TAKING NUTRITIONAL SUPPLEMENTS FOR THREE MONTHS REDUCED BLOOD PRESSURE BUT NOT BLOOD LIPID LEVELS IN STUDENTS

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not significantly affect the blood and saliva chemistry. (*J Chiropr Med* 2006;5:53-59)

Key Indexing Terms: Dietary Supplements; Chiropractic; Blood Pressure

ABSTRACT

Objective: To study the effects of a nutritional supplement on cardiovascular health determined by blood and saliva biochemistry and heart rate variability (HRV) in asymptomatic college students and faculty members.

Methods: Forty subjects were recruited in the study via a school wide email notification and through personal contacts. GreensFirst vegetable supplement drink was tested to document its effect on the blood and saliva biochemistry in relation to cardiovascular health.

Results: After taking the supplement for 90 days, both the systolic and diastolic blood pressure decreased significantly. The heart rate decreased but did not reach statistical significance. Time domain analysis of HRV showed a slight decrease in standard deviation of normal-normal and square root of the mean squared differences but did not reach statistical significance. Frequency analysis of HRV found no significant changes. Saliva dehydroepiandrosterone sulfate (DHEA-S) and Cortisol decreased after taking the supplement, however, they did not reach significant levels. Blood glucose concentration was increased slightly but the change was not significant. Blood serum tests showed no significant changes in the total cholesterol or the low-density lipoprotein levels. However, it was noted that the high-density lipoprotein level was decreased significantly. Triglycerides did not show any significant changes but it was slightly elevated after the treatment period. Homocysteine increased significantly after taking the supplement for 90 days.

Conclusion: For this group of subjects, taking the green vegetable drink for 90 days reduced blood pressure but did

INTRODUCTION

Taking nutritional supplements and engaging in physical activity are considered beneficial to human health.^{1,2} Nutritional counseling has become an integral part of chiropractic practice in recent years. Healthy People 2010,² a nationwide health promotion and disease prevention program, has set goals for dietary counseling: 75% of physician office visits made by patients with cardiovascular disease (CVD), diabetes, or dyslipidemia will include counseling or education related to diet and nutrition by the year 2010. A study on the use of supplements in women showed that nearly 60% of US women took at least one supplement in 2000. Women who were obese or overweight and women who had not had contact with a health professional in the past 12 months were less likely to use supplements.³ Children often have limited understanding of the benefits of green vegetables.⁴

Many studies have investigated CVD risk factors, including hypertension, serum cholesterol, high-density lipoprotein, low-density lipoprotein, body mass index, body weight, dietary fat, energy intake, dietary fiber and exercise.⁵⁻¹¹ It has been reported that eating fruits and vegetables, particularly dark green leafy vegetables, appears to have a protective effect against coronary heart disease,¹² stroke,¹³ and other conditions.¹⁴⁻¹⁷ In a study with male US health professionals 40 to 75 years of age, taking dietary folate, vitamin B-6 and B-12 reduced peripheral arterial diseases (PAD).¹⁸ In this study, all

subjects answered a detailed 131-item questionnaire to assess diet and vitamin supplement use. The study population consisted of 46036 men free of PAD, CVD and diabetes at baseline. The subjects were followed for 12 years, during which time they documented 308 incident PAD cases. This study supports the notion that nutritional supplement improves cardiovascular health.¹⁸

Green leafy vegetables are a rich source of a number of micronutrients and other phytochemicals having antioxidant properties.¹⁹ It was reported that consumption of green leafy vegetables in the raw state had significantly higher values of antioxidant than the cooked state ($P < 0.001$).¹⁹ However, it may not be convenient to shop for fresh vegetables every day for daily consumption. This may contribute to the widely accepted method of improving cardiovascular health by taking green vegetable supplements. GreensFirst is a supplement and is one of the many products that can be found in the market of nutritional supplements. GreensFirst contains fruits and vegetables in powdered form. It has the antioxidant power of over 10 servings of fruits and vegetables. It is fortified with Beta-Glucan, a USDA patented oat fiber, that lowers cholesterol. This supplement contains 49 different super foods, extracts and concentrates, including super greens, vegetables, fruits, probiotics, soluble fiber, insoluble fiber, herbs, spices, natural flavonoids, enzymes and lecithin. The mechanism of the beneficial effects of taking a green vegetable supplement is not well established. It was proposed that green vegetable supplement might provide antagonists on the aryl hydrocarbon receptor.²⁰

