# Vascular Dysfunction and Chronic Obstructive Pulmonary Disease

## The Role of Redox Balance

Stephen J. Ives,* Ryan A. Harris,* Melissa A.H. Witman, Anette S. Fjeldstad, Ryan S. Garten, John McDaniel, D. Walter Wray, Russell S. Richardson

**See Editorial Commentary, pp 444–446**

***Abstract***—Chronic obstructive pulmonary disease (COPD) is characterized by low pulmonary function, inflammation, free radical production, vascular dysfunction, and subsequently a greater incidence of cardiovascular disease. By administering an acute oral antioxidant cocktail to patients with COPD (n=30) and controls (n=30), we sought to determine the role of redox balance in the vascular dysfunction of these patients. Using a double-blind, randomized, placebo-controlled, crossover design, patients with COPD and controls were ingested placebo or the antioxidant cocktail (vitamin C, vitamin E, α-lipoic acid) after which brachial artery flow-mediated dilation and carotid-radial pulse wave velocity were assessed using ultrasound Doppler. The patients exhibited lower baseline antioxidant levels (vitamin C and superoxide dismutase activity) and higher levels of oxidative stress (thiobarbituic acid reactive species) in comparison with controls. The patients also displayed lower basal flow-mediated dilation ($P<0.05$), which was significantly improved with antioxidant cocktail (3.1±0.5 versus 4.7±0.6%; $P<0.05$; placebo versus antioxidant cocktail), but not controls (6.7±0.6 versus 6.9±0.7%; $P>0.05$; placebo versus antioxidant cocktail). The antioxidant cocktail also improved pulse wave velocity in patients with COPD (14±1 versus 11±1 $m \cdot s^{-1}$; $P<0.05$; placebo versus antioxidant cocktail) while not affecting controls (11±2 versus 10±1 $m \cdot s^{-1}$; $P>0.05$; placebo versus antioxidant). Patients with COPD exhibit vascular dysfunction, likely mediated by an altered redox balance, which can be acutely mitigated by an oral antioxidant. Therefore, free radically mediated vascular dysfunction may be an important mechanism contributing to this population's greater risk and incidence of cardiovascular disease. **(*Hypertension*. 2014;63:459-467.)**

**Key Words:** free radicals ■ oxidative stress ■ pulmonary disease, chronic obstructive ■ vascular stiffness

Chronic obstructive pulmonary disease (COPD) is a condition that originates in the pulmonary system but is now well recognized to manifest as a syndrome encompassing other symptomology and comorbidities beyond pulmonary disease, most of which seem to be vascular related.[1–6] Specifically, an increased incidence of hypertension, advanced atherosclerosis, coronary artery disease, peripheral vascular disease, and elevated cardiovascular mortality are now often considered hallmarks of COPD.[4–8] However, the mechanistic link among COPD, vascular dysfunction, and cardiovascular disease (CVD) remains to be elucidated.

Vascular endothelial function has been documented to be related to both CVD risk[9–11] and incidence.[12,13] Consequently, assessments of flow-mediated dilation (FMD), a measure of vascular endothelial function, and pulse wave velocity (PWV), a measure of vascular stiffness, have grown in popularity as independent predictors of CVD risk.[14–16] Using the FMD technique, previous studies have demonstrated that patients with COPD display reduced vascular function compared with age-matched controls.[17–19] Similarly, studies have revealed that patients with COPD exhibit significantly elevated vascular stiffness as assessed by PWV.[20–22] In addition, there is a growing hypothesis that pulmonary vascular dysfunction itself may be a key factor that instigates the development of COPD[23–25] and as a consequence systemic vascular dysfunction follows,[26] although the nature of this relationship is currently not well understood.[27]

Received August 17, 2013; first decision August 28, 2013; revision accepted October 28, 2013.

From the Geriatric Research, Education, and Clinical Center, George E. Whalen VA Medical Center, Salt Lake City, UT (S.J.I., M.A.H.W., R.S.G., D.W.W, R.S.R.); Department of Internal Medicine, Division of Geriatrics (S.J.I., M.A.H.W., R.S.G., D.W.W., R.S.R.) and Department of Neurology (A.S.F.), University of Utah, Salt Lake City; Department of Health and Exercise Sciences, Skidmore College, Saratoga Springs, NY (S.J.I.); Institute of Preventative and Public Health, Georgia Prevention Center, Medical College of Georgia, Augusta (R.A.H.); Sport and Exercise Science Research Institute, University of Ulster, Jordanstown, Co. Antrim, Northern Ireland, United Kingdom (R.A.H.); Department of Exercise and Sport Science, Kent State University, OH (J.M.D.); Louis Stokes VA Medical Center, Cleveland VA Medical Center, OH (J.M.D.); and Department of Exercise and Sport Science, University of Utah, Salt Lake City (D.W.W., R.S.R.).

*These authors contributed equally to this work.

Correspondence to Russell S. Richardson, VA Medical Center Bldg 2, Rm 1D25, 500 Foothill Dr, Salt Lake City, UT 84148. E-mail r.richardson@hsc.utah.edu



*Hypertension* **is available at http://hyper.ahajournals.org** **DOI: 10.1161/HYPERTENSIONAHA.113.02255**

Chronic inflammation associated with COPD[4,20,28–31] may be the instigator of, and related to, the peripheral vascular dysfunction associated with this population.[18] However, the downstream effects of this inflammation[32] and the subsequent role of free radicals in disrupting vascular function in these patients have received little attention. To date, we are unaware of a single study that has aimed to reduce free radicals in patients with COPD to determine whether this can improve vascular function, thus providing a mechanistic link between oxidative stress and the elevated CVD risk, incidence, and mortality in this population.[3]

Previously, it has been documented that an acute antioxidant cocktail (AOC) of known efficacy[33,34] is capable of improving vascular function, as assessed by FMD, in the elderly population[33] and heart transplant recipients with history of heart failure,[35] populations with increased CVD risk and a definitive incident, respectively. The AOC-induced improvement in redox balance[36] likely attenuates free radical–mediated reductions in nitric oxide (NO) bioavailability, thus increasing endothelial-dependent FMD.[33–35] These previous studies highlight the role of free radicals in mediating vascular dysfunction in vulnerable populations and that the AOC-induced improvements in vascular function were mediated by an improvement in redox balance.

Accordingly, by acutely administering an AOC to patients with COPD, we sought to determine the role of redox balance in vascular dysfunction (FMD and PWV) in patients with COPD. Specifically, we used FMD and PWV to assess vascular endothelial function and stiffness after acute ingestion of an oral AOC or placebo in patients with COPD and age-matched controls. We hypothesized that the AOC would improve FMD and reduce PWV in patients with COPD but not in controls, highlighting the role of redox balance in vascular dysfunction in this patient population.

## Methods

### Subjects and General Procedures

Sixty volunteers were recruited for this study: 30 patients with COPD and 30 age- and sex-matched controls (Table 1). Although the majority of patients with COPD had a significant or recent history of smoking (months since quitting: 96±33), current smokers were excluded because acute smoke inhalation is capable of altering endothelial function.[37] A single control subject reported a history of smoking but quit 240 months before the study. In accordance with recent guidelines,[38] the inclusion criterion for patients with COPD was a pulmonary function test that was performed after bronchodilator administration, indicating a forced expiratory volume$_{1.0}$/forced vital capacity ratio <0.70. In addition, none of the patients with COPD reported a recent exacerbation (<3 months; Table 2) and were stable, in terms of symptom severity, during both visits. Subject characteristics, such as prevalence of coronary artery disease and obstructive sleep apnea, unless otherwise indicated, were determined from health histories. The protocol was approved by the Institutional Review Boards of the University of Utah and the Salt Lake City Department of Veterans Affairs Medical Center. Written informed consent was obtained from each subject before participation in this study.

