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ORIGINAL RESEARCH

Translational Research of the Acute Effects of Negative Emotions on Vascular Endothelial Health: Findings From a Randomized Controlled Study

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BACKGROUND: Provoked anger is associated with an increased risk of cardiovascular disease events. The underlying mechanism linking provoked anger as well as other core negative emotions including anxiety and sadness to cardiovascular disease remain unknown. The study objective was to examine the acute effects of provoked anger, and secondarily, anxiety and sadness on endothelial cell health.

METHODS AND RESULTS: Apparently healthy adult participants (n=280) were randomized to an 8-minute anger recall task, a depressed mood recall task, an anxiety recall task, or an emotionally neutral condition. Pre−/post-assessments of endothelial health including endothelium-dependent vasodilation (reactive hyperemia index), circulating endothelial cell-derived microparticles (CD62E+, CD31+/CD42−, and CD31+/Annexin V+) and circulating bone marrow-derived endothelial progenitor cells (CD34+/CD133+/kinase insert domain receptor+ endothelial progenitor cells and CD34+/kinase insert domain receptor+ endothelial progenitor cells) were measured. There was a group×time interaction for the anger versus neutral condition on the change in reactive hyperemia index score from baseline to 40 minutes (P=0.007) with a mean±SD change in reactive hyperemia index score of 0.20±0.67 and 0.50±0.60 in the anger and neutral conditions, respectively. For the change in reactive hyperemia index score, the anxiety versus neutral condition group by time interaction approached but did not reach statistical significance (P=0.054), and the sadness versus neutral condition group by time interaction was not statistically significant (P=0.160). There were no consistent statistically significant group×time interactions for the anger, anxiety, and sadness versus neutral condition on endothelial cell-derived microparticles and endothelial progenitor cells from baseline to 40 minutes.

CONCLUSIONS: In this randomized controlled experimental study, a brief provocation of anger adversely affected endothelial cell health by impairing endothelium-dependent vasodilation.

Key Words: atherosclerosis ■ endothelium ■ psychosocial factors ■ vascular health

Atherosclerosis is a diffuse disease characterized by the deposition of lipid and other blood-borne elements within the arterial wall.[1] Evidence indicates that disruption of an arterial atherosclerotic plaque and subsequent thrombus formation is responsible for the onset of cardiovascular disease (CVD) events.[2] Cardiovascular research efforts have been directed toward the identification of early underlying factors that initiate the pathways contributing to atherosclerosis.[2]

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RESEARCH PERSPECTIVE

What Is New?

- The experience of a negative emotion including anger, anxiety, and sadness is common and associated with an increased risk of cardiovascular disease events. There are scarce data on the effects of provoked anger, anxiety, and sadness on vascular endothelial health.
- In the current study, the provocation of anger adversely affected endothelial health by impairing endothelium-dependent vasodilation. There were no statistically significant adverse effects of provoked anxiety and sadness on endothelium-dependent vasodilation.

What Question Should Be Addressed Next?

- Negative emotions should not be grouped together mechanistically in their associations with increased cardiovascular disease risk. Future investigation into the mechanisms underlying the link between anger and endothelial dysfunction may help identify effective specific intervention targets for a large proportion of individuals at increased cardiovascular disease risk.

Nonstandard Abbreviations and Acronyms

EC	endothelial cell
EDV	endothelium-dependent vasodilation
EMP	EC-derived microparticle
EPC	endothelial progenitor cell
RHI	reactive hyperemia index

Since Friedman and Rosenman first proposed in 1959 that individuals with a behavior pattern defined as highly competitive, ambitious, work driven, time conscious, and aggressive (ie, the type A behavior pattern) were at an increased risk of CVD events,[3] there has been an immense interest in investigating the associations between psychosocial factors and incident CVD events.[4] The experience of negative emotions is associated with an increased risk of incident CVD events, independent of traditional risk factors.[5–7] Among the best-studied negative emotions for triggering CVD events is anger, with population-based studies consistently demonstrating that the acute experience of anger is associated with an increased risk of CVD event onset.[6–11] The mechanism(s) by which the experience of anger acutely affects the pathways that underlie atherosclerosis development and progression remain to be fully characterized.

The endothelium is a key regulator of vascular homeostasis. Vascular endothelial cells (ECs) play essential roles in maintaining vascular tone and the integrity of blood vessels.[12] Evidence suggests that endothelial dysfunction is an early pathogenic process underlying atherosclerosis development and CVD event onset.[13,14] Several studies have demonstrated that a mental stress task such as mental arithmetic or public speaking impairs endothelium-dependent vasodilation (EDV).[15–18] In laboratory studies, reactivity to mental stress tasks is often equated with experiencing a negative emotion such as anger. However, these tasks elicit performance-related reactivity that may not provoke a specific negative emotion, just as life stressor may produce a variety of different emotions, depending on individual and contextual factors. We have previously shown in a small, nonrandomized study that an anger recall task, which promotes an acute reexperience of a prior event that provoked anger, acutely affected EC health by impairing EDV, injuring ECs and disrupting EC reparative capacity.[19] Finally, in addition to anger-provoked CVD events, there is some evidence from population-based studies that indicates that the acute experience of anxiety and sadness may also trigger CVD events.[8,20] However, there are scarce data on the effects of provoked anxiety and sadness on EC health.

