

# The effect of a garlic preparation on plasma lipid levels in moderately hypercholesterolemic adults

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## Abstract

Lipid management is well established as an effective preventive and management tool for cardiovascular disease (CVD). Health claims regarding the cholesterol lowering benefits of garlic are widespread. However, the clinical trial data are inconsistent. The effect of two doses of a commercial garlic preparation on plasma lipids were evaluated, compared to a placebo, in moderately hypercholesterolemic adults (baseline low density lipoprotein cholesterol (LDL-C) =  $157.4 \pm 18.7$ , mean  $\pm$  S.D.). Fifty-one adults, aged  $51.8 \pm 8.3$  years participated in a double-blind, placebo-controlled, parallel treatment trial conducted in an outpatient research clinic. They were randomized to a placebo or a garlic botanical blend providing 500 or 1000 mg dehydrated garlic powder/day (three groups, 16–18 subjects per group). Plasma lipids were assessed every 2 weeks for 12 weeks. The study was designed with sufficient power to detect a 10% relative decline in LDL-C. The absolute mean changes in LDL-C over 12 weeks were  $0.0 \pm 4.3$ ,  $+1.4 \pm 4.8$ , and  $-10.1 \pm 6.8$  mg/dl for the placebo, half-dose and full-dose, respectively. In the full-dose group, the LDL-C decrease of 6.1% was not significantly different from the other groups ( $P = 0.5$ ). No significant differences were observed for total- or high-density lipoprotein cholesterol (HDL-C), or triacylglycerol levels. In conclusion, the garlic powder preparation used in this study among moderately hypercholesterolemic adults did not significantly effect plasma lipids levels. There was no indication of a graded affect by garlic dose over the range of 0, 500 and 1000 mg/day. A small ( $< 10\%$ ) effect on LDL-C levels or a threshold effect requiring larger doses are not eliminated by this study. © 2001 Elsevier Science Ireland Ltd. All rights reserved.

**Keywords:** Cholesterol; Garlic supplements; Hyperlipidemia; Allicin; Low density lipoprotein cholesterol; Clinical trial

## 1. Introduction

Management of plasma cholesterol levels continues to be a cardinal issue in cardiovascular disease (CVD) prevention. Hygienic and pharmacological approaches have been established for lipid management [1]. A growing interest in complementary and alternative medicine [2,3] has led to an increasing number of non-pharmacological therapies for lipid management. Among these is garlic, which in 1997 was one of the top herbal remedies in consumer demand. That year garlic supplements accounted for sales of \$200 million dollars, up 33% from 1995 sales [4]. Garlic preparations have

been reported to have numerous prophylactic and remedial benefits, including decreasing plasma cholesterol levels and blood pressure, enhancing immune function, decreasing platelet aggregation and blood coagulation, protecting low density lipoprotein (LDL) particles from oxidation, and improving vasodilatation [5,6].

A number of clinical trials have examined the role of garlic in lipid management. Although several reviews and at least two meta-analyses have been conducted [7–10], there remains a lack of consensus [11,12]. While many clinical trials have reported a hypocholesterolemic effect of garlic [13–17], others have found no detectable lipid response [18–22]. The lack of concordance may be due to the substantial heterogeneity in the design and conduct of these trials. Many of the garlic trials reported have been limited or flawed. For example, in a meta-analysis conducted by Warshafsky

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et al. [9], of the 28 identified trials, 21 were excluded for having inadequate study designs, or including a substantial proportion of subjects with normal cholesterol levels. Another possible reason for the inconsistency has been the type of garlic preparation used, which has included fresh garlic [23,24], steam-distilled garlic oil [18], aged garlic extract [14], and dehydrated powdered garlic [15,20,22]. Possible confounders in these studies were often unreported, including study adherence level or changes in weight, diet or exercise. Other important differences between these trials include the dosage, duration and characteristics of subjects studied. Although widely promoted commercially for its cholesterol-lowering ability, the benefit of garlic supplementation in lipid management remains uncertain.

