Oral L-Arginine Improves Hemodynamic Responses to Stress and Reduces Plasma Homocysteine in Hypercholesterolemic Men[1,2]

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ABSTRACT When administered intravenously, L-arginine substantially reduces blood pressure (BP) and peripheral vascular resistance in healthy adults and in patients with vascular disease. Oral L-arginine has been shown to improve endothelial function; however, it is not clear whether oral administration has significant effects on systemic hemodynamics. In a randomized, placebo-controlled, crossover study we tested whether oral L-arginine (12 g/d for 3 wk) affected hemodynamics, glucose, insulin, or C-reactive protein in 16 middle-age men with hypercholesterolemia. After each treatment, hemodynamic variables were measured at rest and during 2 standardized stressor tasks (a simulated public-speaking task and the cold pressor). Regardless of treatment, the stressor tasks increased BP and heart rate ($P \leq 0.02$). Relative to placebo, L-arginine changed cardiac output (−0.4 L/m), diastolic BP (−1.9 mm Hg), pre-ejection period (+3.4 ms), and plasma homocysteine (−2.0 umol/L) ($P \leq 0.03$). The change in plasma L-arginine was inversely correlated with the change in plasma homocysteine ($r = -0.57$, $P = 0.03$). Contrary to the results of previous studies of L-arginine administered intravenously, oral administration did not affect total peripheral resistance or plasma insulin. Oral L-arginine also did not affect plasma glucose, C-reactive protein, or lipids. This pattern of findings is consistent with the hypothesis that oral L-arginine reduces BP. This study is the first to describe a hemodynamic mechanism for the hypotensive effect of oral L-arginine and the first to show substantial reductions in homocysteine with oral administration. J. Nutr. 135: 212–217, 2005.

KEY WORDS: • *L-arginine* • *peripheral resistance* • *hemodynamic* • *homocysteine* • *acute stress*

Protein consumption is associated with lower blood pressure (BP)[4] in both epidemiologic studies (1,2) and randomized controlled trials (3,4). L-Arginine may contribute to the hypotensive effects of protein because it is the substrate for the synthesis of nitric oxide (NO). Substantial improvements in endothelium-dependent vasodilation have been reported with L-arginine supplementation in humans (5) and animals (6,7). In addition, several clinical studies have shown significant reductions in BP and peripheral vascular resistance with i.v. L-arginine administration (8–11).

In contrast, substantial reductions in BP have been observed in some (12–14) but not all (5,15–20) studies of oral L-arginine treatment. For example, Siani et al. (12) reported significant reductions in systolic and diastolic BP (ranging from 5 to 7 mm Hg) when healthy subjects consumed 10–14 g/d of L-arginine in the form of a supplement or through the inclusion of arginine-rich foods in the diet. Therefore, increased intake of arginine, in the form of foods or supplements, may be beneficial to people with cardiovascular risk factors. However, in contrast to the more extensive literature on i.v. administration, no previous studies have examined the systemic hemodynamic effects of repeated daily consumption of oral L-arginine.

In this study, we measured changes in BP, cardiac output, and total peripheral resistance in response to a moderate dose of supplemental oral L-arginine (12 g/d for 3 wk) in 16 men with elevated cholesterol in a randomized, placebo-controlled, crossover experiment. We also tested whether increases in plasma L-arginine were associated with changes in circulating homocysteine, lipids, glucose, insulin, or C-reactive protein (CRP). Because of growing interest in the effects of NO on sympathetic nervous system function (21), we also examined L-arginine's effects on cardiovascular responses to acute stressors in the laboratory. This is important because endothelial dysfunction is associated with greater peripheral vascular constriction during acute stressors (22), and we have shown that individuals with exaggerated responses to an acute stress task were at higher risk of developing elevated blood pressure in a 10-y prospective study (23). The present study is the first to examine the effects of L-arginine on hemodynamic stress responses.

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[2] Supported by Kraft Foods Global, Inc., Glenview, Illinois, and by the General Clinical Research Center of The Pennsylvania State University (NIH M01RR10732).

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[4] Abbreviations used: ADMA, asymmetric dimethylarginine; BP, blood pressure; C, cholesterol; CRP, C-reactive protein; DBP, diastolic blood pressure; GI, gastrointestinal; MAP, mean arterial pressure; NO, nitric oxide; SBP, systolic blood pressure; TPR, total peripheral resistance.