All subjects reported to the laboratory twice within 1 week (>48 hours apart) after ingesting either placebo or the AOC in a balanced, double-blind, crossover design. The standardized AOC, taken in the same manner, by all subjects was composed of 2 separate doses of vitamin C, vitamin E, and α-lipoic acid,[33] which has previously been documented to reduce plasma free radicals as measured by electron paramagnetic resonance (EPR) spectroscopy.[34,39] The first AOC dose (300 mg α-lipoic acid, 500 mg vitamin C, 200 IU vitamin E) was taken 90 minutes before testing, whereas the second AOC dose (300 mg α-lipoic acid, 500 mg vitamin C, 400 IU vitamin E) was taken 60 minutes before testing. These doses and the dosing paradigm were chosen based on both practicality (doses found in over-the-counter formulations) and efficacy as assessed by a reduction in the free radical EPR signal.[34,39] Placebo microcrystalline cellulose capsules of similar taste, color, and appearance were likewise consumed in the same manner as the AOC trial. Subjects reported to the laboratory in a fasted state and without caffeine or alcohol use for 12 and 24 hours, respectively. They also had not performed any exercise within the past 24 hours. On arrival, a venous blood sample was obtained for blood chemistry (electrolytes, creatinine, glucose, and so on), lipid panel, complete blood count, and biochemical assays (markers of antioxidant capacity and oxidative stress). After this blood draw, subjects were positioned supine and rested quietly for 20 minutes before PWV and FMD testing.

**Table 1. Characteristics of Patients With COPD and Controls**

| Characteristic | Control (n=30) | COPD (n=30) |
|---|---|---|
| Age, y | 66±2 | 66±2 |
| Female/male, n | 15/15 | 15/15 |
| Height, cm | 169±2 | 166±2 |
| Weight, kg | 74±3 | 73±4 |
| BMI, kg/m$^2$ | 25±1 | 26±1 |
| SBP, mm Hg | 129±3 | 136±4 |
| DBP, mm Hg | 79±1 | 80±2 |
| FEV$_{1.0}$, L | 3.0±0.2 | 1.3±0.1* |
| FEV$_{1.0}$, % predicted | 107±4 | 55±4* |
| FVC, L | 4.0±0.3 | 3.0±0.2* |
| FEV$_{1.0}$/FVC ratio, % | 76±1 | 45±3* |
| GOLD classification, (n/group) | | |
| Mild | … | 3 |
| Moderate | … | 13 |
| Severe | … | 8 |
| Very severe | … | 6 |
| Creatinine, mg/dL | 1.0±0.0 | 1.0±0.1 |
| Urea nitrogen, mg/dL | 16.3±0.6 | 16±1.2 |
| Glucose, mg/dL | 85±3.8 | 87±3.2 |
| Cholesterol, mg/dL | 198±10 | 195±11 |
| HDL cholesterol, mg/dL | 51±3.2 | 57±4.1 |
| LDL cholesterol, mg/dL | 128±8 | 121±9 |
| Triglycerides, mg/dL | 144±19 | 110±12 |
| Erythrocytes, M/μL | 5.1±0.1 | 5.0±0.1 |
| Hemoglobin, g/dL | 15.2±0.3 | 15.4±0.5 |
| Hematocrit, % | 45±0.7 | 47±1.5 |
| Leukocytes, K/μL | 5.6±0.3 | 7.4±0.6* |
| Neutrophils, K/μL | 3.4±0.3 | 4.8±0.5* |
| Lymphocytes, K/μL | 1.6±0.1 | 1.8±0.2 |
| Monocytes, K/μL | 0.5±0.0 | 0.6±0.0* |
| Eosinophils, K/μL | 0.3±0.1 | 0.2±0.0 |
| Basophils, K/μL | 0.1±0.0 | 0.0±0.0 |

Values are expressed as mean±SE. BMI indicates body mass index; COPD, chronic obstructive pulmonary disease; DBP, diastolic blood pressure; FEV, forced expiratory volume; FVC, forced vital capacity; GOLD, Global Initiative for Chronic Obstructive Lung Disease; HDL, high-density lipoprotein; LDL, low-density lipoprotein; and SBP, systolic blood pressure.

*$P$<0.05 control vs COPD.

### Brachial Artery FMD and Reactive Hyperemia

The FMD was performed in accordance with recent guidelines.[40] Briefly, after baseline measurements of brachial artery diameter and

**Table 2. Subject History of Controls (n=30) and Patients With COPD (n=30)**

| History | Control (No. of Cases) | COPD (No. of Cases) |
|---|---|---|
| Hypertension | 7 | 17* |
| Hypercholesterolemia | 7 | 6 |
| Chronic kidney disease | 0 | 0 |
| Diabetes mellitus | 1 | 0 |
| Hypothyroid | 2 | 3 |
| Chronic heart failure | 1 | 2 |
| Anemia | 0 | 2 |
| Coronary artery disease | 1 | 2 |
| Self-reported tobacco smoking history | 1 | 26* |
| Obstructive sleep apnea | 1 | 2 |
| Exacerbation in past 3 mo req hospitalization | 0 | 0 |
| Exacerbation in past 6 mo req corticosteroids | 0 | 3 |
| Prescribed supplemental oxygen | 0 | 7 |
| Taking supplemental oxygen | 0 | 7 |
| Medications | | |
| Pulmonary | | |
| Short-acting anticholinergic bronchodilators | 0 | 5* |
| Long-acting anticholinergic bronchodilators | 0 | 7* |
| Long-acting $\beta_2$-agonist | 1 | 14* |
| Corticosteroids | 0 | 11* |
| Cardiovascular | | |
| Calcium channel inhibitor | 2 | 9* |
| Antiarrhythimc | 1 | 1 |
| Thiazide diruetic | 2 | 4 |
| ACE inhibitor | 2 | 10* |
| Angiotensin receptor blocker | 1 | 2 |
| β-Blocker | 2 | 4 |
| Statin | 6 | 5 |
| Anticoagulant | 0 | 1 |

n=30 controls and n=30 patients with COPD. ACE indicates angiotensin-converting enzyme; COPD, chronic obstructive pulmonary disease; and req, requiring.

*$P$<0.05 control vs patients with COPD.

blood velocity were performed, a blood pressure cuff was inflated to a suprasystolic pressure for 5 minutes. On cuff release, measurements were assessed continuously for 2 minutes. During baseline and cuff release, images were sent in real-time to off-line software and later analyzed using automated edge detection. These analyses were performed by a trained technician who was blinded to both subject group and condition. FMD was quantified as the peak diameter observed postocclusion and expressed as percent change from baseline (%). Reactive hyperemia (RH) was quantified as the cumulative area under the curve (AUC) for brachial artery blood flow during the entire 2-minute postocclusion period.