Taking natural supplements rarely causes side effects. The most common side effects include swelling of the throat and shortness of breath.²¹ It is debated whether these effects are allergic reactions, effects of chemical properties or contaminants, or just coincidences.²¹

In this study, a specific type of supplement drink was tested to document its effect on the cardiovascular system. The specific aims of the study were to understand the effectiveness of the GreensFirst supplements on: 1) blood lipids and cholesterol; 2) saliva cortisol and; 3) blood pressure and heart rate variability (HRV). The hypotheses are: 1) that the supplements are effective in reducing blood cholesterol and lipids that cause CVD; 2) reducing saliva

cortisol, and; 3) reducing blood pressure and increasing HRV.

METHODS

Subjects

Forty students and faculty members were recruited for the study via a school wide email notification and through personal contacts. All study procedures were approved by the institutional review board at Logan College of Chiropractic and explained to each subject prior to testing. All subjects were required to sign a written informed consent before participating in the study.

Inclusion Criteria

Male or female asymptomatic students and faculty members of any age were qualified for the study.

Exclusion Criteria

Individuals with diabetes, heart, kidney, thyroid disorders, neurological diseases, and chronic disease were not recruited in the study. Female subjects who were pregnant or breast feeding were excluded and those using drugs or medications were not permitted to participate in the study. Subjects already taking similar supplements or those that did not sign the informed consent form were not accepted for the study.

Role of Subjects and Treatment

All subjects were given a 90-day supply of the nutritional supplement free of charge. The supplement was supplied by GreensFirst (Doctors for Nutrition, Solana Beach, CA, USA). All subjects answered a pre-screening questionnaire for qualifications. This was to ensure each subject met the inclusion and exclusion criteria. The study began with a baseline test of blood pressure, HRV, blood serum (high-density lipoproteins [HDL], low-density lipoproteins [LDL], total cholesterol, triglycerides, homocysteine), and saliva chemistry (DHEA-S, cortisol). All biochemical tests were performed in a medical lab. Blood and saliva samples were collected at the college research department by a registered nurse. All tests were performed prior to initially taking the supplement and all tests were repeated after the 90-day study period.

Instrumentation

Blood pressure: An Omron automatic blood pressure monitor (Omron, Vernon Hills, IL, USA) was used in the blood pressure measurements before and after the study period.

Blood serum: Blood serum tests were conducted before and after the study period, and included the following (testing procedure and apparatus in parentheses):

- HDL (modified enzymatic, Roche, Hitachi Modular)
- LDL (Friedewald Formula [$LDL = Chol - HDL - [Trig/5]$])
- Total cholesterol (enzymatic, Roche, Hitachi Modular)
- Triglycerides (enzymatic [glycerol phosphate oxidase], Roche, Hitachi Modular)
- Homocysteine (fluorescence polarization immunoassay, Abbott, AxSYM)

Saliva chemistry: Cortisol and DHEA-S were assessed by immunoassay (RIA).

All blood and saliva samples were collected at the college and shipped overnight to Pharmasan Labs (Osceola, WI, USA) for analysis by Drs. Gottfried Kellerman and Mike Bull. All test results were mailed back to the college research department for final data analysis.

HRV: Biocom's Active ECG Scanner (Biocom Technologies, Poughkeepsie, NY) was used in the HRV data collection with digital signal processing software. The heart scanner records an electrocardiograph signal, computing the instantaneous changes of HRV after each recording session. The scanner connects to the subjects through 3 small electrodes that attached to the left arm, right arm and left leg. A baseline HRV was recorded before treatment was given. The HRV recording was taken in the sitting position for 5 minutes for all subjects.