The overall aim of this study was to examine primarily the acute effects of provoked anger, and secondarily, anxiety and sadness on EC health. Our primary hypothesis was that compared with the neutral condition, the anger recall task would impair EDV, injure ECs, and reduce EC reparative capacity. We secondarily hypothesized that compared with the neutral condition, the anxiety, and separately, the sadness tasks have similar effects on EDV, EC injury, and EC reparative capacity.

METHODS

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Study Population

We conducted the National Heart, Lung, and Blood Institute-funded PUME (Putative Mechanisms Underlying Myocardial Infarction Onset and Emotions) study, which was a laboratory-based, randomized controlled experimental study.[21] The PUME study was a single-blind, between-subjects (ie, parallel arm) randomized study design in which 280 participants, enrolled from August 2013 to May 2017, were randomized to 1 of the 4 conditions: an anger recall task, an anxiety recall

task, a depressed mood recall task, and an emotionally neutral condition (Clinicaltrials.gov registration number: NCT01909895). Pre-/post-assessments of EC health using flow-mediated EDV, EC injury as represented by levels of circulating EC-derived microparticles (EMPs), and EC reparative capacity as represented by levels of circulating bone marrow-derived endothelial progenitor cells (EPCs) were conducted.

Participants were recruited from the community surrounding Columbia University Irving Medical Center. Eligible participants were 18 years or older and apparently healthy. Exclusion criteria included (1) any chronic medical condition including prevalent CVD (defined as physician-diagnosed coronary artery disease, coronary revascularization [eg, stent, angioplasty, coronary bypass surgery], stroke, transient ischemic attack, peripheral arterial disease, or heart failure) and traditional risk factors including history of hypertension, diabetes, dyslipidemia; (2) active smoking; (3) any current medication use including over-the-counter drugs and dietary supplements; or (4) history of psychosis, mood disorders, or personality disorder diagnoses. Written informed consent was obtained from all participants, and the study was approved by the Institutional Review Board of Columbia University Irving Medical Center. The Consolidated Standards of Reporting Trials diagram is shown in Figure 1. There were no serious or unexpected adverse events and no unanticipated problems in the study.

Procedures

The prior study protocol[21] and Data S1 describe the study protocol in detail. In brief, information about participant demographics and cardiovascular risk factors were obtained by self-administered standardized questionnaires and interview during the screening visit. Physical activity was assessed using the 7-item International Physical Activity Questionnaire–Short Form.[22] Alcohol consumption was assessed by a self-administered standardized questionnaire that examined alcohol use on a daily and weekly basis.