A randomized, double-blind, placebo-controlled, parallel treatment trial of the effects of a garlic preparation was conducted on plasma lipids in a group of moderately hypercholesterolemic adults (LDL-cholesterol [LDL-C] 130–190 mg/dl). Study compliance, weight, diet and exercise were monitored throughout the study to insure their consistency. Plasma lipids were measured every 2 weeks to examine the possible time-dependent nature of any effects. The purpose of this study was to determine if 12 weeks of taking a garlic preparation would lower plasma LDL-C levels in moderately hypercholesterolemic adults, and if so, to examine a possible dose response using 0, 500 and 1000 mg/day of powdered garlic. Blood pressure was examined as a secondary outcome measure.

## 2. Subjects and methods

### 2.1. Sample

Participants were recruited from the general public and from Stanford University employees, primarily through newspaper advertisements. Men and women who were 30–65 years of age, with fasting plasma LDL-C 130–190 mg/dl, fasting plasma triacylglycerol < 300 mg/dl, and a body mass index (BMI) between 19 and 31 kg/m<sup>2</sup> were invited to enroll. Exclusions were pregnant women, smokers, prevalent heart disease, diabetes, or use of lipid or blood pressure lowering medications within the past month. Following a telephone screening interview, 158 were eligible for

cholesterol testing; of these, 60 were found to have LDL-C and triacylglycerol levels in the eligibility range. Of those eligible, seven chose not to participate and 53 were randomly assigned to the three groups. This investigation was reviewed and approved by the Stanford University Human Subjects Committee. All participants signed an informed consent form prior to enrollment, and the study was carried out according to the guidelines laid down in the Declaration of Helsinki.

### 2.2. Study design

The study was a randomized, double-blind, placebo-controlled, 12-week parallel treatment, clinical trial. Participant enrollment was from June to October, 1997. Participants were scheduled for nine clinic visits following random assignment to one of three treatment groups (Fig. 1).

### 2.3. Materials and garlic preparations

Garlic and placebo tablets were provided by a commercial garlic tablet manufacturer in sealed bottles that each contained a 2-week supply (42 tablets/container). The garlic was prepared from the 1997 harvest of garlic grown in California's Central Valley and processed by proprietary dehydration processes. Each full-dose garlic tablet contained 333 mg garlic powder, and the half-dose tablets contained 167 mg. The 3 tablets/day of the full dose were equivalent to approximately 1.5 cloves of fresh garlic as determined by the manufacturer. Tablet consumption was recommended three times daily with meals. Garlic was the primary component in this botanical blend, which also included extracts of *Fabaceae* and *Brassicaceae* as minor ingredients. The garlic manufacturer certifies that the material provided 1500 ppm allicin yield per 1000 milligrams of garlic powder (i.e. 1500 µg allicin yield/day in the full dose). Both the garlic and placebo tablets were created with the same dextrose and cellulose tablet matrix and were prepared according to Good Manufacturing Practices. There were no plant-based materials in the placebo formula. The coated tablets were formulated to disintegrate in warm water in less than 30 min. All tablets were formulated to be identical in appearance by the use of a colored coating and produced from the same lots of raw materials. In addition, each bottle of tablets included a cotton-rayon filler infused with a drop of garlic oil that imparted a strong odor to disguise the identity of the tablet composition. Each set of six bottles for an individual participant was labeled with a unique three-digit code number to identify its contents. These randomly selected code numbers were generated by the tablet manufacturer using the RAND function from Microsoft® Excel (1997) and provided in a non-sequential list. Upon enrollment, each new participant

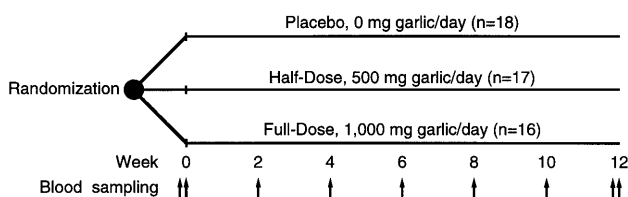


Fig. 1. Study design.

was assigned the next three-digit number on the list. Participants, study staff, laboratory technicians and investigators were blinded to the treatment assignment until the protocol and laboratory work was completed.