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TABLE 1

Baseline characteristics of the participants[1]

Age, *y*	45 ± 1.9 (33–60)
BMI, *kg/m²*	28 ± 1.1 (23–38)
SBP, *mm Hg*	134 ± 3.3 (118–162)
DBP, *mm Hg*	87 ± 1.6 (78–100)
Heart rate, *bpm*	72 ± 2.4 (48–88)
Plasma metabolites	
Total-C, *mmol/L*	6.4 ± 0.2 (5.2–7.3)
LDL-C, *mmol/L*	4.3 ± 0.1 (3.5–5.0)
HDL-C, *mmol/L*	1.2 ± 0.1 (0.9–1.9)
Triglyceride, *mmol/L*	2.0 ± 0.3 (0.7–4.4)
Insulin, *pmol/L*	129.6 ± 17.3 (74.1–289.8)
Glucose, *mmol/L*	5.2 ± 0.1 (4.4–6.1)

[1] Values are least squares means ± SEM (range), $n = 16$.

SUBJECTS AND METHODS

Characteristics of this sample are presented in **Table 1**. Volunteers were recruited via newspaper advertisements and fliers posted in the community. Forty-three subjects met the criteria after a phone screening, and 18 of the men were deemed eligible after a blood draw and physical exam (see below). Of these, 16 completed the study (1 began taking a statin after enrollment, and 1 suffered gastrointestinal (GI) distress while on L-arginine). The study was approved by the Institutional Review Board of Pennsylvania State University, and written informed consent was obtained. All had total cholesterol (C) and LDL-C levels within the 50th to the 90th percentiles according to NHANES III criteria, and LDL and total-C were in the borderline high–high range according to the National Cholesterol Education Program (24). Exclusion criteria included using medications with cardiovascular effects; history of cardiovascular disease/hypertension; being a current smoker; BMI ≥ 38; or abnormal results on a clinical chemistry panel. Participants were instructed to postpone changes in diet or exercise until after the study. Vitamins and other supplements were discontinued 2 wk prior to randomization.

Supplement intervention. A randomized, double-blind, 2-period, crossover design was used. Treatments (3 wk each) were separated by a 1-wk compliance break. Treatment order was randomized (9 subjects were given L-arginine first) and blood samples were drawn after the men had fasted for 12 h at the end of each treatment. Participants were given L-arginine tablets and matched placebos (Jarrow Formulas). Tablets contained 1 g freebase L-arginine or 1 g microcrystalline cellulose. Participants were asked to take the tablets in 4 divided doses throughout the day, to break them in half, and to consume them with a meal or a full glass of liquid. L-Arginine intake was gradually increased during wk 1 of treatment (19) as follows: 4 g/d for 2 d, 8 g/d for 3 d, and 12 g/d thereafter. No participant reported GI distress after the run-in period was instituted. In keeping with Lerman et al. (19), supplements were taken with the evening meal the night before the fasting blood draw. Cardiovascular testing sessions were scheduled on the same day as the blood draws. On testing days, participants were asked to avoid caffeine, alcohol, and vigorous exercise, and they consumed 3 g L-arginine or placebo 2 h before each testing session.

Cardiovascular testing sessions. Laboratory sessions included these events in fixed order: resting baseline (30 min), speech stressor (5 min), recovery (20 min), cold pressor task (2.5 min), and a final recovery (20 min). Measurements were collected every other minute during rest/recovery periods and every minute during stress. As in our previous studies (25,26), task instructions were presented on videotape. During the speech task, participants prepared (2 min) and delivered a speech (3 min) on a hypothetical disagreement (e.g., being falsely accused of shoplifting). To limit habituation, the topic of the speech changed at the second visit. Participants were told that their speech would be videotaped and later judged for creativity, organization, and clarity. During the cold pressor task, participants placed one foot, up to the ankle, in 4°C water for 2.5 min. When standardized instructions are given, these tasks have good test-retest reliability (27).