### Pulse Wave Velocity

Concurrent pilot work in a separate study revealed a positive effect of the AOC on PWV in patients with COPD; therefore, because of the timing of this observation, this measurement was only performed in patients with COPD (n=17) and age- and sex-matched controls (n=17) during the latter portion of the current study. Before the FMD test, ultrasound Doppler measurements were taken at the carotid and radial arteries to assess peripheral arterial stiffness (carotid-radial PWV), an approach previously used in this population[22] and has been documented to detect elevations in PWV in populations with heightened CVD risk.[41,42] In addition, in young healthy individuals, we have found that carotid-radial and carotid-femoral PWV are significantly related, and carotid-radial PWV is, therefore, a predictor of carotid-femoral PWV ($r$=0.5; S. Ives, PhD, unpublished observations, 2013). PWV was calculated using the foot-to-foot ECG-gated method as described previously[43,44] and expressed as meters per second ($m \cdot s^{-1}$).

### Data Analysis

AUC for shear rate and RH was calculated using the trapezoidal rule for the 2 minutes after cuff deflation. Statistical comparisons were performed using 2-way ANOVA, ANCOVA, $t$ tests, and $\chi^2$, where appropriate. The level of significance was established at 0.05. All data are expressed as mean±SEM.

## Results

### Subject Characteristics

The subject characteristics and medical history are presented in Tables 1 and 2, respectively. The subjects were well matched, aside from the typical greater incidence of hypertension in patients with COPD, compared with controls. Of note, because there was no difference in vascular function (FMD) between patients with COPD who had a history of hypertension and those who did not ($P$=0.56), the patients were not divided into 2 groups based on this characteristic. There were no sex differences with regard to basal vascular function (FMD or PWV) in either group or in response to the AOC; therefore, the data for both sexes were combined.

### FMD and RH

Under the placebo condition, patients with COPD displayed significantly lower FMD compared with controls (3.1±0.5 versus 6.7±0.6%; $P$<0.05; COPD versus control; Figure 1). FMD was significantly improved with ingestion of AOC in patients with COPD (3.1±0.5 versus 4.7±0.6%; $P$<0.05; placebo versus AOC) but not in controls (6.7±0.6 versus 6.9±0.7%; $P$>0.05; placebo versus AOC; Figure 1). These results also held true when expressed as absolute change in brachial artery diameter, confirming a lower FMD in patients with COPD under placebo condition (0.01±0.002 versus 0.03±0.003 cmΔ; $P$<0.05; COPD versus control) that was improved by ingestion of the AOC in patients (0.01±0.002 versus 0.02±0.003 cmΔ; $P$<0.05; placebo versus AOC) and not in controls (0.03±0.003 versus 0.03±0.004 cmΔ; $P$>0.05; placebo versus AOC). Shear rate (AUC) was not different between patients with COPD and controls (28 776±3305 versus 31 563±2797 $s^{-1}$; $P$>0.05; COPD versus control), and the AOC had no significant effect on shear rate in either group (COPD: 31 308±3586; control: 31 902±3034 $s^{-1}$; $P$>0.05). Similarly, RH (AUC) was not different between groups (516±68 versus 590±65 mL; $P$>0.05;

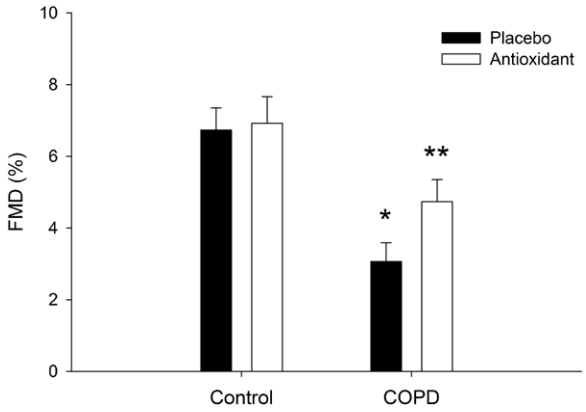


**Figure 1.** Flow-mediated dilation (FMD), expressed as peak relative change in patients with chronic obstructive pulmonary disease (COPD) and age- and sex-matched controls under placebo and antioxidant conditions. *$P$<0.05 COPD vs control placebo condition, **$P$<0.05 placebo vs antioxidant in COPD only.

COPD versus control, respectively), and the AOC had no effect on the RH (AUC) in either group (564±75 versus 582±68 mL; $P$>0.05; COPD versus control, respectively).

To account for potential individual differences in shear rate and thus the stimulus for FMD, the FMD data were normalized for the shear stimulus (FMD/shear rate, AUC). Patients with COPD again exhibited reduced vascular function (0.12±0.03 versus 0.25±0.03 FMD/shear rate; $P$<0.05; COPD versus control), which was significantly improved after acute ingestion of the oral AOC in patients with COPD ($P$<0.05) but not in controls (0.26±0.06 versus 0.28±0.04 FMD/shear rate; $P$>0.05; COPD versus control). Time to peak vasodilation did not differ between control and patients with COPD and was unaffected by the AOC in both groups.

Post hoc analysis of the subject characteristics revealed a significantly greater incidence of hypertension in patients with COPD. With the potential that this specific comorbidity in patients with COPD could have influenced their vascular function, the data were reanalyzed in 3 different ways to determine whether hypertension, per se, played a role. In the first approach, in addition to age and sex matching, the groups were matched for incidence of hypertension (dropping 10 patients with hypertension in the COPD group and 10 patients without hypertension in the healthy group), leaving n=7 hypertensives in each group of 20 subjects (β still >0.8 for all major variables). This approach yielded the same initial result where patients with COPD exhibited significantly reduced FMD compared with controls (absolute [0.009±0.002 versus 0.03±0.002 cmΔ], relative [2.6±0.6 versus 7.7±0.8%FMD], or normalized to shear rate [0.1063±0.03 versus 0.3005±0.05 $s^{-1}$], all $P$<0.05), which was improved after AOC ingestion in the COPD group only, abolishing the difference between the 2 groups. The second approach, filtering out all subjects with hypertension leaving 13 patients with COPD and 23 age-matched controls with no incidence of hypertension, resulted in the same findings: a significantly blunted FMD (absolute [0.0107±0.002 versus 0.0283±0.003 cmΔ], relative [2.6±0.6 versus 6.6±0.7%FMD], or normalized to shear rate [0.09±0.02 versus 0.21±0.03 $s^{-1}$]; all $P$<0.05) in patients with COPD compared with the healthy controls. The third approach, using an ANCOVA with hypertensive status as a covariate and all subjects included (n=60), revealed that the contribution of hypertensive status to basal FMD was not significant (partial $\eta^2$=0.013; $P$=0.40). Regardless of which approach/group of patients with COPD was examined, there was no statistically significant relationship between the indices of COPD severity (Global Initiative for Chronic Obstructive Lung Disease classification, forced expiratory volume$_1$/forced vital capacity ratio, % predicted forced expiratory volume$_1$, and so on) and AOC-induced vascular function improvement.

## Pulse Wave Velocity

Under the placebo condition, patients with COPD exhibited significantly higher PWV (14±1 versus 11±2 m·$s^{-1}$; $P$<0.05; COPD versus controls, respectively; Figure 2). The AOC significantly reduced PWV in patients with COPD (14±1 versus 11±1 m·$s^{-1}$; $P$<0.05; control versus AOC, respectively) but not in controls (11±2 versus 10±1 m·$s^{-1}$; $P$>0.05; control versus AOC, respectively).