#### 2.4. Data collection

At every clinic visit, weight and blood pressure were recorded and  $\geq 12$ -h fasting blood samples were collected by venipuncture in EDTA tubes. Study adherence was monitored by tablet count. Every 2 weeks during clinic visits the study tablet bottles were turned in, the number of tablets that had not been consumed in the 2-week period were recorded, and a new bottle with a 2-week supply was dispensed. In addition, participants were asked to report their level of adherence in an end-of-study questionnaire.

Participants were advised to maintain their weight, as well as their usual diet and activity levels for the study's duration. When weight deviated by more than 2 kg from baseline, participants were counseled to alter their caloric intake accordingly. Food intake was assessed at baseline, mid-study and end-study, using 3-day food records. No restrictions were placed on participants' intake of fresh garlic in order to avoid other dietary changes that may have limited the generalizability of the findings.

Exercise and non-formal exercise behaviors (physical activity behaviors) were assessed using a modified version of the Paffenbarger Physical Activity Questionnaire (1978) [25]. A physical activity score for the previous week was determined by quantifying and combining the number of hours of light to moderate and vigorous activity, in both work and leisure settings. The following examples of these activity levels were provided to participants: light to moderate activity included housework, light sports, regular walking, golf, yard work, lawn mowing, painting, repairing, light carpentry, dancing, or bicycling on level ground; vigorous activity included digging in the garden, strenuous sports, jogging, chopping wood, sustained swimming, brisk walking, heavy carpentry, or bicycling on hills. The questionnaires were administered at baseline, mid-study and at the end of study.

Effectiveness of the blinding and the tolerance of the study tablets were assessed at the end of the study by a self-administered questionnaire. Upon completion of the study, participants were asked if they knew to which of the three treatments they were assigned, or if they 'could not tell'. Participants were also asked to indicate whether they had experienced any of twelve symptoms during the study, and the frequency of each symptom (never, seldom, occasionally, often, frequently or always). The 12 symptoms were: alertness, energy, belching, body odor, stomach gas/bloating, intestinal gas/flatulence, constipation, diarrhea, indigestion/cramps, nausea/vomiting, drowsiness, and insomnia.

Fasting blood samples were collected by venipuncture in EDTA tubes, refrigerated immediately and centrifuged within two hours. Plasma samples were stored in Wheaton vials and frozen at  $-70^{\circ}\text{C}$  until a given participant had completed the protocol, at which time a set of samples for that participant were thawed and analyzed in a single batch to minimize laboratory variability. Plasma levels of total cholesterol and triacylglycerol (with subtraction of a free glycerol blank) were measured by enzymatic procedures using established methods of the Lipids Research Clinics (ABA 200 instrument, Abbott Laboratories) [26,27]. HDL-C was measured by dextran sulfate-magnesium precipitation [28] followed by enzymatic determination of cholesterol [26]. LDL-C was calculated according to the method of Friedewald [29]. The lipoprotein laboratory assays as monitored by the Lipid Standardization Program of the Centers for Disease Control and Prevention (Atlanta, GA), and the National Heart Lung and Blood Institute, were consistently within specified limits. The laboratory precision for measuring total cholesterol, HDL-C and triacylglycerol were assessed monthly and all coefficients of variation were  $< 2.8\%$ . Within individual variability of replicate lipid measurements taken at baseline (2 separate days) and end-study (2 separate days) were assessed as intraclass correlation coefficients and for total-C, LDL-C, HDL-C and triglycerides were determined to be 0.89, 0.81, 0.97 and 0.80, respectively.

Blood pressure was measured manually by sphygmomanometer on the right arm, three times while participants were seated after at least a 5-min rest. Three blood pressure readings were taken at approximately 2-min intervals and the average of the second and third readings were used.

#### 2.5. Statistical analyses

Statistical tests were performed using the SAS computer program (SAS Institute, Cary, NC). Descriptive statistics are presented for the baseline characteristics of the participants. Nutrient analyses were conducted using Food Processor, Version 7.0 (ESHA, Salem, OR). The baseline characteristics and study hypothesis were tested using analysis of variance (ANOVA) and two separate approaches. The first approach was an ANOCOVA test for between-group differences in pre-post changes (delta values) in plasma lipids or blood pressure using the average of two baseline and two end-study measurements, and adjusting for baseline levels as a covariate. A general linear model (GLM) was used to include pre-post changes in saturated fat and weight as additional covariates. A second approach was to use the time-series of nine assessments for each participant to calculate the slope of the response for total-C, LDL-C and HDL-C and triacylglycerols. These data were then used to test for between-group differ-

Table 1  
Baseline characteristics of study population (mean  $\pm$  S.D.)