Hemodynamic measures. Cardiac output (liters per minute) and stroke volume (milliliters per beat) were measured noninvasively via impedance cardiography (28,29). This technique records changes in thoracic impedance across the cardiac cycle via a tetrapolar band electrode system and a standard electrocardiogram, and it has been empirically validated for measuring within-subject change in hemodynamics (30,31). As described by Georgiades et al. (28), we applied 2 voltage electrodes (1 around the base of the neck and 1 around the middle of the chest). Current electrodes were applied parallel to the voltage electrodes. For each reading, a 40-s continuous sample of the impedance cardiogram was collected using a Minnesota Impedance Cardiograph (Model No. 304B, Instrumentation for Medicine). Signals were processed by the Cardiac Output Program (version 5.04, BIT), an empirically validated system (32) that uses the Kubicek equation 33 to estimate stroke volume. We have shown strong test-retest reliability for impedance-derived measurements using this protocol (34).

BP was measured with an automated device (Dinamap Pro 100 Monitor, GE Medical Systems). Mean arterial pressure (MAP) (mm Hg) is a time-weighted mean of systolic (SBP) and diastolic BP (DBP). MAP = DBP + [(SBP − DBP)/3]. The Cardiac Output Program calculates total peripheral resistance (TPR, dyne · s · cm^{-5}) using a conventional formula: TPR = (MAP/cardiac output) × 80.

Biochemical assays. Plasma lipids were assayed in duplicate by American Medical Laboratories. Triglycerides and total cholesterol concentrations were measured enzymatically. HDL-C was measured turbidimetrically and LDL-C was calculated with the Friedwald equation. Interassay CVs were as follows: total cholesterol (1.4%), HDL-C (4.0%), and triglyceride (1.4%); all were within CDC guidelines. Plasma insulin was tested using a human-specific insulin RIA (Linco Research). Plasma glucose was analyzed with a YSI STAT Plus 2300 glucose analyzer (Yellow Springs Instruments).

Plasma C-reactive protein concentrations were measured at the Cytokine Core Laboratory of the General Clinical Research Center by a competitive EIA assay protocol previously described (35). Plasma homocysteine was analyzed as part of a classical aminogram by ion-exchange chromatography (36). Plasma samples were reduced with fresh aqueous 0.8 mol/L dithiothreitol. Samples were deproteinized by the addition of fresh aqueous sulfosalicylic acid. After precipitation and centrifugation, the supernatant was removed, freeze-dried, and resuspended in lithium citrate buffer before filtration (0.22 μm). The filtrate was analyzed by ion-exchange chromatography with postcolumn ninhydrine detection (Amino-System 2500; Bio-Tek). Asymmetric dimethylarginine (ADMA) concentrations were determined using an IDE-approved ELISA assay (Cardiovasics Medical Science Laboratory) (37).

Data analysis. Variables were tested for normality, and treatment effects were examined using the mixed models procedure in SAS (v.8, SAS Institute). Models included treatment, period, and treatment order as fixed effects and subject as a random effect. For hemodynamic variables, means were calculated during each stressor and during the last 6 min of the baseline and recovery periods. Models for analysis of hemodynamic measures also included task (stress vs. rest) and the treatment × task interaction. C-reactive protein and triglyceride concentrations were natural-log transformed prior to analysis, and untransformed values are reported in the text and tables.

Significant effects ($P \leq 0.05$) were investigated with Tukey's post hoc test. Tables and figures depict least squares means ± SE unless otherwise noted. Interrelationships between the variables were estimated by Pearson correlations. Correlations were also conducted on change scores (change = level during L-arginine treatment − level during placebo treatment). Our previous work suggests that this study was adequately powered ($\beta = 0.90$) to detect a 15% change in total peripheral resistance (34).

RESULTS

Acute stress effects on hemodynamic variables. Relative to resting values, the speech task increased BP, heart rate, and cardiac output and reduced the pre-ejection period (main

effects of task period, $P \leq 0.0001$). The cold pressor increased BP and total peripheral resistance ($P \leq 0.001$). Diastolic BP and mean arterial pressure remained elevated during the post-task recovery periods.

L-Arginine effects on hemodynamic variables. Treatment-related changes in the hemodynamic measures were evident at rest and during stress, and there were no interactions between task and treatment (**Table 2**). Collapsing across the rest and stress periods, L-arginine was associated with reductions in diastolic BP (mean change = −1.9 mm Hg, $P = 0.007$), arterial pressure (−1.9 mm Hg, $P = 0.02$), and cardiac output (−0.4 L/min, $P = 0.03$). Stroke volume (−2.7 mL/beat, $P = 0.09$) and systolic BP (−2.7 mm Hg, $P = 0.07$) tended to be lower during L-arginine treatment, whereas the pre-ejection period was longer (+3.4 ms, $P = 0.03$). Contrary to our hypotheses, total peripheral resistance and heart rate were not altered by L-arginine.