## Blood Assays

Analysis of the patients' blood revealed lower initial levels of vitamin C (10±1 versus 14±1 μg/mL; $P$<0.05; COPD versus control), whereas the plasma levels of both groups were significantly ($P$<0.05; placebo versus AOC) increased after AOC ingestion, and patients with COPD still exhibited lower levels of vitamin C even after ingestion of the AOC (18±2 versus 24±2 μg/mL; $P$<0.05; COPD versus control; Figure 3). Global antioxidant capacity assessed using the ferric reducing ability of plasma was similar at baseline (1.0±0.05 versus 1.0±0.05 mmol/L; $P$<0.05; COPD versus control) and was significantly ($P$<0.05, placebo versus AOC) increased in both groups after AOC ingestion (1.2±0.06 versus 1.1±0.05 mmol/L; $P$<0.05; COPD versus control, respectively; Figure 3). Superoxide dismutase activity was lower in patients at baseline (5.1±0.2 versus 8.6±0.7 U/mL; $P$<0.05; COPD versus control) and only significantly increased in patients with COPD after AOC ingestion ($P$<0.05). Despite this AOC-induced increase, superoxide

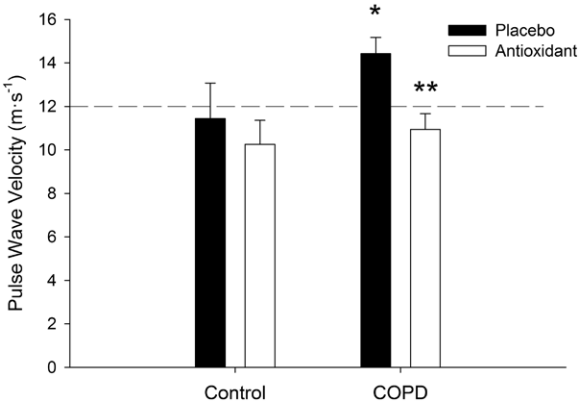


**Figure 2.** Carotid-radial pulse wave velocity under placebo and antioxidant conditions in both patients with chronic obstructive pulmonary disease (COPD), and age- and sex-matched controls. *$P$<0.05 COPD vs control placebo condition, **$P$<0.05 placebo vs antioxidant in COPD only. Dashed line indicates the recommended 12 m·$s^{-1}$ cutoff, as established by the Reference Values for Arterial Stiffness Collaboration, indicating elevated risk for cardiovascular disease.[45]

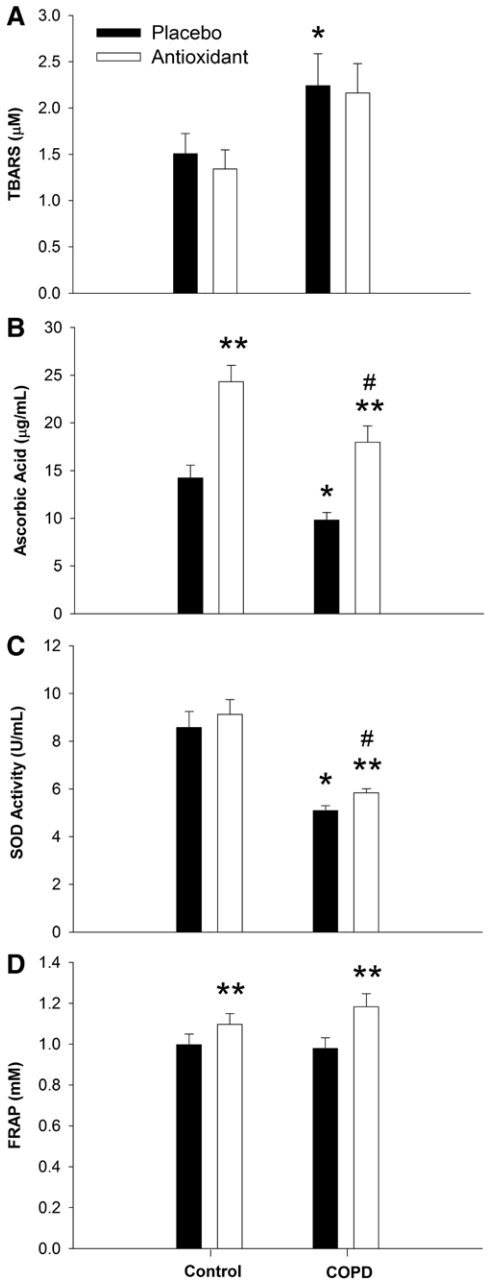


**Figure 3.** Oxidative stress (**A**) and antioxidant (**B–D**) assessments under placebo and antioxidant conditions in patients with chronic obstructive pulmonary disease (COPD) and age- and sex-matched controls. *$P$<0.05 control vs COPD placebo condition, **$P$<0.05 within group, #$P$<0.05 control vs COPD antioxidant condition.

dismutase (SOD) activity remained lower in the patients compared with controls (5.8±0.2 versus 9.1±0.6 U/mL; $P$<0.05; COPD versus control; Figure 3). Oxidative stress as assessed by thiobarbituric acid reactive substances (TBARS; an index of lipid peroxidation) was significantly higher in patients with COPD (2.2±0.3 versus 1.5±0.2 μmol/L; $P$<0.05; COPD versus control) and was unchanged ($P$>0.05) in either group after AOC ingestion (2.2±0.2 versus 1.3±0.2 μmol/L; $P$>0.05; COPD versus control; Figure 3). In a subset of the patients with COPD (n=16), free radical levels, directly assessed by EPR spectroscopy, were significantly reduced after AOC ingestion (8.5±2 versus 3.8±1 AU; $P$<0.05; placebo versus AOC). Regarding inflammation, the complete blood count obtained during the placebo trial revealed significantly elevated leukocytes (7.4±0.6 versus 5.6±0.3 K/μL), neutrophils (4.8±0.5 versus 3.4±0.3 K/μL), and monocytes (0.58±0.04 versus 0.46±0.03 K/μL) in patients with COPD compared with controls. However, although statistically different, these elevated values were still within the normal range (Table 1).

## Discussion

The goal of this study was to determine the role of redox balance in the vascular dysfunction associated with COPD. Patients with COPD did, indeed, display impaired vascular function, as assessed by FMD and PWV, compared with age-matched controls. Blood analyses revealed greater basal oxidative stress (TBARS) and lower endogenous antioxidant capacity (ascorbic acid and SOD) in patients with COPD compared with controls. After AOC ingestion, both groups displayed a similar increase in plasma ascorbic acid and total antioxidant capacity, indicating an equivalent initial effect of the AOC. Under these conditions, the differences in FMD and PWV between patients with COPD and controls were mitigated. Therefore, collectively, these data reveal that patients with COPD exhibit an altered redox balance that seems to negatively affect vascular function and stiffness, likely predisposing this patient population to greater risk and incidence of CVD.