	Supplementation group		
	Placebo	Half-dose	Full-dose
Males/females ( <i>n</i> )	9/9	7/10	10/6
Age (years)	51.6 $\pm$ 8.1	53.4 $\pm$ 8.9	50.2 $\pm$ 8.3
Weight (kg)	77.8 $\pm$ 10.2	76.9 $\pm$ 17.7	76.3 $\pm$ 12.1
Body mass index (kg/m <sup>2</sup> )	26.9 $\pm$ 2.8	26.0 $\pm$ 4.2	25.4 $\pm$ 3.6
<i>Dietary composition<sup>a</sup></i>			
Total energy/day (Kcal)	1960 $\pm$ 490	1840 $\pm$ 375	2075 $\pm$ 600
Total fat (% energy)	26.6 $\pm$ 8.6	26.4 $\pm$ 7.4	27.0 $\pm$ 7.5
Saturated fat (% energy)	8.5 $\pm$ 4.3	8.4 $\pm$ 3.1	8.1 $\pm$ 3.1
Dietary fiber (g/1000 Kcal)	11.3 $\pm$ 6.1	11.6 $\pm$ 3.7	11.9 $\pm$ 4.4
Alcohol (% energy)	2.2 $\pm$ 3.1	4.7 $\pm$ 4.8	2.6 $\pm$ 2.9
Physical activity score <sup>b</sup>	19.9 $\pm$ 15.7	25.0 $\pm$ 23.1	22.2 $\pm$ 18.2
<i>Outcome measures</i>			
Total-C (mg/dl)	230 $\pm$ 24	231 $\pm$ 21	235 $\pm$ 27
LDL-C (mg/dl)	158 $\pm$ 20	151 $\pm$ 18	163 $\pm$ 18
HDL-C (mg/dl)	46 $\pm$ 10	54 $\pm$ 18	46 $\pm$ 12
Triacylglycerol (mg/dl)	133 $\pm$ 48	125 $\pm$ 63	126 $\pm$ 56
<i>Blood pressure (mmHg)</i>			
Systolic	117.0 $\pm$ 11.9	115.0 $\pm$ 11.0	118.1 $\pm$ 14.5
Diastolic	75.8 $\pm$ 7.6	74.9 $\pm$ 7.8	79.1 $\pm$ 7.3

<sup>a</sup> Does not include participants with missing baseline or end-study food records (placebo *n* = 3, half-dose *n* = 1, full-dose *n* = 1).

<sup>b</sup> Includes combined hours per week of light to moderate and intense physical activity, at work or leisure (examples provided in methods).

None of the between-group differences were statistically significant at baseline.

completed nutrition study (unpublished) to predict the S.D. of pre-post change in LDL-C concentrations (16 mg/dl), the study was designed to have a power of 80% ( $1 - \beta$ ) to detect a 10% difference in 12-week LDL-C change between treatment arms ( $\alpha = 0.05$ ).

### 3. Results

Table 1 shows the three groups were similar at baseline in age, weight, diet composition, plasma lipids and blood pressure. Fifty-one participants completed the study. Two participants dropped out in the first week because of unforeseen scheduling conflicts, one each from the half-dose and full-dose groups.

Minimal side effects were reported. Of the 12 symptoms enumerated in the questionnaire, participants reported seldom to occasional incidences of belching, body odor, stomach gas, flatulence and constipation. Eleven, 47 and 24% of the placebo, half-dose and full dose group, respectively, reported at least one of these gastrointestinal symptoms. However, the symptoms were mild in severity and did not impair participation or study adherence. The blinding technique proved to be effective. The study tablets were indistinguishable by sight and smell. Twenty-eight percent of the placebo group, 27% of the half-dose and 0% of the full-dose group correctly guessed their assigned dose.