L-Arginine effects on circulating amino acids, lipids, and C-reactive protein. Consumption of L-arginine was associated with a 69% increase in plasma L-arginine relative to the placebo period ($P < 0.0001$, **Table 3**). Ornithine increased by 68% and homocysteine decreased by 10% during L-arginine treatment ($P < 0.03$). L-Arginine did not affect any of the other amino acids (Table 3). Circulating glucose, insulin, total cholesterol, LDL-C, HDL-C, ADMA, and C-reactive protein concentrations were also unaffected (**Table 4**). Fasting triglyceride concentrations were 27% lower ($P = 0.05$) after L-arginine relative to placebo; however, there was no difference in triglyceride concentrations when L-arginine treatment was compared to the screening value.

Pearson correlations. The change in plasma L-arginine was inversely correlated with the change in plasma homocysteine ($r = -0.57$, $P = 0.03$, **Fig. 1**). Individuals with the largest increases in plasma L-arginine had the largest decreases in homocysteine. Paradoxically, subjects with the smallest increases in plasma L-arginine after 3 wk of treatment showed the largest reductions in diastolic BP ($r = 0.53$, $P = 0.04$; **Fig. 2**). There were no significant correlations between change in plasma L-arginine and change in any of the other hemodynamic variables.

TABLE 2

Effects of placebo and L-arginine treatment on cardiovascular variables at rest and during stress in hypercholesterolemic men[1,2]

	SBP[a,b]			DBP[a,b,c]		
	Placebo	L-Arginine	Change	Placebo	L-Arginine	Change
	mm Hg			*mm Hg*		
Base	115.2 ± 4.2	114.6 ± 4.2	−0.60 ± 3.51	72.2 ± 2.2	70.9 ± 2.2	−1.24 ± 1.69
Speech	145.8 ± 4.3	145.4 ± 4.3	−0.44 ± 3.64	87.0 ± 2.2	85.7 ± 2.2	−1.34 ± 1.69
Recovery	122.3 ± 4.2	119.9 ± 4.2	−2.48 ± 3.52	75.9 ± 2.2	74.5 ± 2.2	−1.42 ± 1.63
Cold	142.2 ± 4.3	138.6 ± 4.3	−3.66 ± 3.64	87.0 ± 2.2	85.7 ± 2.2	−1.34 ± 1.69
Mean	131.2 ± 3.6	128.5 ± 3.6	−2.69 ± 1.48	80.2 ± 2.0	78.3 ± 2.0	−1.88 ± 0.69
	Cardiac output[a,c]			Total peripheral resistance[b]		
	L/m			*dyne · s · cm^{-5}*		
Base	7.3 ± 0.4	6.9 ± 0.4	−0.41 ± 0.37	987.2 ± 80.9	1053.2 ± 80.9	65.99 ± 51.19
Speech	9.0 ± 0.5	8.9 ± 0.4	−0.18 ± 0.39	1002.8 ± 82.1	1008.4 ± 82.2	5.62 ± 54.87
Recovery	7.4 ± 0.4	6.6 ± 0.4	−0.75 ± 0.37	1043.6 ± 81.5	1094.1 ± 82.1	50.51 ± 54.08
Cold	7.3 ± 0.4	7.1 ± 0.4	−0.26 ± 0.37	1225.5 ± 80.9	1279.9 ± 81.5	54.50 ± 52.18
Mean	7.8 ± 0.4	7.5 ± 0.4	−0.35 ± 0.15	1059.7 ± 73.9	1087.6 ± 74.0	27.95 ± 21.71
	Heart rate[a]			Stroke volume		
	bpm			*mL/beat*		
Base	68.0 ± 3.7	67.6 ± 3.7	−0.44 ± 2.73	110.9 ± 8.3	105.5 ± 8.3	−5.42 ± 3.80
Speech	83.9 ± 3.7	82.5 ± 3.7	−1.41 ± 2.73	113.9 ± 8.4	111.5 ± 8.4	−2.41 ± 3.99
Recovery	68.5 ± 3.7	66.7 ± 3.7	−1.71 ± 2.73	110.4 ± 8.3	104.2 ± 8.3	−6.25 ± 3.80
Cold	72.5 ± 3.7	72.0 ± 3.7	0.50 ± 2.73	105.1 ± 8.3	104.4 ± 8.3	−0.74 ± 3.80
Mean	74.3 ± 3.3	73.0 ± 3.3	−1.31 ± 1.13	109.2 ± 8.0	106.5 ± 8.0	−2.67 ± 1.59
	Pre-ejection period[a,c]			Arterial pressure[a,b,c]		
	ms			*mm Hg*		
Base	132.8 ± 4.4	138.4 ± 4.4	5.56 ± 3.63	86.6 ± 2.7	85.5 ± 2.7	−1.11 ± 1.99
Speech	121.9 ± 4.6	123.9 ± 4.5	2.03 ± 3.82	106.3 ± 2.8	106.0 ± 2.8	−0.28 ± 2.13
Recovery	133.1 ± 4.4	138.7 ± 4.4	5.56 ± 3.63	91.4 ± 2.7	89.7 ± 2.7	−1.69 ± 1.99
Cold	133.7 ± 4.4	134.8 ± 4.4	1.10 ± 3.63	104.9 ± 2.7	103.2 ± 2.8	−1.63 ± 2.02
Mean	129.4 ± 3.8	132.8 ± 3.8	3.35 ± 1.52	97.1 ± 2.4	95.2 ± 2.4	−1.92 ± 0.84