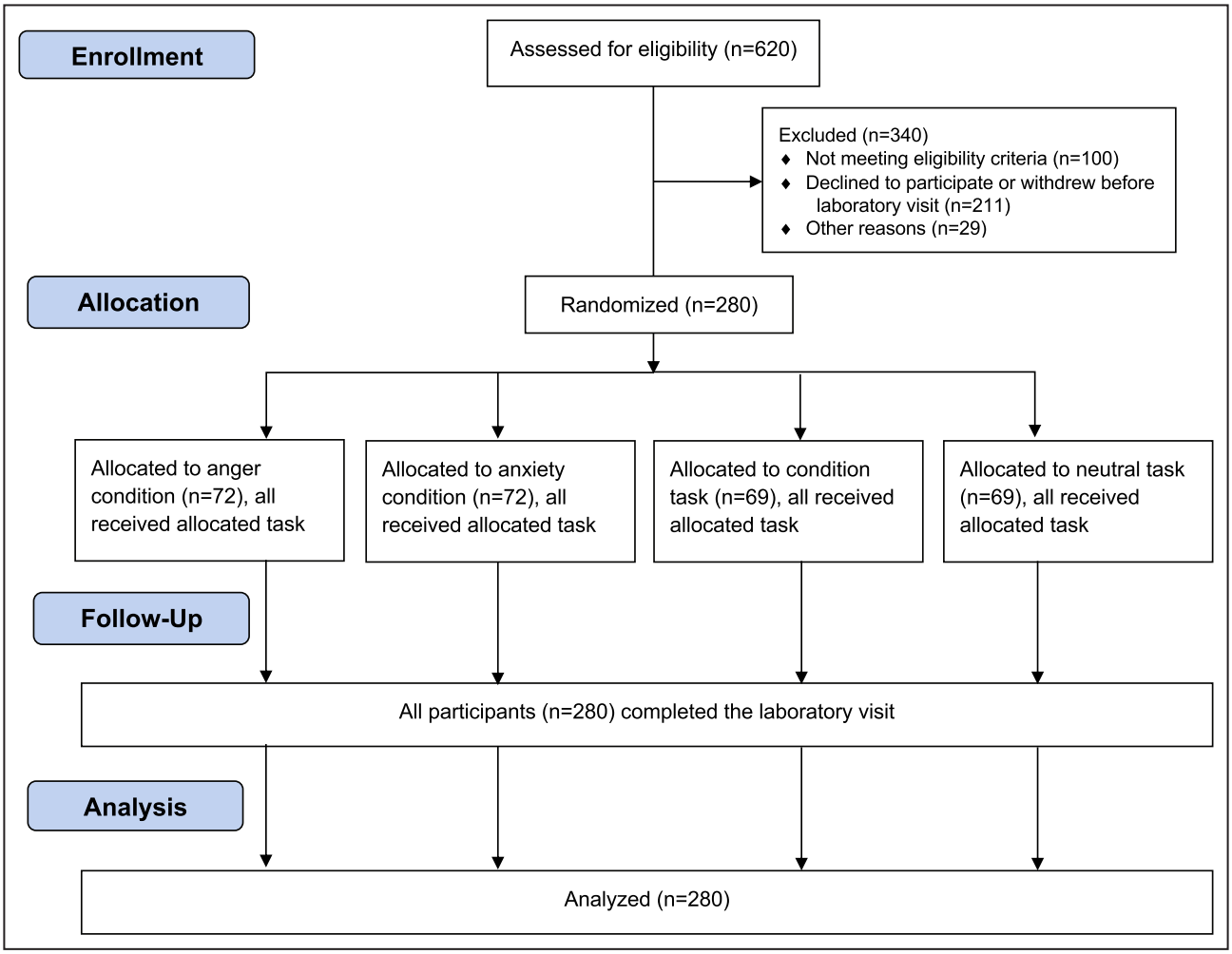

Figure 1. Consolidated Standards of Reporting Trials flow diagram.

Environmental exposure to tobacco was assessed using a questionnaire from the CARDIA (Coronary Artery Risk Development in Young Adults) study.[23]

Laboratory Visit

Participants arrived at the research laboratory at 8:30 AM and were escorted to a temperature-controlled study room and seated in a comfortable chair for the entire visit. The study procedures began before 9:30 AM. An appropriately sized cuff was placed on the nondominant upper arm for blood pressure (BP) measurement. A 20-gauge intravenous catheter was inserted into the antecubital vein of the dominant arm. Afterwards, a finger probe for the EndoPAT2000 device (Itamar Medical, Inc; ZOLL Medical Corporation) was placed on the first digit of each hand for the assessment of EDV.[21] A BP cuff was placed on the nondominant forearm for inducing reactive hyperemia for EDV testing. The participant was then instructed to relax for 30 minutes, during which they were not allowed to talk, use their phones, read any documents, or sleep. After this resting period, time point 1 (baseline) measures were obtained. Two BP with corresponding heart rate measurements were obtained 1 minute apart using a validated device (BpTru, Model BPM-200) and an appropriately sized cuff, and then EDV testing was conducted. Blood was drawn in the collection tubes including a citrated tube, an EDTA tube and a serum separate tube. Visual Analog Scale (VAS) ratings of anger, anxiety, and sadness were performed. After completion of these baseline measurements, the 8-minute negative emotion induction task or neutral condition was administered. The same measurements at baseline were repeated at 3 minutes (time point 2), 40 minutes (time point 3), 70 minutes (time point 4), and 100 minutes (time point 5) after the negative emotion induction task or neutral task was completed.

Negative Emotion Induction and Neutral Condition

After baseline measures were obtained, participants were randomized to 1 of 4 conditions (anger induction, anxiety induction, sadness induction, and neutral condition). The recall technique was used for provoking anger and anxiety, and the Velten mood induction technique was used for provoking sadness.[24–26] The recall technique consisted of asking the participant to recall relevant personal memories that would evoke either associated anger or anxiety over a period of 8 minutes. The Velten mood induction technique consisted of having the participant read descriptors that evoke sadness over a period of 8 minutes. For the neutral condition, which controlled for the potential effects of speech, participants were asked to count aloud by ones, starting with 1 and ending with 100, over and over, until 8 minutes had elapsed, with the participant choosing the pace of counting. After the task, the participants sat quietly in the chair, and were not allowed to talk, use their phones, read any documents, or sleep.

EC Health Measures

Details about EDV, EMPs, and EPCs have been provided in Data S1.