### Baseline Vascular Dysfunction in Patients With COPD

In agreement with previous work,[18] using current methodologies for the assessment of FMD,[40] this study has demonstrated that patients with COPD are characterized by reduced vascular function, which is likely specific to the endothelium. In support of this contention, although not assessed in the current study, others have determined that there was no difference between controls and patients with COPD in terms of endothelium-independent dilation using sublingual nitroglycerin[17] or intra-arterial infusion of sodium nitroprusside and verapamil.[20] However, it is also important to note that not all agree that impaired endothelium-dependent dilation is an obligatory component of COPD[20] and may actually depend on exacerbation status.[17] Interestingly, another vascular assessment performed in the current study, RH, was not different between patients with COPD and controls or as a result of ingestion of the AOC. RH, which reflects microvascular responsiveness, seems to be mediated through both endothelium-dependent and endothelium-independent mechanisms[46] but also seems to be predictive of CV events.[47] Thus, a differential response between conduit artery (FMD) and microvascular function (RH) is not surprising and is in agreement with previous work,[17,18] suggesting that RH alone may not be as sensitive as FMD. However, because RH plays an important role as a component of shear rate, the stimulus for FMD,[48,49] normalizing FMD for the increase in the shear rate, evoked by cuff occlusion and release, can be an important consideration, although normalizing FMD to shear rate had no effect on the interpretation of the current findings.

In support of the current FMD data, revealing attenuated vascular function in patients with COPD, the PWV assessment

independently revealed significantly higher carotid-radial PWV in these patients compared with controls under the placebo condition. Of note, placebo PWV in the patients was above the recommended 12 $m \cdot s^{-1}$ as established by the Reference Values for Arterial Stiffness Collaboration, indicating elevated risk for CVD,[45] which was significantly reduced after acute AOC ingestion. Although PWV is traditionally thought of as purely an estimate of vascular stiffness, more recent evidence suggests that PWV may be related to endothelial function,[50–52] which may explain the AOC-mediated reduction in PWV. Taken together, reduced FMD and elevated PWV provide significant evidence of vascular dysfunction in patients with COPD, which likely contributes to the elevated CVD risk and prevalence in this population.[4–8]

## Mechanisms of Vascular Dysfunction in Patients With COPD

According to the current findings, the mechanisms responsible for vascular dysfunction in patients with COPD seem to be partly mediated by an alteration in the redox balance by an attenuated antioxidant capacity and elevated oxidative stress because the oral AOC improved vascular function only in those with COPD. Interestingly, in contrast to our previous work that focused on aging, in which the elderly exhibited a significant improvement in FMD after ingestion of the AOC,[33] the relatively old subjects in the current study were not affected by the AOC. However, it is important to note that the control group for patients with COPD were, on average, actually half a decade younger than the subjects in the previous study and contained individuals as young as 36 years of age who likely do not benefit from or may even respond negatively to such exogenous antioxidant treatment.[33] In support of this interpretation, the placebo FMD of the control group in the current study was ≈7%, whereas the FMD of the aged group in the article published by Wray et al[33] was ≈5%; thus, there is less vascular dysfunction in the current control group likely attributable to a relatively younger cohort, albeit still lower vascular function than young individuals. However, more importantly, the current study reveals that, in comparison with age-matched controls, patients with COPD exhibit reduced vascular function, which can be restored after ingestion of the AOC, suggestive of redox imbalance.

Previous work suggests that antioxidant capacity, as measured by Trolox equivalent antioxidant capacity, was reduced in patients with COPD, resulting in greater superoxide levels in the blood.[53] Numerous mechanisms could contribute to this reduced total antioxidant capacity, such as reduced activity of superoxide dismutase, catalase, and glutathione peroxidase.[36,54,55] In the current study, basal levels of factors contributing to the antioxidant defense system, including superoxide dismutase activity and vitamin C, were lower in patients with COPD compared with controls (Figure 3). In parallel, TBARS, which estimates lipid peroxidation, a footprint of oxidative stress, was elevated in patients with COPD (Figure 3) and is in agreement with previous literature suggesting that elevated oxidative stress is a characteristic of COPD.[28] Although we did not see an effect of the acute AOC on TBARS in either group, this lack of an effect agrees with previous work.[35,56] Specifically, such acute treatments may reduce free radicals either through the inhibition of pro-oxidant enzymes (nicotinamide adenine dinucleotide phosphate oxidase or xanthine oxidase) or through direct molecular quenching, resulting in functional changes, but there is a delay in terms of when the downstream effect of the oxidative stress can be detected (eg, TBARS). The EPR data reported here suggest a reduction in free radicals with ingestion of the AOC in patients with COPD, but with such an approach we cannot ascertain all of the potential mechanisms or the exact origin of the free radicals. Future studies might consider the use of an inhaled formulation to better target the likely source of the inflammation and oxidative stress in the lungs.[4] Considering both the antioxidant status and footprint of oxidative stress (TBARS), the patients with COPD seem to have an altered redox state because of a reduced antioxidant and pro-oxidant environment.

An oxidant imbalance contributes to greater levels of the free radicals superoxide and peroxynitrite, both potent endogenous competitors to the endothelial NO-vascular smooth muscle pathway.[57,58] Superoxide binds avidly with NO, reducing NO bioavailability, which increases the levels of peroxynitrite. Subsequently, peroxynitrite, itself, can oxidize tetrahydrobiopterin, an essential cofactor for endothelial NO synthase, leading to the uncoupling of endothelial NO synthase that also reduces levels of NO via reduced production.[57,59,60] In vitro evidence suggests that vitamin C and likely other antioxidants are capable of decreasing the oxidation of tetrahydrobiopterin and preventing the uncoupling of endothelial NO synthase.[57,61] It is likely that the reduced antioxidant status of patients with COPD resulted in greater free radical–mediated reductions in NO bioavailability either directly through the interaction of superoxide with NO or indirectly through oxidation of tetrahydrobiopterin, which contributed to the blunted endothelial-mediated FMD. In support of the contention that elevated free radicals are mediating the vascular dysfunction in patients with COPD, in the current study there was a significant increase in SOD in response to the AOC, likely because of a sparing of SOD, and a parallel reduction in total free radical signal, as measured by EPR in patients with COPD. Similarly, in agreement with the FMD findings, the PWV data indicated a COPD-related elevation in arterial stiffness, which was significantly reduced after ingestion of the AOC to within the values recommended for lower CVD risk (Figure 2).[45] These results highlight that PWV is also likely dependent on free radical/antioxidant redox balance and NO bioavailability, affecting vasomotor tone and ultimately arterial distensibility[50–52,62] in patients with COPD. As always, the effect of disease-specific medications on vascular function in a patient population such as this cannot be ruled out as playing a role in these findings.

## Perspectives

Patients with COPD in this study had elevated oxidative stress, lower vitamin C levels, and lower SOD activity, each likely contributing to impaired vascular function, as assessed by FMD and PWV. These findings contrast sharply with the age- and sex-matched controls. Also, of note, although not clearly demonstrating altered vascular function compared with other patients, 17 of the patients with COPD compared with 7 of the controls exhibited medically controlled hypertension.