Weight, dietary composition, physical activity, and study adherence remained constant throughout the study. The differences between groups for these potential confounders were modest and not statistically significant. The average difference from baseline in weight was less than 1 kg for each group at each clinic visit, and at end-study the differences from baseline (mean  $\pm$  S.D.) were  $+0.4 \pm 1.6$ ,  $0.0 \pm 1.6$  and  $+0.8 \pm 1.6$  kg for placebo, half-dose and full-dose, respectively. The change from baseline in % energy from saturated fat was determined at both mid-study and end-study to be:  $+0.1 \pm 3.0$  and  $+0.8 \pm 3.6\%$  respectively, for placebo (*n* = 15 with complete data);  $+1.8 \pm 3.7$  and  $+2.6 \pm 3.1\%$  for half-dose (*n* = 16 with complete data); and  $+1.8 \pm 2.4$  and  $+1.4 \pm 3.5\%$  for full-dose (*n* = 14 with complete data). The change from baseline in mean activity scores (exercise plus physical activity) at mid-study and end-study was:  $-2.6 \pm 14.3$  and  $+2.3 \pm 8.4$  h/week, respectively, for placebo;  $-5.8 \pm 16.8$  and  $-5.3 \pm 12.0$  h/week for half-dose; and  $+0.5 \pm 11.0$  and  $+1.9 \pm 7.8$  h/week for full-dose (complete data for all but placebo with *n* = 1 missing). Study adherence as assessed separately by tablet count and by a self-administered questionnaire was  $\geq 92\%$  across all treatment groups.

Plasma lipid changes are presented in Fig. 2 as group averages, and the LDL-C change data from each individual participant are presented in Fig. 3. Mean

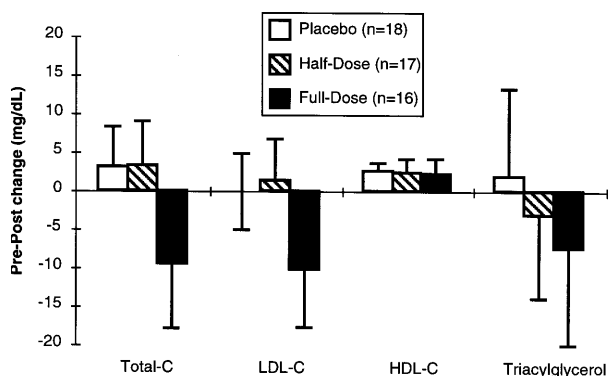


Fig. 2. Mean  $\pm$  S.E.M. 12-week changes in plasma lipids. Analysis of covariance detected no significant between-group differences.

ences in slopes in a GLM using ANCOVA with adjustment for baseline lipid level as a covariate. All statistical tests were two-tailed using a significance level ( $\alpha$ ) of 0.05. Using empirical data from a previously

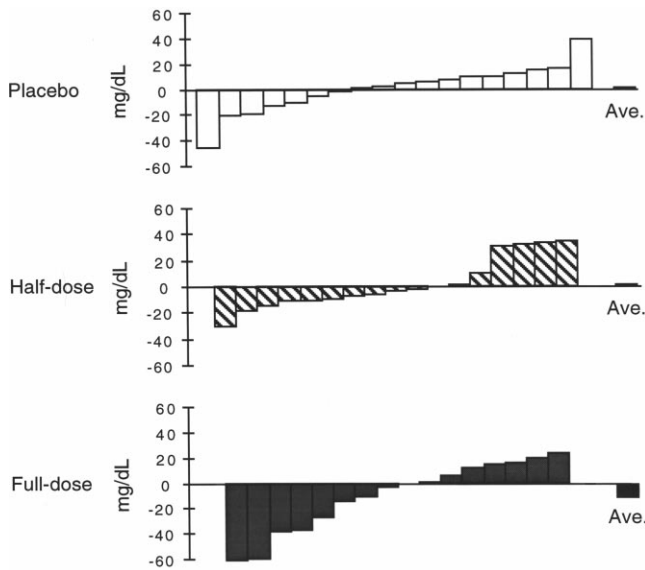


Fig. 3. Individual and mean 12-week changes in low density lipoprotein cholesterol (LDL-C) by treatment group.

