

Applied nutritional investigation

Beneficial effects of aged garlic extract and coenzyme Q10 on vascular elasticity and endothelial function: The FAITH randomized clinical trial

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ABSTRACT

Objective: Aged garlic extract (AGE) is associated with a significant decrease in atherosclerotic plaque progression and endothelial function improvement. Similarly, coenzyme Q10 (CoQ10) has significant beneficial effects on endothelial function. A stressful lifestyle is a well-known risk factor for the presence and progression of atherosclerosis. This study investigated the effect of AGE plus CoQ10 on vascular elasticity measured by pulse-wave velocity (PWV) and endothelial function measured by digital thermal monitoring (DTM) in firefighters.

Methods: Sixty-five Los-Angeles County firefighters who met the eligibility criteria were enrolled in this placebo-controlled, double-blinded randomized trial. The firefighters were randomized to four tablets of AGE (300 mg/tablet) plus CoQ10 (30 mg/tablet) or placebo. The participants underwent quarterly visits and 1-year follow-up. PWV and DTM were measured at baseline and at the 1-year follow-up.

Results: There were no significant differences in age, cardiovascular risk factors, PWV, and DTM between the AGE/CoQ10 and placebo groups at baseline ($P > 0.5$). At 1-y, PWV and DTM significantly improved in the AGE/CoQ10 compared with the placebo group ($P < 0.05$). After an adjustment for cardiovascular risk factors and statin therapy, the mean decrease in vascular stiffness (PWV) was 1.21 m/s in the AGE/CoQ10 compared with the placebo group ($P = 0.005$). Similarly, the mean increase in the area under the temperature curve, the DTM index of endothelial function, was 31.3 in the AGE/CoQ10 compared with the placebo group ($P = 0.01$).

Conclusion: The combination of AGE and CoQ10 was independently associated with significant beneficial effects on vascular elasticity and endothelial function in firefighters with high occupational stress, highlighting the important role of AGE and CoQ10 in atherosclerotic prevention of such individuals.

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Introduction

The risk of major adverse cardiovascular events is three-fold higher in firefighters compared with the general population [1]. The leading cause of line-of-duty death in firefighters is sudden cardiac death, accounting for approximately 45% of duty deaths [2]. Studies have shown that self-perceived psychological stresses and occupational stressors such as fire-fighting are

independent risk factors for coronary artery disease (CAD) and major cardiac adverse events such as myocardial infarction [3,4].

A growing body of literature supports the linkage of endothelial dysfunction, inflammation, and immunologic factors to the acceleration of the atherosclerotic process from the early subclinical phase to overt clinical CAD with complications [5]. Our group previously reported that aged garlic extract (AGE) has cardiovascular protective effects by decreasing the progression of the coronary atherosclerotic burden, oxidative biomarkers, and blood cholesterol levels in intermediate-risk individuals [6].

A primary prevention method to retard the progression of atherosclerosis at the earliest subclinical stages in individuals with highly stressful occupational or environmental lifestyles has not been well validated. The Firefighter Aged Garlic Extract Investigation with Coenzyme (CoQ10) as a Treatment for Heart

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Disease (FAITH), a randomized, placebo-controlled trial, was designed to assess the effects of AGE plus CoQ10 on vascular elasticity and endothelial function in firefighters.

Materials and methods

Sixty-five asymptomatic Los Angeles County firefighters who met the eligibility criteria (Table 1) signed a written informed consent that has been approved by the institutional review board of the Los Angeles Biomedical Research Institute and were enrolled in the placebo-controlled, double-blinded randomized FAITH clinical trial (Clinicaltrials.gov identifier, NCT00860847). Demographic and cardiovascular risk factors and serum lipid profiles were obtained using standard techniques at baseline and during the 1-y course of the trial. Pulse-wave velocity (PWV) and digital thermal monitoring (DTM) tests were measured at baseline and after the 1-y follow up.

Firefighters were randomized to four identical tablets of AGE/CoQ10 or placebo at a 1:1 ratio using a computer-generated randomization method. The AGE/CoQ10 tablets contained AGE 300 mg and CoQ10 30 mg, which were provided by Wakanuga Inc. of America (Mission Viejo, CA, USA). The participants underwent quarterly visits for compliance assessment, drug refills, and diet and nutritional consultations.