[1] Values are least squares means ± SEM, $n = 16$. [a] Significant speech task effect (vs. baseline level) overall, collapsing across treatments. $P \leq 0.0001$. [b] Significant cold pressor task effect (vs. baseline level) overall, collapsing across treatments. $P \leq 0.0001$. [c] Significant effect of treatment collapsing across all tasks, $P \leq 0.05$.

[2] Mean change scores were calculated using speech preparation and cold pressor recovery although those individual tasks are not shown.

TABLE 3

Effects of placebo and L-arginine treatment on amino acid concentrations in hypercholesterolemic men[1]

	Placebo	L-Arginine	Change
	μmol/L		%
Alanine	523.7 ± 24.6	544.3 ± 36.4	5.8 ± 8.6
Arginine	125.8 ± 11.5	193.8 ± 15.1[a]	68.7 ± 12.0
ADMA	0.42 ± 0.06	0.40 ± 0.08	−4.0 ± 4.0
Asparagine	39.2 ± 1.7	47.7 ± 4.1	19.7 ± 8.2
Aspartic acid	5.0 ± 0.3	5.3 ± 0.3	7.3 ± 6.2
Citrulline	50.1 ± 4.9	53.3 ± 6.5	6.4 ± 5.1
Cysteine	499.2 ± 28.3	549.8 ± 30.9	13.7 ± 7.9
Glutamic acid	36.1 ± 5.9	44.2 ± 7.2	34.1 ± 12.9
Glutamine	712.5 ± 29.2	713.4 ± 36.6	1.5 ± 5.0
Glycine	284.5 ± 17.1	286.7 ± 14.2	1.2 ± 5.1
Histidine	101.3 ± 3.9	112.6 ± 5.7	12.7 ± 6.6
Homocysteine	14.1 ± 1.2	12.0 ± 0.8[a]	−9.7 ± 4.2
Isoleucine	91.6 ± 5.8	89.3 ± 6.2	−0.7 ± 5.0
Leucine	187.1 ± 9.0	200.1 ± 11.6	7.7 ± 5.5
Lysine	246.4 ± 11.5	257.8 ± 11.9	6.0 ± 6.3
Methionine	34.8 ± 1.2	36.4 ± 2.3	3.1 ± 6.0
Ornithine	63.5 ± 4.8	99.5 ± 7.2[a]	68.4 ± 10.9
Phenylalanine	78.1 ± 3.1	85.1 ± 5.6	7.5 ± 5.0
Proline	253.7 ± 16.9	246.3 ± 20.6	−1.4 ± 8.6
Serine	97.4 ± 4.3	106.5 ± 6.5	8.9 ± 6.1
Taurine	83.0 ± 4.6	97.5 ± 7.9	16.6 ± 7.9
Threonine	150.8 ± 7.7	154.9 ± 9.6	1.2 ± 4.7
Tyrosine	82.0 ± 4.0	86.4 ± 5.5	5.0 ± 6.1
Valine	361.6 ± 22.0	391.6 ± 26.1	8.7 ± 5.7
Total amino acids	4196.2 ± 114.5	4431.5 ± 188.1	6.2 ± 5.6

[1] Values are means ± SEM, $n = 16$. [a] Different from the placebo period, $P < 0.05$.