Data Treatment and Analysis

A Student t-test was used to assess the differences of blood and saliva biochemistry and the blood pressure and HRV changes before and after taking the supplement for 90-days. Significance was determined at $p < 0.05$. SPSS 11.5 (SPSS, Inc, Chicago, IL,

USA) statistical software was used for the data analysis.

RESULTS

Forty subjects (19 male) were recruited and completed the study. The average age of the subjects was 27 ± 5 years old (range 21–48). The study population consisted of 2 faculty members and 38 students currently enrolled at the college.

After taking the supplement for 90 days, both the systolic and diastolic blood pressure decreased significantly from 127 ± 13 to 123 ± 14 mmHg (systolic) and 82 ± 10 to 77 ± 10 mmHg (diastolic) with $p < 0.05$ (Fig 1). The heart rate did not show significant changes from 71.98 ± 11.9 to 71.81 ± 11.89 (Fig 2). Time domain analysis of HRV showed an increase in standard deviation of normal-normal (44.0 ± 150.0 to 64.8 ± 56.9 ms) but a decrease in square root of the mean squared differences (55.6 ± 36.0 to 56.2 ± 28.8 ms) but both did not reach statistical significance ($p > 0.05$) (Fig 3). Frequency analysis of HRV found a lower total power (1636 ± 1547 to 1561 ± 1253) and a slightly higher high frequency (315 ± 334 to 319 ± 359) but both did not reach a significant level ($p > 0.05$). Changes in very low frequency (592 ± 590 to 516 ± 465) and low frequency (761 ± 809 to 725 ± 661) were not significant ($p > 0.05$) (Fig 4). Decreases in saliva DHEA-S (436 ± 338 to 350 ± 291 Pg/ml) and cortisol (2.43 ± 2.61 to 2.15 ± 1.39 ng/ml) were present after taking the supplement, however, they did not reach any significant levels ($p > 0.05$) (Fig 5 and 6). Blood glucose concentration was decreased slightly (102 ± 28 to 101 ± 25 mg/dL) but the change was

Figure 1. Blood pressure readings before and after taking supplement, showing a statistically significant change. ($p < 0.01$ for systolic and diastolic).

Figure 2. Heart rate averages before and after taking the supplement were not statistically significant ($p > 0.05$).

Figure 3. Time domain analysis of HRV (SDNN = standard deviation of normal-normal; RMS-SD = square root of the mean squared differences; for all $P > 0.05$).

Figure 4. Frequency analysis of HRV (VLF = very low frequency; LF = low frequency; HF = high frequency; for all $p > 0.05$).

not significant ($p > 0.05$) (Fig 7). Blood serum tests showed a slight decrease in the total cholesterol level from 183 ± 38 to 180 ± 39 mg/dL ($p > 0.05$). The LDL level was slightly elevated from 104.92 ± 32.2 to 106.25 ± 33.38 mg/dL ($p > 0.05$) (Fig 8 and 9). However, it was noted that the HDL level decreased significantly from 65.32 ± 98.69 to

Figure 5. Changes in DHEA-S were not significant ($p > 0.05$).

Figure 6. Changes in salivary cortisol levels were not significant ($p > 0.05$).

46.64 ± 12.14 mg/dL ($p < 0.05$) (Fig 9). Triglycerides did not show any significant changes but were slightly elevated after the treatment period (135.61 ± 96.25 to 137.4 ± 94.99 mg/dL). Homocysteine increased significantly after taking the supplement for 90 days (7.32 ± 2.34 to 8.43 ± 2.39 uMol/L) (Fig 10).

DISCUSSION

The research question of whether the vegetable supplement has any physiological effects on the cardiovascular system as determined by blood pressure, HRV, saliva and blood chemistries resulted in mixed responses.

The favorable response was indicated by the decrease in systolic and diastolic blood pressure after taking the supplement for 90 days. It is known that hypertension can be caused by atherosclerosis, imbalances in the rennin-angiotension system, and

Figure 7. Changes in blood glucose concentration were not significant ($p > 0.05$).