Pulse-wave velocity

Carotid and radial PWVs were measured by the SphygmoCor device (AtCor Medical, Sydney, Australia) based on the tonometry in two steps. The first step is used to simultaneously record carotid pulse waves and the electrocardiogram; the second step is the recording of the radial pulse wave and the electrocardiogram. The electrocardiographic recording during the measurements was used for the synchronization of the carotid and radial pulse-wave times. The transit time between the carotid and radial pressure waves was calculated using the wave foot-to-foot method. Wave “foots” are identified using intersecting tangent algorithms. The PWV is calculated as the ratio of the pulse-wave travel distance from the carotid to the radial artery to its transit time (meters per second) [7].

Digital thermal monitoring

The DTM measurements were performed in a quiet, dimmed room at a controlled ambient temperature of 23.5°C to 25.0°C. The DTM of both hands was obtained during a 5-min stabilization, a 2-min cuff inflation to 50 mmHg greater than systolic blood pressure, and a 3-min deflation using an automated, operator-independent protocol (VENDYS, Endothelix Inc., Houston, TX, USA). Thermal changes during a 5-min arm cuff-induced reactive hyperemia test were monitored continuously in the fingertips of the occluded and non-occluded arms using VENDYS software. The device consists of a computer-based thermometric system (0.005°C thermal resolution) with two fingertip, resistance temperature detector, fast-response probes, designed to minimize the skin-probe contact area and fingertip pressure, attached to the pulp of the index finger on each hand. The system includes a common automated sphygmomanometric cuff, a cuff-inflation pump, and a release valve to permit the non-invasive measurement of arterial pressure and the control of occlusive hyperemia. Dual-channel temperature data are simultaneously acquired at a 1-Hz sampling rate. The area under the temperature curve (TMP-AUC), a DTM index of endothelial function, was used to

assess the difference in response in endothelial function between the AGE/CoQ10 and placebo groups [8].

Statistical analysis

Mean \pm standard deviation and proportions were used to summarize the characteristics of the study sample. Continuous variables were compared by *t* test, and categorical variables were compared by the chi-square test. Multivariate logistic regression analyses were employed to assess the relation of vascular elasticity and endothelial function to the AGE/CoQ10 versus placebo treatment before and after an adjustment for conventional cardiovascular risk factors. All statistical analyses were performed with SAS 9.1 (SAS Institute, Cary, NC, USA; <http://www.sas.com>). The level of significance was set at $P < 0.05$ (two-tailed). The study protocol and consent form were approved by the institutional review board committee of the Los Angeles Biomedical Research Institute at Harbor UCLA Medical Center (Torrance, CA, USA).

Results

Table 2 lists the baseline and 1-y measurements of the studied markers between the AGE/CoQ10 and placebo groups. There was no significant difference between the placebo and AGE/CoQ10 groups in age, systolic and diastolic blood pressures, triacylglycerols, low-density lipoprotein cholesterol level, prevalence of hypertension, diabetes mellitus and family history of premature CAD at baseline ($P > 0.05$). The baseline PWV was no different in the AGE/CoQ10 group than in the placebo group (8.11 ± 0.91 versus 8.26 ± 1.15 m/s, $P = 0.6$). Similarly, there was no difference between the AGE/CoQ10 and placebo groups in baseline TMP-AUC (61.4 ± 8.3 versus 47.3 ± 7.3 , $P = 0.3$). After 1 y of the study, the mean decrease in PWV was 1.21 m/s in the AGE/CoQ10 group compared with the placebo group ($P = 0.005$), and the mean increase in TMP-AUC was 31.28 in the AGE/CoQ10 group compared with the placebo group ($P = 0.01$) after an adjustment for all risk factors (Table 2, Fig. 1A). An increase in TMP-AUC, which corresponds to faster temperature rebound response after induced ischemia, is a marker of improved endothelial function. Because blood traveling through more elastic vessels takes a longer time, a decrease in PWV also corresponds with an improvement in vascular elasticity.

Compared with the placebo treatment, the relative risk of the AGE/CoQ10 treatment in decreasing the PWV was 1.21 (95% confidence limit -2.1 to -0.32 , $P = 0.005$), and a relative risk of 31.3 (95% confidence limit 1.67–60.91, $P = 0.01$), increased the TMP-AUC, providing strong evidence of an improvement in vascular elasticity and endothelial functions, respectively. Based on these two different methods of vascular function evaluation,

Table 1
Inclusion and exclusion criteria for asymptomatic Los Angeles County firefighters ($n = 65$)