Endothelium-Dependent Vasodilation

EDV was defined as the reactive hyperemia index (RHI), assessed using EndoPAT2000 (Itamar Medical, Inc; ZOLL Medical Corporation). The primary outcome, RHI score, was calculated as the ratio of the average amplitude of the peripheral arterial tonometry signal over a 90- to 120-second period post deflation divided by the average amplitude of the peripheral arterial tonometry signal of a 2-minute period before cuff inflation.[27]

EC-Derived Microparticles

EC injury was assessed by measuring circulating EMPs using flow cytometry.[28–30] EMPs were defined as the number of particles with size <1.5 μm, which were positively labeled by CD62E+ (EMPs expressing CD62E); positively labeled by CD31 and negatively labeled by CD42 (CD31+/CD42- EMPs); and positively labeled by FITC-conjugated Annexin V (CD31+/Annexin V+ EMPs). The primary outcome was CD62E+ EMPs. Secondary outcomes were CD31+/CD42- EMPs and CD31+/Annexin V+ EMPs.

Endothelial Progenitor Cells

EC reparative capacity was assessed by measuring circulating EPCs using flow cytometry.[19,31–34] The percentages of the mononuclear lymphocytic populations that consist of CD34+/CD133+/kinase insert domain receptor (KDR)+ cells, and separately, CD34+/KDR+ cells were determined. The primary outcome was CD34+/CD133+/KDR+ cells, and the secondary outcome was CD34+/KDR+ cells.

The primary outcome measures were chosen a priori and were based on our prior nonrandomized study, which suggested that anger provocation decreased RHI score, increased CD62E+ EMPs, and decreased CD34+/CD133+/KDR+ cells.[19]

Hemodynamic Parameters

Systolic BP, diastolic BP, and heart rate, which were assessed in duplicate (as described previously) at each time point, were averaged.

Laboratory Testing

Blood obtained at the baseline time point was used to measure total cholesterol, triglycerides, high-density lipoprotein cholesterol, low-density lipoprotein cholesterol, and glucose.

Statistical Analysis

The analysis was based on a 4 (negative emotion induction/neutral task groups)×5 (time points) factorial design. To align with our a priori primary and secondary hypotheses, we focused the analyses on the contrast between the neutral condition versus each of the 3 emotion conditions. Based on our prior studies,[19,35] we focused on the first 3 time points as we expected that the impact of negative emotion induction may be greatest at time points 2 and 3 and would then begin to dissipate after time point 3. We analyzed the 5 time points to evaluate if the return of the endothelial measures to baseline would differ between the conditions. We used orthogonal polynomial contrast codes to represent the linear effect of time. We specified an unstructured variance–covariance matrix across the time points. The critical analysis was the group × time (linear) interaction. For a sample size of 280 participants, the study had >80% power to detect the estimated effects for the primary outcome measures: RHI score, CD62E+ EMPs, and CD34+/CD133+/KDR+ cells. Please see Data S1 for further details about the SDs and correlations used to determine the sample size. Finally, as the study was not powered for the secondary outcome measures, CD31+/CD42- EMPs, CD31+/Annexin V+ EMPs, and CD34+/KDR+ cells, these analyses were considered hypothesis-generating.

Sensitivity Analysis

The analyses were repeated after adjusting for the change in noncondition VAS ratings (ie, examining the effect of the anger induction task on EC health measures adjusting for the changes in VAS scores of anxiety and sadness). A P value of <0.05 was set as statistically significant.

RESULTS

Sample Characteristics

Table 1 shows the characteristics of the 280 study participants.

Effects of Provoked Anger, Anxiety, and Sadness on VAS Ratings

VAS anger, anxiety, and sadness ratings changed across the 5 time points with each of the negative emotion task conditions (Figure 2). There were group differences in the VAS anger, anxiety, and sadness ratings between the 4 conditions. The greatest increases in VAS anger, VAS anxiety, and VAS sadness rating were present in the anger condition, the anxiety condition, and the sadness condition, respectively.

Effects of Provoked Anger, Anxiety, and Sadness on RHI Score

Across the first 3 time points (baseline, 3 minutes, 40 minutes), there was an induction group×time interaction for the anger versus neutral condition on the change in RHI score from baseline to 40 minutes (Table 2). The greatest effect was observed at 40 minutes; the mean±SD change in RHI score was 0.20±0.67 and 0.50±0.60 in the anger and neutral conditions. There was no evidence of an anxiety versus neutral by time interaction or sadness versus neutral by time interaction on RHI score. The mean±SD change in RHI score in the anxiety and sadness conditions was 0.26±0.74 and 0.37±0.60, respectively, at 40 minutes. The condition by time interaction analyses across the 5 time points are provided in Table 2. In a sensitivity analysis, controlling for the change in noncondition VAS ratings as covariates in the analyses did not change the results.

