2.15           | 0.257   |
| Vitamin-C (mg/dl)           | 0.51        | 0.56           | 0.145   |

*Significant; **Highly significant

Table 4: Descriptive biochemical characteristics with smoking habits in cases.

| Biochemical characteristics | Smokers (n=26) | Non Smokers (n=49) | P value |
|-----------------------------|---------------|--------------------|---------|
| Cholesterol (mg/dl)         | 257.96        | 239.35             | 0.045*  |
| Triglycerides (mg/dl)       | 232.47        | 204.93             | <0.001* |
| HDL (mg/dl)                 | 55.15         | 62.27              | 0.031*  |
| LDL (mg/dl)                 | 134.00        | 144.21             | 0.039*  |
| VLDL (mg/dl)                | 59.99         | 61.16              | 0.844   |
| LDL-C/HDL-C (mg/dl)         | 2.30          | 2.35               | 0.885   |
| Vitamin-C (mg/dl)           | 0.53          | 0.52               | 0.775   |

*Significant; **Highly significant
Table 5: Descriptive biochemical characteristics with diet in cases.

| Biochemical characteristics | Non Vegetarian (n=42) | Vegetarian (n=33) | p value |
|-----------------------------|-----------------------|-------------------|---------|
| Cholesterol (mg/dl)         | Mean 259.87 SD 32.16  | Mean 241.29 SD 38.95 | 0.030* |
| Triglycerides (mg/dl)       | Mean 219.28 SD 23.52  | Mean 205.92 SD 27.27 | 0.028* |
| HDL (mg/dl)                 | Mean 61.88 SD 12.89   | Mean 68.65 SD 13.72 | 0.031* |
| LDL (mg/dl)                 | Mean 138.94 SD 15.82  | Mean 135.75 SD 15.36 | 0.381  |
| VLDL (mg/dl)                | Mean 56.47 SD 14.29   | Mean 65.39 SD 13.43 | 0.007* |
| LDL-C/HDL-C                 | Mean 2.98 SD 1.45     | Mean 2.51 SD 1.34   | 0.150  |
| Vitamin-C (mg/dl)           | Mean 0.56 SD 0.15     | Mean 0.49 SD 0.13   | 0.034* |

*Significant; **Highly significant

Table 6: Descriptive biochemical characteristics according to area in cases.

| Biochemical characteristics | Rural (n=28) | Urban (n=47) | p value |
|-----------------------------|-------------|--------------|---------|
| Cholesterol (mg/dl)         | Mean 238.47 SD 40.11 | Mean 266.59 SD 46.77 | 0.007* |
| Triglycerides (mg/dl)       | Mean 219.83 SD 25.28 | Mean 202.40 SD 23.79 | 0.004* |
| HDL (mg/dl)                 | Mean 64.39 SD 13.71 | Mean 54.19 SD 12.53 | 0.002* |
| LDL (mg/dl)                 | Mean 131.28 SD 12.08 | Mean 139.48 SD 11.21 | 0.005* |
| VLDL (mg/dl)                | Mean 57.92 SD 23.20 | Mean 64.55 SD 25.60 | 0.253  |
| LDL-C/HDL-C                 | Mean 2.91 SD 1.45   | Mean 2.56 SD 1.35   | 0.303  |
| Vitamin-C (mg/dl)           | Mean 0.56 SD 0.15   | Mean 0.49 SD 0.14   | 0.049* |

*Significant; **Highly significant

Figure 1: The correlations of serum concentrations of vitamin-c with lipid profile parameters in cases.

There was statistically significant difference in lipid levels among smokers and nonsmokers among TC (257.96±36.41 v/s 239±39.79), TG (232.47±15.11 v/s 204.93±15.40), HDL (55.15±13.77 v/s 62.27±12.60) and LDL levels (134.00±20.61 v/s 144.21±18.73). When vitamin C levels were compared there was no statistically significant difference in their levels (Table 4). There was significant difference in lipid levels (TC 259.87 V/s...
TC, TG, HDL and LDL showed statistically significant difference in rural and urban according to area in cases.

Vitamin C levels (0.56 V/s 0.49) shows statistically significant difference (p<0.05). (Table 6).

In present study the results indicate that in cases (study) group there was no significant correlation between serum vitamin C levels and TC (r=-0.020, p>0.05), TG (r=-0.07, p>0.05), HDL (r=0.105, p>0.05), VLDL levels (r=0.02, p>0.05) and LDL-C/HDL-C (r=0.05, p>0.05) (Figure 1).

DISCUSSION

This study was conducted on 150 subjects, divided into two groups (a) normal control subjects (75) and (b) cases (75), who visited cardiology department for the treatment/ diagnosis/ management of cardiovascular disease at Geetanjali Medical College and hospital, Udaipur and their blood samples were evaluated in the central laboratory of Biochemistry department. The data so obtained was analysed and is being compared with other studies. The mean age of case patients was 54.15±13.35 yrs and of controls was 50.29 ±16.10 yrs. There were 42 (56%) non vegetarians and 33(44%) vegetarians in cases as compared to 38(50.67%) non vegetarians and 37(49.33%) vegetarians in controls. There were 28(37.33%) rural and 47(62.67%) urban in cases and 30(40%) rural and 45(60%) urban in controls. There were 49(65.33%) nonsmokers and 26(34.67%) smokers in cases and 46(61.33%) nonsmokers and 29(38.67%) smokers in controls and 26(34.67%) high SES and 49(65.33%) middle SES in cases and 38(50.67%) high SES and 37(49.33%) middle SES in control group patients. In our study there were no significant differences between the two groups for the above characteristics (Table 1).