Because the recruitment process was similar across all subjects, this difference highlights the elevated prevalence and risk of CVD among patients with COPD. It was certainly possible that the greater incidence of hypertension in patients with COPD could have explained the differences between the patients and age-matched controls; however, using several different statistical approaches, there was no evidence that this was the case. Therefore, based on the current findings, it seems reasonable to propose that, in an attempt to improve vascular health and reduce CVD risk in patients with COPD, an increase in antioxidant capacity could be targeted perhaps by exogenous antioxidant supplementation or endogenously through exercise training,[63] thereby reducing free radicals and improving endothelial function. Although the success of the clinical trials using antioxidants as an intervention in CVD has been mixed,[64–68] it is important to note that most trials have used a single antioxidant and not a cocktail containing both water and fat-soluble vitamins as in the current study. The antioxidant-induced improvement in vascular function, assessed by FMD and PWV, observed in the current study is suggestive of a reduction in the risk of CVD associated with COPD; however, a randomized, controlled trial with long-term supplementation is needed to confirm this hypothesis.

### Experimental Considerations

As with most studies, especially those performed on clinical populations, there are experimental considerations related to this work that need to be discussed. In patients with COPD, there were a disproportionate number of subjects with hypertension compared with controls, which proved not to influence the conclusions of the current study based on post hoc matching of subjects and ANCOVA with hypertension as a covariate. However, this does raise the question of what would be the effect of this AOC on subjects with a primary diagnosis of hypertension, but this is beyond the scope of the current study that focused on COPD. It should also be recognized that there was not a statistically significant relationship between the indices of COPD severity and AOC-induced vascular function improvement, implying that other uncontrolled variables such as occult obstructive sleep apnea, coronary artery disease, and the effect of several COPD-specific medications (eg, long-acting anticholinergics and β2-agonists) may have contributed to the variance in this study. A larger sample size and more proactive assessments of pathologies that do not come to light through health histories would be required to avoid these issues.

### Conclusions

Patients with COPD exhibited altered redox balance as evidenced by blunted endogenous antioxidant capacity and elevated oxidative stress compared with age- and sex-matched controls. Vascular function, as measured by FMD and PWV, was impaired in patients with COPD compared with controls. After an acute AOC, antioxidant capacity was improved in patients with COPD, which coincided with significant improvements in vascular function but not in controls. These results highlight the role of redox balance in vascular function of patients with COPD, which likely contributes to the disproportionate risk for CVD in this population.

## Acknowledgments

We thank the participants who volunteered for this study. We also thank Jia Zhao and Van Reese for performing the blood assays.

## Sources of Funding

We acknowledge financial support from National Institutes of Health (P01 HL-091830 awarded to R.S. Richardson), Department of Veterans Affairs (Merit grant E6910R awarded to R.S. Richardson), and the American Heart Association (10SDG3050006 awarded to R.A. Harris and 0835209N awarded to D.W. Wray). Advanced Fellowships in Geriatrics from the Department of Veterans Affairs supported S.J. Ives and M.A.H. Witman, in addition to a Career Development Award for J. McDaniel (E7560W).

## Disclosures

None.

## References

1. Divo M, Cote C, de Torres JP, Casanova C, Marin JM, Pinto-Plata V, Zulueta J, Cabrera C, Zagaceta J, Hunninghake G, Celli B; BODE Collaborative Group. Comorbidities and risk of mortality in patients with chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. 2012;186:155–161.
2. Macnee W, Maclay J, McAllister D. Cardiovascular injury and repair in chronic obstructive pulmonary disease. *Proc Am Thorac Soc*. 2008;5:824–833.
3. Konecny T, Somers K, Orban M, Koshino Y, Lennon RJ, Scanlon PD, Rihal CS. Interactions between COPD and outcomes after percutaneous coronary intervention. *Chest*. 2010;138:621–627.
4. Barnes PJ, Celli BR. Systemic manifestations and comorbidities of COPD. *Eur Respir J*. 2009;33:1165–1185.
5. Luppi F, Franco F, Beghé B, Fabbri LM. Treatment of chronic obstructive pulmonary disease and its comorbidities. *Proc Am Thorac Soc*. 2008;5:848–856.
6. Sin DD, Wu L, Man SF. The relationship between reduced lung function and cardiovascular mortality: a population-based study and a systematic review of the literature. *Chest*. 2005;127:1952–1959.
7. Matsuoka S, Yamashiro T, Diaz A, Estépar RS, Ross JC, Silverman EK, Kobayashi Y, Dransfield MT, Bartholmai BJ, Hatabu H, Washko GR. The relationship between small pulmonary vascular alteration and aortic atherosclerosis in chronic obstructive pulmonary disease: quantitative CT analysis. *Acad Radiol*. 2011;18:40–46.
8. Stone IS, Barnes NC, Petersen SE. Chronic obstructive pulmonary disease: a modifiable risk factor for cardiovascular disease? *Heart*. 2012;98:1055–1062.
9. Celermajer DS, Sorensen KE, Gooch VM, Spiegelhalter DJ, Miller OI, Sullivan ID, Lloyd JK, Deanfield JE. Non-invasive detection of endothelial dysfunction in children and adults at risk of atherosclerosis. *Lancet*. 1992;340:1111–1115.
10. Yeboah J, Crouse JR, Hsu FC, Burke GL, Herrington DM. Brachial flow-mediated dilation predicts incident cardiovascular events in older adults: the Cardiovascular Health Study. *Circulation*. 2007;115:2390–2397.
11. Yeboah J, Folsom AR, Burke GL, Johnson C, Polak JF, Post W, Lima JA, Crouse JR, Herrington DM. Predictive value of brachial flow-mediated dilation for incident cardiovascular events in a population-based study: the multi-ethnic study of atherosclerosis. *Circulation*. 2009;120:502–509.
12. Anderson TJ, Uehata A, Gerhard MD, Meredith IT, Knab S, Delagrange D, Lieberman EH, Ganz P, Creager MA, Yeung AC. Close relation of endothelial function in the human coronary and peripheral circulations. *J Am Coll Cardiol*. 1995;26:1235–1241.
13. Yeboah J, Sutton-Tyrrell K, Mcburnie M, Burke G, Herrington D, Crouse J. Association between brachial artery reactivity and cardiovascular disease status in an elderly cohort: the cardiovascular health study. *Atherosclerosis*. 2008;197:768–776.
14. Vlachopoulos C, Aznaouridis K, Stefanadis C. Prediction of cardiovascular events and all-cause mortality with arterial stiffness: a systematic review and meta-analysis. *J Am Coll Cardiol*. 2010;55:1318–1327.
15. Chirinos JA, Kips JG, Jacobs DR Jr, Brumback L, Duprez DA, Kronmal R, Bluemke DA, Townsend RR, Vermeersch S, Segers P. Arterial wave reflections and incident cardiovascular events and heart failure: MESA (Multiethnic Study of Atherosclerosis). *J Am Coll Cardiol*. 2012;60:2170–2177.