Figure 8. Changes in total cholesterol were not significant ($p > 0.05$).

Figure 9. Changes in HDLs (significant decrease, $p < 0.05$), LDLs (insignificant increase, $p > 0.05$) and total triglycerides (insignificant increase, $p > 0.05$).

hyperinsulinemia. Hypertension is the leading cause for heart disease, kidney disease and stroke.²² Studies have found that vegetable products in vegetarian-diet trials reduced blood pressure in normotensive and hypertensive individuals.^{22,23} In a large randomized controlled study, diet and blood pressure were evaluated in subjects consuming vegetables and fruits daily for 8 weeks. The experimental group showed a significant reduction in blood pressure. In addition, the hypertensive subjects had

Figure 10. Homocysteine blood serum levels increased significantly ($p < 0.05$) after taking the supplement for 90 days.

greater reductions in blood pressure than did the normotensive subjects.²⁴ In the current study, the subjects were normotensive individuals and the results reflected a small but significant reduction of blood pressure, which is consistent with the literature. This reduced blood pressure was also consistent with reduced heart rate over the 3-month period. However, the decrease in heart rate was not statistically significant. Decreased heart rate is a beneficial response to the cardiovascular system. It has been known for years that reduced heart rate increases cardiac reserve, reduces cardiac work-load and reduces heart attacks.²⁵

The HRV analysis showed an increase in the total power but it was not statistically significant after taking the supplement. It was hypothesized that the HRV would increase with consumption of the supplement; however, it was not observed in the study. An increased total power was a sign of increased HRV, which was beneficial to the cardiovascular system. Increased total power reduces the chance of heart attacks in people with cardiac diseases.²⁶ The lack of a significant increase in the HRV suggests that the supplement might not exert effects on the autonomic nervous system under the current conditions. There are many factors that could affect the outcome of the HRV, such as exercise, diet, and emotional states during the period in which the HRV was recorded. Another important factor in the HRV analysis is the duration of the study period. It was reasoned that a study of longer duration, such as greater than 6 months of treatment, might produce significant changes in HRV. A treatment period of longer duration is needed to change the physiological set point of the autonomic nervous system.²⁵

Research studies have demonstrated that risk factors for coronary artery disease include: elevated serum

total cholesterol, LDL-cholesterol, and triacylglycerol concentrations, as well as reduced HDL-cholesterol concentrations.^{27,28} Treatment of hypercholesterolemia has focused on increasing fecal excretion of cholesterol and bile acids and reducing hepatic cholesterol synthesis through diet modification and use of pharmacologic agents. The hypocholesterolemic effects of vegetables and fruit and their constituents have been studied in recent years.^{27,28}

Despite the reported positive findings of the vegetable supplement on the blood lipids, this study did not find any significant changes in the lipids. In fact, it was surprising to see a slight increase in total cholesterol, LDL and triglycerides, though they were not statistically significant. Homocysteine level increased to such a high level, which was unexpected. The reason for the increase was not clear. It was speculated by the researchers that the reason for not seeing a decrease in blood lipids was the life style of the student population in the study. Most students in the study did not perform daily exercise and they spent a majority of their time sitting in the classroom and studying in the evening. This may have contributed to the slight increase in the blood lipid levels. It was suggested by the researchers that further investigations should be carried out in a more diverse population under more strict controls. A control group would help eliminate some of the mixed responses. Perhaps for further studies, a population that initially has increased lipid levels may serve as a more reliable sample. Another limiting factor is the fluctuations in the blood chemistry and blood pressure measurement that could affect the results of the study. If funding and resources are available, repeated measurements at more intervals could improve the reliability of the study.

CONCLUSIONS

After taking the GreensFirst supplement for 90 days, a decrease in both the systolic and diastolic blood pressures was observed in the 40 asymptomatic subjects. There were no significant changes in heart rate and total power in the HRV analysis. No significant changes were found in the blood cholesterol, LDL and Triglycerides levels.

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