Inclusion criteria	Exclusion criteria
Age 35–84 y	known hypersensitivity to AGE
Agree to refrain from supplemental garlic or significant dietary garlic	current tobacco use
Agatston CAC score >20	current intake of garlic supplements
Willing to sign informed consent	current use of anticoagulants (except for antiplatelet agents)
	chronic renal failure
	weight ≥ 325 lbs.
	diabetic subjects with HbA1c $>12\%$
	serum creatinine >1.4 mg/dL
	triacylglycerols >400 at visit 1
	NYHA class III or IV heart failure
	known bleeding disorder
	history of myocardial infarction stroke or life-threatening arrhythmia
	presence of metal clips (i.e., patients with bypass) or intracoronary stenting precluding accurate measurement of coronary calcification
	partial ileal bypass or known gastrointestinal disease limiting drug absorption

AGE, aged garlic extract; CAC, coronary artery calcium; HbA1c, hemoglobin A1c; NYHA, New York Heart Association

Table 2

PWV and TMP-AUC in AGE/CoQ10 versus placebo group at baseline and absolute change of the same markers after 1-y follow-up for the same groups

Variable	Placebo (n = 32)	AGE/CoQ10 (n = 33)	P
Baseline			
Age (y)	55 ± 6	54 ± 5	0.6
Men	100%	100%	–
Diabetes mellitus	6%	3%	0.5
Hypertension	19%	24%	0.6
Hypercholesterolemia	62%	40%	0.07
Family history of CHD	94%	96%	0.6
History of smoking	26%	27%	0.9
BMI	28 ± 3	29 ± 4	0.5
PWV (m/s)	8.11 ± 0.91	8.26 ± 1.15	0.6
CRP	1.9 ± 2.1	1.9 ± 2.4	0.9
TMP-AUC	61.4 ± 8.3	47.3 ± 7.3	0.3
Absolute change after 1-year follow-up			
PWV (m/s)	0.74 ± 0.28	–0.47 ± 0.33	0.009
CRP	0.97 ± 0.44	–0.34 ± 0.38	0.02
TMP-AUC	–1.79 ± 9.9	29.49 ± 10.47	0.02

AGE, aged garlic extract; BMI, body mass index; CHD, coronary heart disease; CoQ10, coenzyme 10; CRP, C-reactive protein; PWV, pulse-wave velocity; TMP-AUC, area under the temperature curve

the intermediate risk in firefighters with 1 y of AGE/CoQ10 consumption also had less atherosclerotic progression compared with placebo (Tables 3 and 4). After a 1-y consumption of AGE plus CoQ10, an increase in C-reactive protein (CRP) was more common in the placebo group (PWV decrease <0.18), and firefighters in the AGE/CoQ10 group who had a PWV decrease greater than 0.18 showed the greatest CRP decrease, supporting an anti-inflammatory effect of AGE plus CoQ10 (Fig. 1B).

Discussion

This is the first study to demonstrate a benefit with a combination of AGE and CoQ10 on atherosclerotic progression in intermediate-risk firefighters with high occupational stress. The present study demonstrates that, after 1 y of AGE plus CoQ10, the vascular elasticity and endothelial function in firefighters improved significantly.

Endothelial dysfunction is an independent predictor of future cardiovascular events in patients with atherosclerotic risk factors, stable ischemic heart disease, or acute coronary syndromes. Thus, endothelial dysfunction might be a systemic vascular process that not only mediates the development of atherosclerotic plaque but also may modulate its clinical course [5].

Fingertip DTM of vascular reactivity is a non-invasive, operator-independent test based on changes in fingertip temperature during and after arm cuff occlusion. Our previous studies have shown that DTM correlates with the burden of coronary atherosclerosis in asymptomatic individuals and patients with CAD, measured by coronary artery calcium [9], nuclear perfusion imaging [10], and computed tomographic angiography [8].

Several studies have demonstrated strong correlations between vascular elasticity and cardiovascular risk factors and reported the additive prognostic value of PWV in clinical management [11]. In this study, vascular elasticity, measured by PWV, and endothelial function, measured by DTM, improved in the AGE/CoQ10 group. Those improvements are consistent with retardation of the overall atherosclerotic process. Although some theories have suggested that endothelial function improvement is a vasoprotective mechanism of CoQ10, little clinical information in this regard is available. CoQ10 supplementation in