16. Weber T, Auer J, O'Rourke MF, Kvas E, Lassnig E, Berent R, Eber B. Arterial stiffness, wave reflections, and the risk of coronary artery disease. *Circulation*. 2004;109:184–189.
17. Ozben B, Eryüksel E, Tanrikulu AM, Papila-Topal N, Celikel T, Başaran Y. Acute exacerbation impairs endothelial function in patients with chronic obstructive pulmonary disease. *Turk Kardiyol Dern Ars*. 2010;38:1–7.
18. Eickhoff P, Valipour A, Kiss D, Schreder M, Cekici L, Geyer K, Kohansal R, Burghuber OC. Determinants of systemic vascular function in patients with stable chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. 2008;178:1211–1218.
19. Barr RG, Mesia-Vela S, Austin JHM, Basner RC, Keller BM, Reeves AP, Shimbo D, Stevenson L. Impaired flow-mediated dilation is associated with low pulmonary function and emphysema in ex-smokers. *Am J Respir Crit Care Med* 2007;176:1200–1207.
20. Maclay JD, McAllister DA, Mills NL, Paterson FP, Ludlam CA, Drost EM, Newby DE, Macnee W. Vascular dysfunction in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. 2009;180:513–520.
21. Maclay JD, McAllister DA, Rabinovich R, Haq I, Maxwell S, Hartland S, Connell M, Murchison JT, van Beek EJ, Gray RD, Mills NL, Macnee W. Systemic elastin degradation in chronic obstructive pulmonary disease. *Thorax*. 2012;67:606–612.
22. Vivodtzev I, Minet C, Wuyam B, Borel JC, Vottero G, Monneret D, Baguet JP, Lévy P, Pépin JL. Significant improvement in arterial stiffness after endurance training in patients with COPD. *Chest*. 2010;137:585–592.
23. Alford SK, van Beek EJ, McLennan G, Hoffman EA. Heterogeneity of pulmonary perfusion as a mechanistic image-based phenotype in emphysema susceptible smokers. *Proc Natl Acad Sci U S A*. 2010;107:7485–7490.
24. Arao T, Takabatake N, Sata M, Abe S, Shibata Y, Honma T, Takahashi K, Okada A, Takeishi Y, Kubota I. *In vivo* evidence of endothelial injury in chronic obstructive pulmonary disease by lung scintigraphic assessment of (123)I-metaiodobenzylguanidine. *J Nucl Med*. 2003;44:1747–1754.
25. Peinado VI, Barbera JA, Ramirez J, Gomez FP, Roca J, Jover L, Gimferrer JM, Rodriguez-Roisin R. Endothelial dysfunction in pulmonary arteries of patients with mild COPD. *Am J Physiol*. 1998;274(6 Pt 1):L908–L913.
26. Chao J, Wood JG, Gonzalez NC. Alveolar macrophages initiate the systemic microvascular inflammatory response to alveolar hypoxia. *Respir Physiol Neurobiol*. 2011;178:439–448.
27. Sabit R, Shale DJ. Vascular structure and function in chronic obstructive pulmonary disease: a chicken and egg issue? *Am J Respir Crit Care Med*. 2007;176:1175–1176.
28. Folchini F, Nonato NL, Feofiloff E, D'Almeida V, Nascimento O, Jardim JR. Association of oxidative stress markers and C-reactive protein with multidimensional indexes in COPD. *Chron Respir Dis*. 2011;8:101–108.
29. Eagan TM, Ueland T, Wagner PD, Hardie JA, Mollnes TE, Damås JK, Aukrust P, Bakke PS. Systemic inflammatory markers in COPD: results from the Bergen COPD Cohort Study. *Eur Respir J*. 2010;35:540–548.
30. Pinto-Plata VM, Müllerova H, Toso JF, Feudjo-Tepie M, Soriano JB, Vessey RS, Celli BR. C-reactive protein in patients with COPD, control smokers and non-smokers. *Thorax*. 2006;61:23–28.
31. Celli BR, Locantore N, Yates J, et al; ECLIPSE Investigators. Inflammatory biomarkers improve clinical prediction of mortality in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. 2012;185:1065–1072.
32. Noguera A, Batle S, Miralles C, Iglesias J, Busquets X, MacNee W, Agustí AG. Enhanced neutrophil response in chronic obstructive pulmonary disease. *Thorax*. 2001;56:432–437.
33. Wray DW, Nishiyama SK, Harris RA, Zhao J, McDaniel J, Fjeldstad AS, Witman MA, Ives SJ, Barrett-O'Keefe Z, Richardson RS. Acute reversal of endothelial dysfunction in the elderly after antioxidant consumption. *Hypertension*. 2012;59:818–824.
34. Wray DW, Uberoi A, Lawrenson L, Bailey DM, Richardson RS. Oral antioxidants and cardiovascular health in the exercise-trained and untrained elderly: a radically different outcome. *Clin Sci (Lond)*. 2009;116:433–441.
35. Witman MA, Fjeldstad AS, McDaniel J, Ives SJ, Zhao J, Barrett-O'Keefe Z, Nativi JN, Stehlik J, Wray DW, Richardson RS. Vascular function and the role of oxidative stress in heart failure, heart transplant, and beyond. *Hypertension*. 2012;60:659–668.
36. Berg D, Youdim MB, Riederer P. Redox imbalance. *Cell Tissue Res*. 2004;318:201–213.
37. Ijzerman RG, Serne EH, van Weissenbruch MM, de Jongh RT, Stehouwer CD. Cigarette smoking is associated with an acute impairment of microvascular function in humans. *Clin Sci (Lond)*. 2003;104:247–252.
38. Pauwels RA, Buist AS, Calverley PM, Jenkins CR, Hurd SS; GOLD Scientific Committee. Global strategy for the diagnosis, management, and prevention of chronic obstructive pulmonary disease. NHLBI/WHO Global Initiative for Chronic Obstructive Lung Disease (GOLD) Workshop summary. *Am J Respir Crit Care Med*. 2001;163:1256–1276.
39. Richardson RS, Donato AJ, Uberoi A, Wray DW, Lawrenson L, Nishiyama S, Bailey DM. Exercise-induced brachial artery vasodilation: role of free radicals. *Am J Physiol Heart Circ Physiol*. 2007;292:H1516–H1522.
40. Harris RA, Nishiyama SK, Wray DW, Richardson RS. Ultrasound assessment of flow-mediated dilation. *Hypertension*. 2010;55:1075–1085.
41. Zhang M, Bai Y, Ye P, Luo L, Xiao W, Wu H, Liu D. Type 2 diabetes is associated with increased pulse wave velocity measured at different sites of the arterial system but not augmentation index in a Chinese population. *Clin Cardiol*. 2011;34:622–627.
42. McEleavy OD, McCallum RW, Petrie JR, Small M, Connell JM, Sattar N, Cleland SJ. Higher carotid-radial pulse wave velocity in healthy offspring of patients with Type 2 diabetes. *Diabet Med*. 2004;21:262–266.
43. Baguet JP, Kingwell BA, Dart AL, Shaw J, Ferrier KE, Jennings GL. Analysis of the regional pulse wave velocity by Doppler: methodology and reproducibility. *J Hum Hypertens*. 2003;17:407–412.
44. Laurent S, Cockcroft J, Van Bortel L, Boutouyrie P, Giannattasio C, Hayoz D, Pannier B, Vlachopoulos C, Wilkinson I, Struijker-Boudier H; European Network for Non-invasive Investigation of Large Arteries. Expert consensus document on arterial stiffness: methodological issues and clinical applications. *Eur Heart J*. 2006;27:2588–2605.
45. Reference Values for Arterial Stiffness C. Determinants of pulse wave velocity in healthy people and in the presence of cardiovascular risk factors: 'Establishing normal and reference values'. *Eur Heart J*. 2010;31:2338–2350.
46. Meredith IT, Currie KE, Anderson TJ, Roddy MA, Ganz P, Creager MA. Postischemic vasodilation in human forearm is dependent on endothelium-derived nitric oxide. *Am J Physiol*. 1996;270(4 Pt 2):H1435–H1440.
47. Huang AL, Silver AE, Shvenke E, et al. Predictive value of reactive hyperemia for cardiovascular events in patients with peripheral arterial disease undergoing vascular surgery. *Arterioscler Thromb Vasc Biol*. 2007;27:2113–2119.
48. Mitchell GF, Parise H, Vita JA, Larson MG, Warner E, Keaney JF Jr, Keyes MJ, Levy D, Vasan RS, Benjamin EJ. Local shear stress and brachial artery flow-mediated dilation: the Framingham Heart Study. *Hypertension*. 2004;44:134–139.
49. Pyke KE, Dwyer EM, Tschakovsky ME. Impact of controlling shear rate on flow-mediated dilation responses in the brachial artery of humans. *J Appl Physiol (1985)*. 2004;97:499–508.
50. Naka KK, Tweddel AC, Doshi SN, Goodfellow J, Henderson AH. Flow-mediated changes in pulse wave velocity: a new clinical measure of endothelial function. *Eur Heart J*. 2006;27:302–309.
51. Kinlay S, Creager MA, Fukumoto M, Hikita H, Fang JC, Selwyn AP, Ganz P. Endothelium-derived nitric oxide regulates arterial elasticity in human arteries in vivo. *Hypertension*. 2001;38:1049–1053.
52. Wilkinson IB, Qasem A, McEniery CM, Webb DJ, Avolio AP, Cockcroft JR. Nitric oxide regulates local arterial distensibility in vivo. *Circulation*. 2002;105:213–217.
53. Rahman I, Morrison D, Donaldson K, MacNee W. Systemic oxidative stress in asthma, COPD, and smokers. *Am J Respir Crit Care Med*. 1996;154(4 Pt 1):1055–1060.
54. Madamanchi NR, Vendrov A, Runge MS. Oxidative stress and vascular disease. *Arterioscler Thromb Vasc Biol*. 2005;25:29–38.
55. Arthur JR. The glutathione peroxidases. *Cell Mol Life Sci*. 2000;57:1825–1835.
56. Silvestro A, Scopacasa F, Oliva G, de Cristofaro T, Iuliano L, Brevetti G. Vitamin C prevents endothelial dysfunction induced by acute exercise in patients with intermittent claudication. *Atherosclerosis*. 2002;165:277–283.
57. Kuzkaya N, Weissmann N, Harrison DG, Dikalov S. Interactions of peroxynitrite, tetrahydrobiopterin, ascorbic acid, and thiols: implications for uncoupling endothelial nitric-oxide synthase. *J Biol Chem*. 2003;278:22546–22554.
58. Somers MJ, Mavromatis K, Galis ZS, Harrison DG. Vascular superoxide production and vasomotor function in hypertension induced by deoxycorticosterone acetate-salt. *Circulation*. 2000;101:1722–1728.
59. Schmidt TS, Alp NJ. Mechanisms for the role of tetrahydrobiopterin in endothelial function and vascular disease. *Clin Sci (Lond)*. 2007;113:47–63.
60. Crabtree MJ, Tatham AL, Al-Wakeel Y, Warrick N, Hale AB, Cai S, Channon KM, Alp NJ. Quantitative regulation of intracellular endothelial nitric-oxide synthase (eNOS) coupling by both tetrahydrobiopterin-eNOS stoichiometry and biopterin redox status: insights from cells