changes in total-cholesterol were +1.4, +1.4 and –4.0% for the placebo, half-dose and full-dose groups, respectively. Corresponding changes in LDL-C levels were 0.0, +0.9 and –6.1%; changes in HDL-C levels were +5.6, +4.5 and +4.6%; and changes in triacylglycerol levels were +1.4, –2.5 and –5.9%. Although the full-dose group had the most favorable lipid changes in 12-weeks of intervention, none of the between-group differences was statistically significant.

In order to make the best use of the data from the nine blood samples for each participant, a second

statistical approach was used to test for differences in the slopes of the 12-week changes. Again, no statistically significant differences in lipid changes were detected. For placebo, half-dose and full-dose, respectively, the slopes (mg/dl per day) were: total-C,  $-0.04 \pm 0.39$ ,  $0.03 \pm 0.28$  and  $-0.11 \pm 0.32$ ; LDL-C,  $-0.02 \pm 0.27$ ,  $0.03 \pm 0.25$  and  $-0.07 \pm 0.41$ ; HDL-C,  $0.02 \pm 0.07$ ,  $0.03 \pm 0.06$  and  $0.03 \pm 0.08$ ; and triacylglycerols,  $-0.14 \pm 0.52$ ,  $0.03 \pm 0.44$  and  $-0.10 \pm 0.42$ . Results of the 0, 2, 4, 6, 8, 10 and 12-week plasma lipid assessments are presented in Table 2. There were no significant between-group differences in blood pressure during the study in this normotensive population (data not presented).

#### 4. Discussion

In this study, no significant difference in LDL-C response was detected after 12-weeks of daily intake of 1000 or 500 mg of a dehydrated garlic powder preparation versus placebo in a group of moderately hypercholesterolemic adults. There were also no significant pre-post differences observed between the three groups for total-C, HDL-C, triacylglycerols or blood pressure.

The finding of a non-significant LDL-C lowering effect lends itself to several possible interpretations. It is possible that garlic supplementation, at least in this form, has no effect on LDL-C. It is also possible that it has an effect smaller than this study was able to detect. The sample size was chosen to detect a 10% relative decline in LDL-C; in the full dose group only a 6%

Table 2  
Plasma lipid measures (mg/dl, mean  $\pm$  S.D.) over 12 weeks

	Base-line	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12	P-value <sup>a</sup>
<i>Total-C</i>								
Placebo	230 $\pm$ 24	226 $\pm$ 27	223 $\pm$ 19	227 $\pm$ 23	227 $\pm$ 26	228 $\pm$ 23	233 $\pm$ 28	0.3
Half-dose	231 $\pm$ 21	235 $\pm$ 27	232 $\pm$ 19	239 $\pm$ 23	233 $\pm$ 23	235 $\pm$ 26	234 $\pm$ 26	
Full-dose	235 $\pm$ 27	232 $\pm$ 29	232 $\pm$ 29	235 $\pm$ 18	230 $\pm$ 18	228 $\pm$ 24	225 $\pm$ 23	
<i>LDL-C</i>								
Placebo	158 $\pm$ 20	160 $\pm$ 21	149 $\pm$ 19	152 $\pm$ 17	153 $\pm$ 26	156 $\pm$ 19	158 $\pm$ 25	0.5
Half-dose	151 $\pm$ 18	153 $\pm$ 23	151 $\pm$ 15	158 $\pm$ 18	152 $\pm$ 18	149 $\pm$ 19	153 $\pm$ 20	
Full-dose	163 $\pm$ 18	160 $\pm$ 21	158 $\pm$ 12	159 $\pm$ 20	155 $\pm$ 17	154 $\pm$ 18	153 $\pm$ 22	
<i>HDL-C</i>								
Placebo	46 $\pm$ 10	45 $\pm$ 12	46 $\pm$ 12	46 $\pm$ 12	47 $\pm$ 12	47 $\pm$ 12	48 $\pm$ 12	0.9
Half-dose	54 $\pm$ 18	53 $\pm$ 16	54 $\pm$ 16	55 $\pm$ 17	55 $\pm$ 16	58 $\pm$ 15	56 $\pm$ 18	
Full-dose	46 $\pm$ 12	46 $\pm$ 12	46 $\pm$ 14	50 $\pm$ 13	51 $\pm$ 11	49 $\pm$ 14	49 $\pm$ 14	
<i>Triacylglycerol</i>								
Placebo	133 $\pm$ 48	158 $\pm$ 70	140 $\pm$ 57	145 $\pm$ 52	134 $\pm$ 49	126 $\pm$ 53	135 $\pm$ 39	0.7
Half-dose	125 $\pm$ 63	142 $\pm$ 72	131 $\pm$ 59	129 $\pm$ 69	132 $\pm$ 69	137 $\pm$ 69	122 $\pm$ 64	
Full-dose	126 $\pm$ 56	135 $\pm$ 65	122 $\pm$ 48	133 $\pm$ 71	118 $\pm$ 51	129 $\pm$ 58	119 $\pm$ 52	