Effects of Provoked Anger, Anxiety, and Sadness on EMPs and EPCs

Compared with the neutral condition, there was no evidence of an anger versus neutral by time interaction, anxiety versus neutral by time interaction, or sadness versus neutral by time interaction on the primary outcomes of CD62E+ EMPs and CD34+/CD133+/KDR EPCs (Table S1) across the first 3 time points. At 40 minutes, the mean±SD change in CD62E+ EMPs was –245.7±452.1, –207.1±427.6, –178.3±392.2, and –113.6±293.5 for the anger, anxiety, sadness, and neutral conditions, respectively. At 40 minutes, the mean±SD change in CD34+/CD133+/KDR EPCs was –0.0001±0.00073, 0.0000±0.00008, –0.0002±0.00124, and –0.0003±0.00120 for the anger, anxiety, sadness, and neutral conditions, respectively. The condition by time interaction analyses across the 5 time points for the primary outcome measures of EMPs and EPCs, and across the first 3 time points and 5 time points for the secondary outcome measures are provided in Table S1.

Effects of Provoked Anger, Anxiety, and Sadness on BP and Heart Rate

Compared with the neutral condition, there was an anger versus neutral by time interaction for the change in systolic BP and diastolic BP across the

Table 1. Sample Characteristics by Randomization Group

Characteristics	Anger condition (N=72)	Anxiety condition (N=70)	Sadness condition (N=69)	Neutral condition (N=69)
Age, y	26.3±9.2	25.9±5.2	26.1±7.7	26.8±6.8
Female sex, %	39 (54)	31 (44)	40 (58)	35 (50)
Hispanic or Latino, %	20 (28)	20 (29)	25 (36)	15 (22)
Race				
White, %	40 (44)	24 (34)	27 (39)	30 (44)
Black, %	15 (21)	9 (13)	6 (9)	8 (12)
Asian, %	6 (8)	16 (23)	14 (20)	16 (23)
Native Hawaiian or other Pacific Islander	0 (0)	1 (1)	0 (0)	0 (0)
American Indian or Alaska Native, %	0 (0)	1 (1)	0 (0)	0 (0)
More than 1 race, %	10 (14)	16 (23)	13 (19)	11 (16)
Unknown/not reported, %	5 (7)	3 (4)	9 (13)	4 (6)
Body mass index, kg/m^2	25.3±4.1	24.8±3.9	24.5±4.7	24.0±4.1
Total cholesterol, mg/dL	158.7±28.2	152.4±23.7	152.3±28.3	159.3±29.1
Triglycerides, mg/dL	77.3±42.3	70.8±25.2	66.2±28.9	68.7±26.4
High-density lipoprotein, mg/dL	54.7±14.2	55.1±12.5	55.0±15.5	56.8±16.8
Low-density lipoprotein, mg/dL	88.5±23.8	83.2±23.1	84.0±24.8	88.8±24.8
Glucose, mg/dL	86.9±7.9	86.8±6.5	86.6±7.3	87.2±6.5
Median (25th percentile, 75th percentile) combined total physical activity, metabolic equivalents, min/wk	2160 (1109, 3341)	2016 (1069, 3652)	2186 (837, 4267)	2133 (1125, 4092)
Heavy alcohol consumption*, %	9 (12.7)	2 (2.9)	9 (13.6)	7 (10.1)
Exposure to environmental tobacco smoke at home, %	5 (7.0)	5 (7.1)	6 (9.0)	10 (14.5)
Blood pressure (baseline)				
Systolic blood pressure, mm Hg	105.9±10.3	105.0±11.9	105.7±8.3	105.6±11.1
Diastolic blood pressure, mm Hg	68.6±7.8	68.1±8.2	68.5±7.0	68.6±8.3
Heart rate (baseline), beats per minute	64.3±7.9	64.9±11.6	64.1±9.7	61.8±10.2
Reactive hyperemia index score (baseline), unitless	2.44±0.78	2.41±0.90	2.31±0.82	2.22±0.77
EMPs expressing CD62E (baseline), #/μL	884.4±612.4	978.3±551.6	907.7±503.9	799.6±361.4
EMPs expressing CD31 (baseline), #/μL	549.3±279.4	621.4±318.0	528.2±265.1	599.5±307
EMPs expressing CD31 and Annexin V (baseline), #/μL	182.9±109.4	233.2±165.0	185.6±88.0	201.8±102.4
CD34+/CD133+/KDR+ EPCs (baseline), %	0.0010±0.00130	0.0011±0.00176	0.0011±0.00153	0.0009±0.00124
CD34+/KDR+ EPCs (baseline), %	0.0234±0.02857	0.0210±0.01719	0.0253±0.03013	0.0228±0.02112
VAS score for anger (baseline), unitless	0.68±1.45	0.56 ±1.00	0.67±1.07	0.74±1.57
VAS score for anxiety (baseline), unitless	1.53±1.94	1.66±1.79	1.74±1.74	1.61±1.47
VAS score for sadness (baseline), unitless	0.78±1.38	0.94±1.39	1.04±1.60	1.12±1.65

Data are expressed as number (percentage) or mean±SD, unless otherwise indicated.
EMP indicates endothelial cell-derived microparticle; EPC, endothelial progenitor cell; KDR, kinase insert domain receptor; and VAS, visual analog scale.
*Heavy alcohol consumption was defined as consuming >14 drinks per week for men and >7 drinks per week for women.

first 3 time points and the 5 time points (Table S2; Figure S1). Similar results were seen for the anxiety induction task.