with tet-regulated GTP cyclohydrolase I expression. *J Biol Chem*. 2009;284:1136–1144.
61. Vásquez-Vivar J, Martásek P, Whitsett J, Joseph J, Kalyanaraman B. The ratio between tetrahydrobiopterin and oxidized tetrahydrobiopterin analogues controls superoxide release from endothelial nitric oxide synthase: an EPR spin trapping study. *Biochem J*. 2002;362(Pt 3):733–739.
62. Ramsey MW, Goodfellow J, Jones CJ, Luddington LA, Lewis MJ, Henderson AH. Endothelial control of arterial distensibility is impaired in chronic heart failure. *Circulation*. 1995;92:3212–3219.
63. Donato AJ, Uberoi A, Bailey DM, Wray DW, Richardson RS. Exercise-induced brachial artery vasodilation: effects of antioxidants and exercise training in elderly men. *Am J Physiol Heart Circ Physiol*. 2010;298:H671–H678.
64. Jialal I, Devaraj S. Antioxidants and atherosclerosis: don't throw out the baby with the bath water. *Circulation*. 2003;107:926–928.
65. Thomson MJ, Puntmann V, Kaski JC. Atherosclerosis and oxidant stress: the end of the road for antioxidant vitamin treatment? *Cardiovasc Drugs Ther*. 2007;21:195–210.
66. Stephens NG, Parsons A, Schofield PM, Kelly F, Cheeseman K, Mitchinson MJ. Randomised controlled trial of vitamin E in patients with coronary disease: Cambridge Heart Antioxidant Study (CHAOS). *Lancet*. 1996;347:781–786.
67. Hodis HN, Mack WJ, LaBree L, Mahrer PR, Sevanian A, Liu CR, Liu CH, Hwang J, Selzer RH, Azen SP; VEAPS Research Group. Alpha-tocopherol supplementation in healthy individuals reduces low-density lipoprotein oxidation but not atherosclerosis: the Vitamin E Atherosclerosis Prevention Study (VEAPS). *Circulation*. 2002;106:1453–1459.
68. Queiroz TM, Guimarães DD, Mendes-Junior LG, Braga VA. α-lipoic acid reduces hypertension and increases baroreflex sensitivity in renovascular hypertensive rats. *Molecules*. 2012;17:13357–13367.

## Novelty and Significance

### What Is New?

- This study has revealed that in comparison with healthy age-matched controls, vascular endothelial function is impaired and vascular stiffness elevated in patients with chronic obstructive pulmonary disease. This may help to explain the increased risk and prevalence of cardiovascular disease (CVD) in patients with chronic obstructive pulmonary disease. An acute oral antioxidant cocktail restored vascular function back to that of controls, highlighting the significant role that redox imbalance likely plays in this population.

### What Is Relevant?

- Vascular endothelial function and stiffness have been related to CVD, such as hypertension, coronary artery disease, or heart failure. Endothelial dysfunction has been demonstrated to precede the development of CVD. Restoring redox balance, via exogenous antioxidants, in patients with chronic obstructive pulmonary disease may reduce CVD-related morbidity and mortality in this population.

### Summary

Patients with chronic obstructive pulmonary disease exhibit impaired vascular endothelial function and elevated vascular stiffness, which seems to be mediated by a redox imbalance and may help to explain the increased risk of CVD in this population.

# Vascular Dysfunction and Chronic Obstructive Pulmonary Disease: The Role of Redox Balance

Stephen J. Ives, Ryan A. Harris, Melissa A.H. Witman, Anette S. Fjeldstad, Ryan S. Garten, John McDaniel, D. Walter Wray and Russell S. Richardson

*Hypertension.* 2014;63:459-467; originally published online December 9, 2013;
doi: 10.1161/HYPERTENSIONAHA.113.02255
*Hypertension* is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231

Print ISSN: 0194-911X. Online ISSN: 1524-4563