DISCUSSION

In men with high cholesterol, L-arginine (12 g/d for 3 wk) lowered diastolic BP by 2 mm Hg and homocysteine by 2.0 μmol/L. Subjects with the largest increases in plasma L-arginine also showed the largest decreases in homocysteine. In contrast to previous studies that used i.v. administration of high-dose L-arginine, BP reductions in the present study were smaller in magnitude and were mediated by reductions in cardiac output rather than total peripheral resistance. To our knowledge, this study is the first to show that oral L-arginine supplements significantly reduce plasma homocysteine to a similar degree as a single high-dose infusion (38), and it is the first to suggest a hemodynamic mechanism for the blood pressure lowering effects of oral L-arginine.

The hemodynamic pattern we observed (lower cardiac output with no change in peripheral resistance) is quite different from the transient vasodilation typically reported with i.v. L-arginine (8–11). However, postinfusion plasma concentrations of L-arginine are 30–50 times higher than the concentrations reported here (8,38). In adults with hypercholesterolemia, it has been documented that high plasma ADMA, a competitive inhibitor of NO synthase, impairs NO signaling (20,39). However, in the present study, it is possible that supplemental L-arginine failed to enhance the production and/or the activity of NO (despite a marked increase in the arginine:ADMA ratio) because ADMA concentrations were not very high in our volunteers. In support of this hypothesis,

TABLE 4

Effects of placebo and L-arginine treatment on plasma concentrations and ratios of markers of coronary risk in hypercholesterolemic men[1]

	Placebo	L-Arginine
Total cholesterol, *mmol/L*	6.1 ± 0.2	6.0 ± 0.2
LDL-C, *mmol/L*	4.0 ± 0.2	4.0 ± 0.2
HDL-C, *mmol/L*	1.1 ± 0.1	1.2 ± 0.1
Triglycerides, *mmol/L*	2.3 ± 0.3	1.7 ± 0.3[a]
nonHDL-C, *mmol/L*	4.9 ± 0.2	4.8 ± 0.2
Total cholesterol/HDL-C	0.14 ± 0.01	0.14 ± 0.01
LDL-C/HDL-C	0.09 ± 0.01	0.09 ± 0.01
Insulin, *pmol/L*	132.8 ± 13.3	135.5 ± 13.3
Glucose, *mmol/L*	5.4 ± 0.1	5.4 ± 0.1
CRP,[2] *mg/L*	0.28	0.22

[1] Values are least squares means ± SEM unless otherwise indicated, $n = 16$. [a] Different from the placebo period but not from the study entry value.

[2] Because of skewed distributions, medians are presented for C-reactive protein.

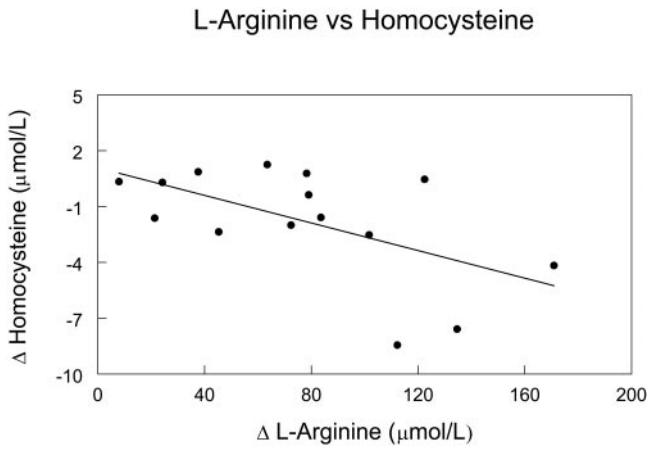

FIGURE 1 Change in plasma L-arginine was inversely correlated with change in plasma homocysteine in hypercholesterolemic men ($r = -0.57$, $P = 0.03$). Subjects with the largest increase in plasma L-arginine showed the largest decreases in plasma homocysteine.