<sup>a</sup> Between group differences in 12-week plasma lipid changes.

decline was seen. In addition, the S.D. of the change was larger than anticipated, contributing to the lack of statistical significance. Based on the changes and inter-individual variation seen in this study, the sample size in each treatment arm would need to be 75 to demonstrate statistical significance. The effect size of 10% was chosen to be significant in a clinical setting, so it is possible to conclude from this study that garlic in this form does not seem to produce a clinically important effect in individuals with LDL-C levels in the 130–190 mg/dl range. The lack of any apparent dose-response (i.e. no difference in the intermediate dose group) favors this null interpretation. On the other hand, if every 1% decrease in plasma cholesterol is associated with a 2–3% decrease in relative risk for coronary heart disease [1], then the absolute change in LDL-C reported here could have public health relevance at the population level.

The findings are consistent with existing literature. The 95% confidence intervals around the plasma lipid level changes in the full-dose treatment arm would include the main effects of most of the clinical trials that have been published to date. The conclusions from two meta-analyses of clinical trials were that plasma lipid lowering therapy with various forms of garlic supplementation resulted in a 9–12% reduction in plasma cholesterol [8,9]. The majority of published studies to date have used a powdered garlic preparation, similar to the preparation method used in this study. Considerable variability in outcomes exists between these studies. For example, Adler et al. [13], using a commercial dehydrated garlic tablet, reported a significant net drop of 13.1% in LDL-C levels relative to the placebo group in 12 weeks, and Jain et al. [15], using the same product and a similar design, reported a significant net decrease of 8% in LDL-C levels in moderately hypercholesterolemic adults. However, three other studies [19,20,22], using the same dosage of the same commercial dehydrated garlic powder product (Kwai<sup>®</sup>, Lichtwer Pharmaceuticals) reported no significant effect. The dose of powdered garlic tablets used in the five studies just cited, 900 mg/day, was similar to the full dose of 1000 mg/day used in this study. The allicin content of the tablets used in this study, 1500 µg/day in the full dose, was lower than the amount used in other studies with powdered garlic preparations. Other types of garlic preparations used in lipid lowering trials have included aged garlic extract and steamed garlic oil. Steiner et al. [14] used a large dose, 9 tablets/day, of aged-garlic extract, and reported a statistically significant 4.6% lowering of plasma LDL-C levels. In contrast, a recent study using steamed garlic oil supplementation reported no significant effect on cholesterol levels in hypercholesterolemic adults after 12 weeks [18]. One explanation could be that the oil is not as effective as dehydrated garlic powder because it con-

tains different sulfur-containing phytochemicals [30]. Some of the discrepancies reported in these studies can be explained by the heterogeneity that exists among them in terms of study design, duration, subject characteristics, adherence, or confounders such as weight, diet, and exercise. Taken as a whole, this current body of evidence suggests that the hypocholesterolemic effect of garlic intake, when taken in the form of supplementation, is marginal to negligible, similar to the findings here.