There was no evidence of a sadness versus neutral by time interaction for the change in systolic BP and diastolic BP across time. At 3 minutes, the mean±SD

Figure 2. Visual analog scale (VAS) scores for anger (upper panel), anxiety (middle panel), and sadness (lower panel) in the four randomized conditions.
Data are expressed as mean VAS scores at each time point. VAS anger ratings changed across the 5 time points in the anger condition, the anxiety condition, and the sadness condition (P<0.001). VAS anxiety ratings changed across the 5 time points in the anxiety condition, the anger condition, and the sadness condition (P<0.001). VAS sadness ratings changed across the 5 time points in the sadness condition, the anger condition, and the anxiety condition (P<0.001). There were statistically significant group differences (P<0.001) in the VAS anger ratings between the 4 conditions. There were also statistically significant group differences in the VAS anxiety ratings (P=0.001) and VAS sadness ratings (P<0.001) between the 4 conditions.

change in systolic BP/diastolic BP was 9.4±7.1/5.6±7.8 mm Hg, 9.3±10.4/5.2±8.5 mm Hg, 3.2±5.6/2.6±5.3 mm Hg, and 1.6±4.6/1.8±4.1 mm Hg for the anger, anxiety, sadness, and neutral conditions, respectively. Finally, compared with the neutral condition, there was no evidence of a task by time interaction for the