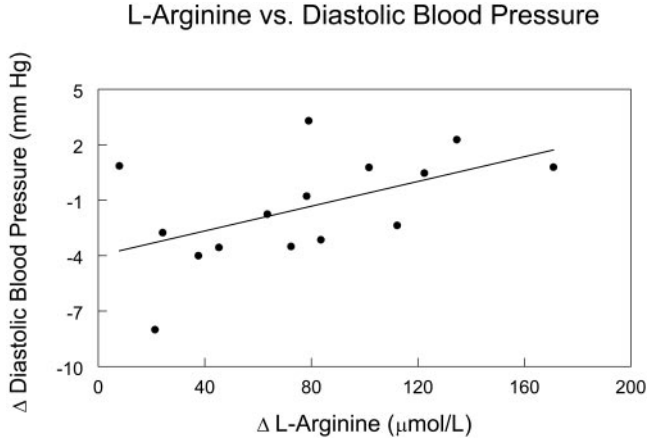

FIGURE 2 Change in plasma L-arginine was positively correlated with change in diastolic BP in hypercholesterolemic men ($r = 0.53$, $P = 0.04$). Subjects with the smallest increases in plasma L-arginine after 3 wk of treatment showed the largest BP reductions.

we found substantial increases in plasma ornithine (68%) but only small, nonsignificant ($P = 0.23$) changes in citrulline (6%), suggesting that supplemental L-arginine was metabolized by arginase rather than NO synthase.

It is surprising that subjects with the largest treatment-related increases in plasma L-arginine also showed the largest increases in diastolic BP with treatment. However, it is important to note that amino acid concentrations were tested in fasting subjects (10–12 h after the last dose with the evening meal) and hemodynamic measures were collected 2 h after the last dose of L-arginine. Simultaneous measurements of amino acids and BP should be collected in future studies to better understand this effect.

It is unlikely that the null effects for total peripheral resistance were a result of low statistical power. Furthermore, we have shown that total peripheral resistance can be reliably measured in the absence of changes in diet and exercise (34) and that it is sensitive to the vasodilatory effects of acute exercise (40) and hormone replacement therapy (41,42). Given the importance of NO in regulating renal function (43), it is possible that L-arginine's effects on cardiac output were a result of enhanced natriuresis. When given intravenously, L-arginine substantially reduces renovascular resistance (44) and increases sodium excretion in healthy subjects (45). In the present study, L-arginine was associated with small but significant increases in the pre-ejection period, suggesting a generalized sympatholytic effect of L-arginine that may also result in lower renovascular resistance and reduced blood volume. Further work is needed to understand the mechanisms and dose-dependency of these effects. Given the inconsistencies across studies on the magnitude of BP lowering with oral L-arginine, future studies should carefully document the background diet of participants to confirm whether habitual diet modifies L-arginine's hypotensive effects (12,46,47).

Few studies have examined L-arginine's effects on homocysteine, although it has been suggested (48) that L-arginine might actually increase flux from *S*-adenosyl methionine to *S*-adenosyl homocysteine in response to the high methylation demand for the synthesis of creatine from arginine. Our results are in agreement with those of Cassone-Faldetta and colleagues (38), who found significant (but transient) reductions in plasma homocysteine 30 min after administration of 3 g i.v. L-arginine. In keeping with the present study, homocysteine reductions were largest in subjects with the largest increases in plasma L-arginine. Our study suggests that L-arginine supplements may actually reduce homocysteine and that the magnitude of this effect is dependent on plasma concentrations of L-arginine.

In conclusion, adults in western countries typically consume 4–5 g/d of L-arginine (49,50). This study showed that increased intake of L-arginine through dietary supplements was associated with small reductions in diastolic BP and moderate reductions in plasma homocysteine. The 12 g/d dose should also be sufficient to reduce platelet aggregation (51) and monocyte adhesiveness (52), although improvements in endothelium-dependent vasodilation may require higher doses (5,15,53). Most importantly, hemodynamic changes were evident at rest and during 2 standardized stressor tasks, suggesting that they would also be evident during daily activities.

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