There was statistically highly significant difference between mean lipid profile characteristics of controls and cases. The study cases have significantly higher lipid values than those of controls (Table 2). Similar to the study that of Zhao et al, in which lipid abnormality was more prevalent in nonelderly patients (74% vs. 67%, P<0.05).12

The proportion of double- abnormality in nonelderly patients was significantly higher than that in elderly patients (27% vs. 21%, P<0.05), whereas no difference was seen. When vitamin C levels were compared between males and females, males had non-significant difference than females (Table 3). Findings similar to those of present study shows gender differences in lipid/lipoprotein metabolism reported by Russo et al, Kolovou et al, Wang et al, and others.13-15 Similarly in our study when the biochemical characteristics across genders in control patients were compared there was no statistically significant difference in lipid levels, both had equally higher levels of lipids. Whereas when vitamin C levels were compared between males and females, males had significantly lower levels than females.

When the biochemical characteristics were studied across smoking habits in cases, there was statistically significant difference in lipid levels among smokers and nonsmokers among TC (257.96±36.41 v/s 239±39.79), TG (232.47±15.11 v/s 204.93±15.40), HDL (55.15±17.37 v/s 62.27±12.60) and LDL levels (134.00±20.61 v/s 144.21±18.73). When vitamin C levels were compared there was no statistically significant difference. Jain RB et al, (2018) evaluated the impact of smoking on the adjusted and unadjusted concentrations of low-density lipoprotein (LDL) cholesterol, high-density lipoprotein (HDL) cholesterol, total cholesterol (TC), and triglycerides (TG).16 Adjusted levels of LDL and TC did not vary among smokers and nonsmokers. Smokers had lower adjusted levels of HDL than nonsmokers (48.8 vs. 51.4 mg/dl, p<0.01) and higher adjusted levels of TG (124.4 vs. 111.9 mg/dl, p < 0.01) than nonsmokers.

In our study the biochemical characteristics across diet in normal patients revealed no difference in lipid, vitamin C levels. There was statistically significant difference in lipid levels among cases non vegetarians and vegetarians (TC 259.87 V/s 241.29), TG (219.28 V/s 205.92), HDL (68.65 V/s 61.88), VLDL (56.47 V/s 65.39). Vitamin C also showed significantly higher values (0.56) in vegetarians than non-vegetarians (0.49) (Table 5).

Biochemical characteristics compared according to area in controls showed TG and HDL levels having statistically significant difference in rural and urban populations. Vitamin C levels were significantly higher in rural than urban (Table 6). TC (238.47 V/s 266.59), TG (219.83 V/s 202.40), HDL (64.39 V/s 54.19) and LDL (131.28 V/s 139.48) showed statistically significant difference in rural and urban according to area in cases. Vitamin C levels (0.56 V/s 0.49) shows statistically significant difference (p<0.05). Potential explanations for the urban-rural differences in blood lipids include differences in socioeconomic status, diet as well as occupational activities.

People living in certain urban areas are often referred to as slums-poses grave health risks due to the poor living conditions in such neighborhoods and may negatively impact individuals’ lifestyles.19

In addition, urban areas, in general, are characterized by a relatively high availability of fast food outlets and are
conducive to the adoption of more western diets, rich in salt, sugar and saturated fat, potentially contributing to the unfavourable blood lipids observed. Another possible explanation is that in urban areas, occupations often involve office work that generally requires less physical activity as compared with labour in rural, agricultural settings.\(^9\) Also, less heterogeneity might exist between urban and rural areas at the level of occupation-related physical activity, food availability and dietary habits and social-economic status. Another potential limitation is that there is no generally accepted definition of urban and rural.\(^9\)

The correlation of serum concentrations of trace element vitamin C included in the study with serum concentrations of lipid profile parameters (TC, TG, LDL-C, HDL-C and LDL-C/ HDL-C ratio) of hyperlipidemic patients (Figure 1) showed non-significant changes. In our study the relationship between trace element vitamin-C and lipid parameters didn't show any significant correlation between them, i.e. the vitamin C levels couldn't be used to predict the lipid concentrations in cardiovascular disease patients. Authors couldn't find studies on this angle of the study.

**CONCLUSION**

The physical characteristics such as diet, residence, smokers, nonsmokers, showed significant difference in the controls and cases. Women seemed to have higher proportion of lipid abnormality than men. It is concluded from this study that lower levels of ascorbic acid definitely increase the risk of cardiovascular disease and inclusion of ascorbic acid in the normal diet will control the level of lipids.

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