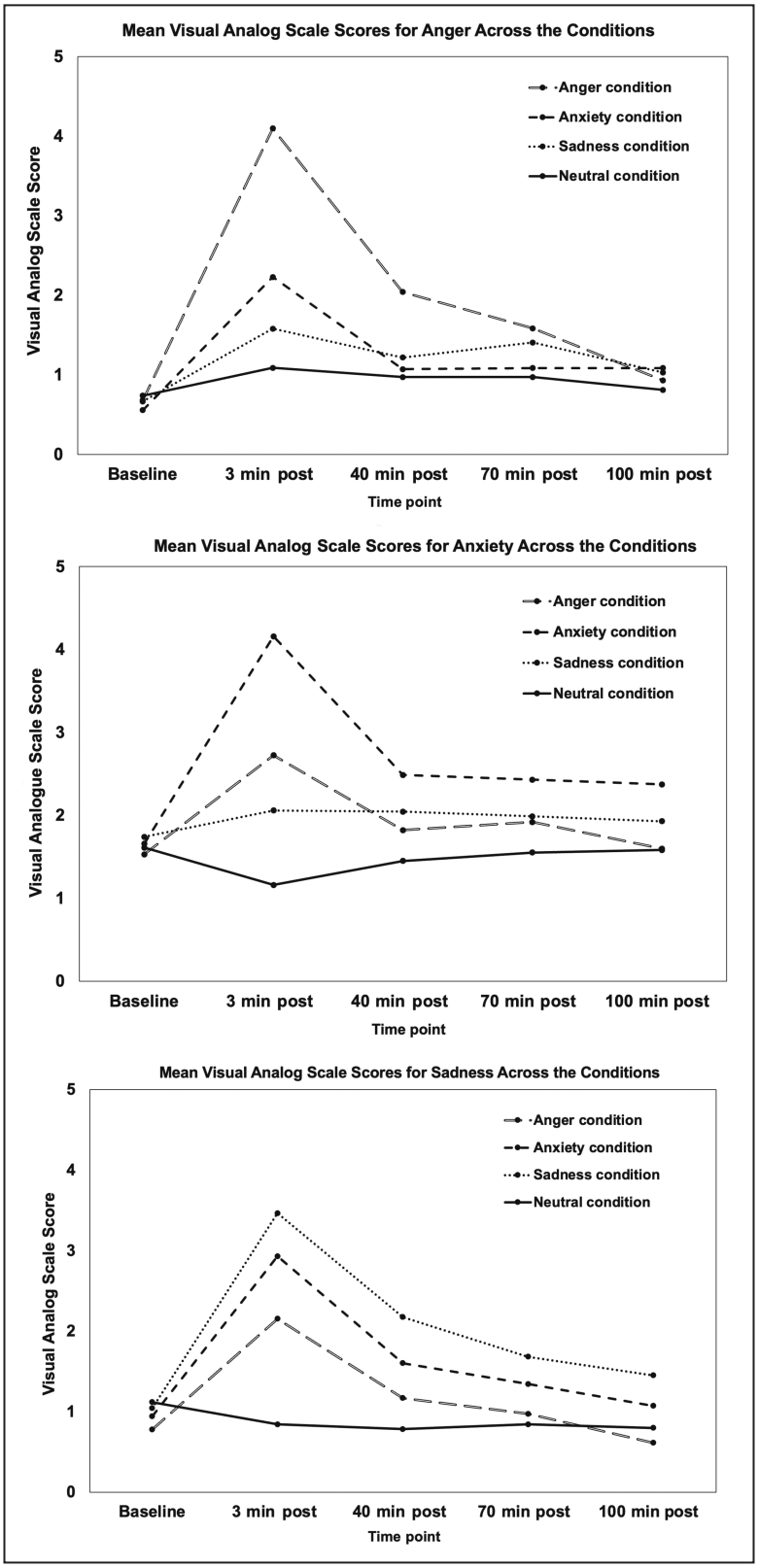

Table 2. Change in Reactive Hyperemia Index From Time Point 1 (Baseline) by Randomized Condition Over Time

	Time point 2 (3 min)	Time point 3 (40 min)	Time point 4 (70 min)	Time point 5 (100 min)	Condition×time *P* value (baseline to time point 3)	Condition×time *P* value (baseline to time point 5)
Anger induction task	0.12±0.65	0.20±0.67	0.38±0.90	0.44±0.84	0.007*	0.5
Anxiety induction task	0.11±0.70	0.26±0.74	0.31±0.83	0.60±0.85	0.054†	0.7
Sadness induction task	0.20±0.56	0.37±0.60	0.33±0.62	0.47±0.72	0.2‡	0.028
Neutral condition	0.20±0.51	0.50±0.60	0.55±0.62	0.63±0.71	–	–

Data are expressed as mean±SD change from time point 1 (baseline).
P values >0.1 were rounded to the tenth decimal place.
VAS indicates visual analog scale.
*P=0.008 after adjusting for the changes in anxiety and sadness VAS ratings.
†P=0.054 after adjusting for the changes in anger and sadness VAS ratings.
‡P=0.2 after adjusting for the changes in anger and anxiety VAS ratings.

change in heart rate across time for the anger, anxiety, or sadness induction tasks.

DISCUSSION

Using a randomized controlled design, our experimental study examined the acute effects of 3 core negative emotions, anger, anxiety, and sadness, on EC health. There were several important findings. First, after each of the negative emotion induction tasks, the greatest increase in the corresponding self-rated negative emotion was observed, indicating that the induction tasks had their intended effects on provoking the targeted negative emotion. These findings highlight the high fidelity of our negative mood induction tasks. Second, compared with the neutral condition, provoked anger led to an impairment in RHI score from 0 to 40 minutes post induction. The impairment in RHI score in the anger versus neutral condition was no longer present after the 40-minute postinduction assessment, indicating the acute effects of anger provocation on RHI score. Third, compared with the neutral condition, there were no statistically significant changes in RHI score with the anxiety and sadness conditions. Fourth, the results did not change after adjusting for noncondition VAS ratings, suggesting that the effects of anger versus neutral condition on RHI score were not influenced by nonspecific increases in anxiety and sadness. Finally, there were no changes in EMPs and EPCs due to any of the induction tasks.

Our study was not designed to examine how the acute transient effects of negative emotions on EC health relate to long-term cardiovascular risk. Although the effects of anger and anxiety on EDV were transient, the induction of these negative emotions was accomplished by having the participant think and speak about a recent incident that had provoked these emotions. It is possible that their effects on EC health occur routinely throughout the day or week, with potentially long-term consequences. Repeated episodes of a negative emotion may affect cardiovascular physiology over time, causing delayed recovery and eventually

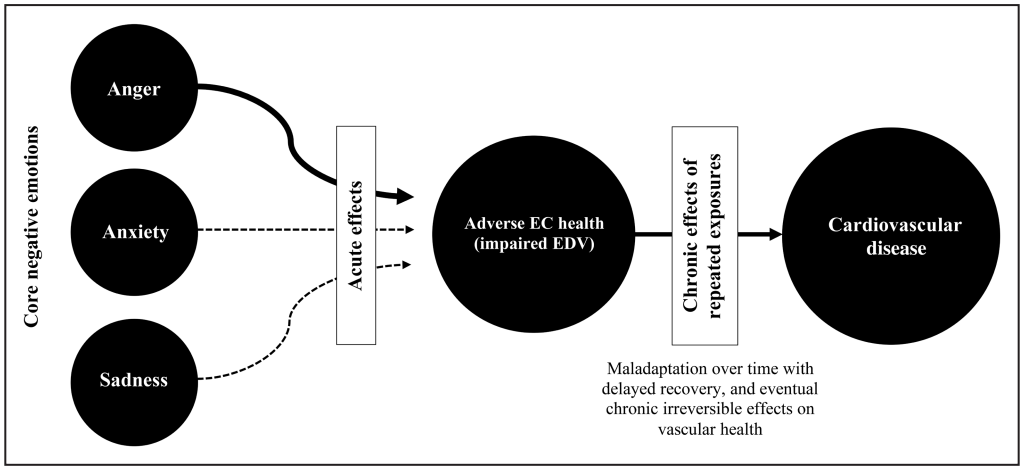

Figure 3. The effects of the experiencing negative emotions on endothelial cell health, and the potential pathway to cardiovascular disease onset.
The solid black line between anger and impaired EDV indicates an effect. The dotted lines between anxiety and impaired EDV, and sadness and impaired EDV indicate no evidence supporting this link in the current study. EC indicates endothelial cell; and EDV, endothelium-dependent vasodilation.