The possibility that a significant cholesterol-lowering effect of garlic supplementation was missed in the current study might be raised if there were strong mechanistic data to support this. Chi et al. suggested that garlic supplemented diets may decrease the activities of lipogenic enzymes and enhance the excretion of neutral steroids and bile acids as was observed in a rat study [31]. Other studies using rat hepatocytes have demonstrated that garlic supplemented diets resulted in a dose-dependent inhibition of cholesterol biosynthesis at different enzymatic steps [32–35]. Despite the reported inhibition in these studies, a limitation of *in vitro* and animal models is that the results may not be physiologically relevant *in vivo* in humans. The one clinical trial that addressed this in humans reported no effect of a steamed garlic oil preparation on cholesterol synthesis or on the excretion of neutral steroids [18]. Further work on a possible mechanistic link between garlic compounds and plasma lipid metabolism is warranted.

A recent review by Lawson [36] adds another important perspective and raises the possibility that a true hypocholesterolemic effect of garlic supplements could go undetected if the *in vivo* production of a cholesterol lowering agent in garlic was inefficient and inconsistent. It has been suggested, but not definitively established, that the cholesterol lowering effect of garlic is attributable to the bioavailable yield of allicin and its derivatives [36,37]. Allicin yield is related to the enzymatic conversion of alliin to allicin by alliinase. Lawson points out that the digestive process might inactivate alliinase before it is allowed to complete the conversion of alliin to allicin. It appears that standardization and documentation of both 'effective allicin yield' [36] and other phytochemical contents in future supplement studies is warranted.

There were several strengths of this investigation. The clinic visits every 2 weeks promoted study adherence and allowed for multiple blood sampling. The use of multiple serial blood samples added sensitivity to the plasma lipid analyses. The documentation of weight, diet and exercise stability, and the high study adherence suggest that consistent garlic tablet consumption was achieved and that this study was shielded from important confounding variables. The inclusion of a simultaneous placebo group in the trial further supports the adequacy of the trial for testing the hypothesis that

garlic supplementation lowers plasma LDL-C. Finally, to the authors' knowledge, the inclusion of a half-dose in the design was an approach that had not been used before, which allowed for the examination of possible intermediate dose or threshold effects, which were not observed.

This trial was also subject to several limitations. Garlic intake from food sources was not controlled during the study. Participants were asked to refrain from consuming large quantities of garlic, but were also told they should maintain their habitual diets and did not have to abstain completely from garlic in their food while on-study. It was felt that this approach made the results more generalizable, although differences in garlic intake between the study groups could have confounded the results. Random assignment and a low-drop out rate should have reduced this concern. Another limitation was the small sample size that had limited statistical power to detect a difference in the order of the magnitude that was found or to test for different effects in subsets of the study population. A larger sample size might have detected a statistical significance, but an LDL-C change of <10% in an efficacy trial is of borderline clinical significance. Another issue was the relatively low allicin content of the garlic preparation used in this study compared to other garlic supplementation trials, although the role of allicin in human lipoprotein metabolism remains equivocal. Finally, this trial focused on a single product, at two doses, in a specified population over a defined time-period of 12 weeks, and the effects on plasma lipids. This leaves many questions unaddressed regarding the effect of garlic intake, in various forms, in different dosages, in other subsets of the population, over longer time periods, on a wide range of other human health factors, such as blood pressure, immune function, blood coagulation, anti-oxidant status, vasodilatation and plaque formation [5,6,38].

#### 4.1. Implications

These data do not support the sole use of garlic preparations containing 1500 ppm allicin to substantially lower total-C, LDL-C, or triacylglycerol levels in moderately hypercholesterolemic adults. Results may differ by preparation, dose or individual characteristics not addressed in this study. Few studies with fresh garlic have been done because of the difficulties in blinding participants, but more of these studies may be warranted to determine whether the effects of fresh garlic are similar to those of supplements. It is also important to consider the food choices that accompany raw or cooked garlic intake. For example, vegetable fiber in a stir-fry containing garlic and plant-based foods, or saturated fat from butter on garlic bread would have opposite effects on plasma lipids, regardless

of the effect or lack of effect of the garlic. Finally, increased garlic intake, ingested in its various forms or methods of preparation, may confer other health outcomes or benefits not measured in this study.

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