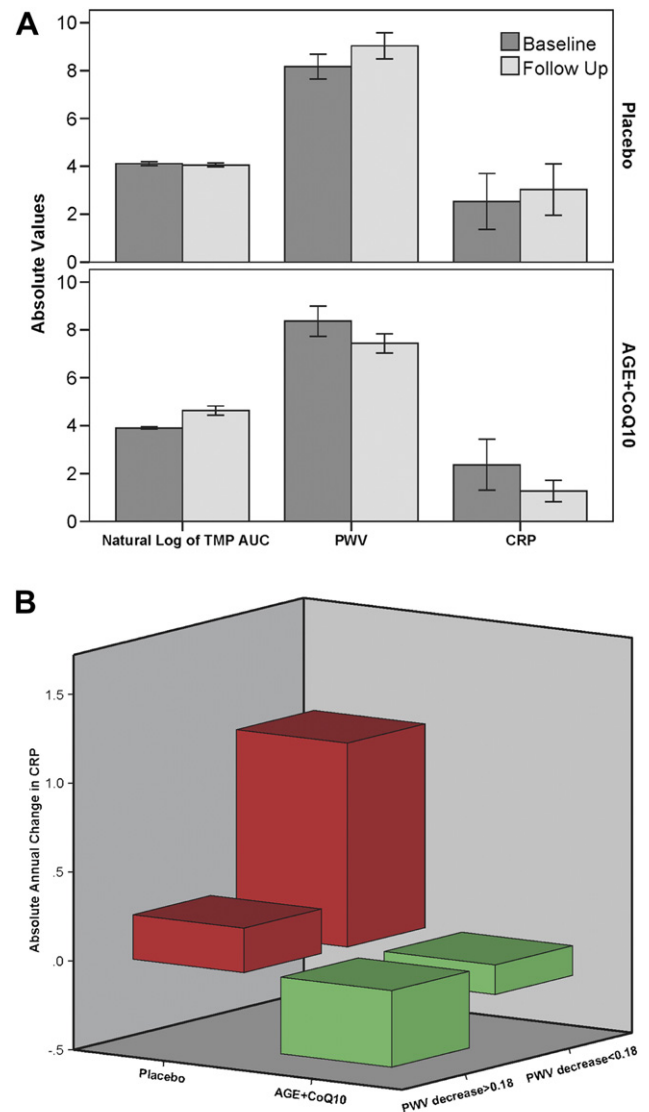


Fig. 1. Absolute annual changes in (A) PWV and the natural logarithm of TMP AUC and (B) CRP with and without a PWV decrease lower than 0.18 in the AGE/CoQ10 and placebo groups after the 1-y study. AGE, aged garlic extract; CoQ10, coenzyme 10; CRP, C-reactive protein; PWV, pulse-wave velocity; TMP AUC, area under the temperature curve.

patients with CAD has been associated with a significant improvement in peak oxygen consumption [12] and with a significant decrease in the plasma lactate/pyruvate ratio, providing evidence to indicate that supplementation decreases

Table 3

Relative risk of PWV and TMP-AUC in AGE/CoQ10 versus placebo group

Model	Placebo	AGE/CoQ10	P
PWV*	1.0 (reference)	–1.21 (–2.1 to –0.32)	0.01
CRP	1.0 (reference)	–1.31 (–2.5 to –0.14)	0.02
TMP-AUC*	1.0 (reference)	31.3 (1.67–60.91)	0.01

AGE, aged garlic extract; CoQ10, coenzyme 10; CRP, C-reactive protein; PWV, pulse-wave velocity; TMP-AUC, area under the temperature curve. Values are presented as odds ratio (95% confidence interval). The model was adjusted for age and conventional risk factors

* Linear regression analysis.

Table 4
Relative risk of PWV and TMP-AUC in AGE/CoQ10 versus placebo group

Model	Placebo	AGE/CoQ10	P
Decrease in PWV*	1.0 (reference)	6.94 (2.1–22.6)	0.001
Decrease in CRP*	1.0 (reference)	7.67 (2.7–21.9)	0.001
Increase in TMP-AUC*	1.0 (reference)	12.60 (3.9–20.6)	0.0001

AGE, aged garlic extract; CoQ10, coenzyme 10; CRP, C-reactive protein; PWV, pulse-wave velocity; TMP-AUC, area under the temperature curve. Values are presented as odds ratio (95% confidence interval). The model was adjusted for age, gender, conventional risk factors, and statin therapy. Logistic regression analysis was used.

* Median annual change: TMP-AUC >9.48 versus ≤9.48, CRP <0 versus ≥0, PWV <0.18 versus ≥0.18.

mitochondrial dysfunction in patients with ischemic left ventricular systolic dysfunction [13]. Our study demonstrated that AGE plus CoQ10 significantly improved vascular elasticity and endothelial function. This provides supplemental evidence of a significant additive value of CoQ10 compared with our first AGE-only trial [6].

The manufacturing and handling processes of garlic modify the chemical characteristics, efficacy, and safety of the final garlic preparations. It is well known that extraction generally increases the potency and bioavailability of various crude botanicals, including garlic, and eliminates harsh and toxic characteristics. Although not all active ingredients of garlic are well known, one of the active ingredients in garlic preparations is S-allyl-cysteine, which has been shown to provide protection against oxidation, free radicals, and cardiovascular diseases and has less toxicity than allicin and diallyl disulfide [14]. Those effects can also be attributed to its rich content of various organic sulfur compounds, including allicin, diallyl sulfide, diallyl disulfide, diallyl trisulfide, and S-allyl-cysteine, which has shown to have hypolipidemic and antithrombotic effects [15]. AGE increases intracellular glutathione levels, glutathione disulfide reductase, and superoxide dismutase (SOD) activity in endothelial cells, whereas the level of glutathione disulfide decreases. These results suggest that the antioxidant effect of AGE may be due to its modulation of the glutathione redox cycle and SOD activity in vascular endothelial cells. AGE also increases nitric oxide levels by stimulating constitutive nitric oxide synthase, but not inducible nitric oxide synthase, or arginine contained in AGE, which could be useful for the prevention of cardiovascular disease. Cardiovascular risk factors have been found to be modulated by the long-term intake of AGE [16]. One recent study has demonstrated anti-inflammatory protection and cytoprotection against oxidative stress characterized by hydrogen sulfide, which is inherently released by garlic [17]. Because the preparation of garlic used in the present and two previous studies [6,18] did not contain allicin, it is unlikely to be the active ingredient associated with the benefit of garlic supplementation.

Coenzyme Q10 is a key component of the mitochondrial respiratory chain and has been recognized to have multiple antioxidant properties. Lee et al. [19] in a recent clinical trial showed significant antioxidant and some anti-inflammatory responses in patients with CAD after a 12-wk consumption of CoQ10 60 and 150 mg/d. They also reported more prominent decreases in SOD and CRP in the group taking higher doses of CoQ10 (60 versus 150 mg/d). Those findings are in line with the present study in which we observed a significant decrease in CRP with the consumption of CoQ10 120 mg/d and AGE 1200 mg/d. Littarru and Tiano [20] in a recent review article on the fundamental role of CoQ10 reported the effect of CoQ10 in improving endothelium-bound extracellular SOD, an enzyme that is thought

to protect blood vessels against oxidant-induced damage, as a possible mechanism in endothelial function improvement.

Clinical significance

Our study demonstrated that AGE plus CoQ10 significantly improved vascular elasticity and endothelial function and for the first time provides supplemental evidence of benefiting high-risk individuals such as firefighters. Primary and secondary preventions for atherosclerosis are critical steps, and primary care physicians are in a position to advise at-risk individuals to use alternative and complementary therapies such as AGE plus CoQ10 to decrease the morbidity and mortality of cardiovascular disease, thus minimizing the economic and social costs associated with the number-one killer in the USA.

Limitations

The present study has several limitations. The sample was relatively small, but we were able to show a clear association between the consumption of AGE and CoQ10 and the retardation of atherosclerotic progression. The present study was not designed to assess which components of the AGE/CoQ10 capsule were most responsible for the observed benefits, and a larger study randomizing patients to individual components of the therapy would be beneficial. Given these limitations, our study might be beneficial for larger cohorts of multiple occupational groups and to assess the possible benefits in more detailed fashion.

Conclusion

The present study demonstrated that compared with the placebo group, AGE plus CoQ10 was effective in slowing the progression of atherosclerosis in intermediate-risk firefighters with high occupational stress, independent of baseline blood pressure, statin use, or other cardiovascular risk factors. There was a strong association between the improvement of vascular elasticity assessed by PWV and the endothelial function evaluated by DTM with AGE plus CoQ10, independent of age, gender, and conventional cardiovascular risk factors. Furthermore, AGE/CoQ10 consumption exhibited improvement in vascular elasticity and endothelial function after 1 y. This robust overall deceleration necessitates more preventive care for firefighters and other similar at-risk groups. This study further highlights the benefits for primary prevention with AGE plus CoQ10. Long-term follow-up clinical trials are warranted to assess the extent of the benefit from this primary prevention in stressful occupations to improve the cardiovascular and psychological health of individuals exposed to highly stressful environments.

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- 3) to discuss solutions to address worldwide micronutrient deficiencies.

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