irreversible damage leading to increased CVD risk.[36–38] It is also possible that repeated episodes of a negative emotion contribute to long-term CVD risk, only in combination with other CVD risk or environmental factors.[39–41]

There have been few studies that have examined the acute effect of anger induction on EDV with each study demonstrating that anger provocation acutely impairs EDV.[19,35] However, these studies had small sample sizes (n=14–38) and did not randomize participants to a neutral condition. A neutral condition is essential because talking, which is part of the anger-recall task, can lead to physiological changes, and it is unclear from these prior studies whether it was anger provocation per se that led to an impairment in EDV. EDV has a diurnal variation with increases from the early morning to later in the day,[42,43] which is an important factor to control for in the analyses. Increases in EDV in the neutral condition, consistent with a diurnal variation, were observed in the current study. Finally, a neutral condition also controls for the effects of the repeated conduct of experimental procedures themselves on the measures of EC health.

In a recent, small randomized trial of 43 patients with recent ST-segment–elevation myocardial infarction and low trait anger control, anger management by cognitive behavioral therapy versus control condition led to a greater improvement in EDV at 3-month follow-up.[44] However, both groups had an improvement in trait anger control with no statistically significant difference between groups. Therefore, whether the improvement in EDV associated with cognitive behavioral therapy was due to an improvement in anger is unclear. There are scarce data on the effects of anxiety on EDV. In our study, anxiety induction had no statistically significant effects on EDV, compared with the neutral condition. In a recent randomized trial of 72 adults with moderate to high trait anxiety, a behavioral intervention, acceptance and commitment therapy versus control condition, led to a reduction in trait anxiety at 8-week follow-up yet had no effect on EDV.[45] These findings suggest that impaired EDV may not be a major mechanism underlying the link between anxiety and increased risk of CVD events. To our knowledge, no prior study has examined the effect of sadness on EDV. In our study, there was no strong indication that sadness induction impaired EDV. Future studies should test whether interventions that target trait versus state anger and anxiety improve EDV.

Studies of animal and human models of stress have demonstrated that acute or chronic stress is associated with adverse effects on EC health.[15–18,46] EDV is a common outcome measure in human studies that examined the effect of a mental stress task on EC health.[15–18] These studies did not examine other EC health measures including EMPs and EPCs. In our prior small, nonrandomized study, compared with baseline, the anger recall task not only impaired EDV (ie, RHI score) but also increased EMPs (ie, CD62E+ EMPs) and decreased EPCs (ie, CD34+/CD133+/KDR+ cells) among 30 apparently healthy individuals.[19] In contrast, in a separate experiment involving 6 age- and sex-matched controls, a neutral task did not adversely affect these EC health measures. In the current randomized controlled study, compared with the neutral condition, provoked anger, anxiety, and sadness did not acutely affect EMPs and EPCs, providing strong evidence that the acute experience of these negative emotions does not injure ECs or reduce EC reparative capacity. Our study was designed to examine 3 different primary outcomes measures of EC health, and the results indicate that EDV was the most important EC-specific pathway that was affected by provoked anger, and to a lesser extent, anxiety.

The underlying biological pathways by which anger impairs EDV are unknown. A common paradigm found in the literature is that the biological consequences of stress are primarily explained by activation of the autonomic nervous system. Indeed, sympathetic nervous system activation is associated with an impairment in EDV.[47,48] Other potential pathways include a dysregulated hypothalamic–pituitary–adrenal axis, impaired nitric oxide bioavailability, oxidative stress, increased arterial inflammation from enhanced bone marrow activity, and increased endothelin-1, which are each associated with psychological stress and contribute to endothelium-dependent vascular impairment.[15,49–53]

There were several potential limitations. The sample was young and apparently healthy without comorbidities. It is unclear whether the findings of our study are generalizable to older adults with comorbidities, who mostly likely would be taking medications. The study was intentionally designed to exclude individuals with prevalent CVD, CVD risk factors, and cardiovascular medications, which all reduce EDV and may have confounded the results because baseline EDV would already be impaired. Future studies should be designed to investigate whether similar effects are observed among older adults with comorbidities. The study did not include a within-subjects design where all participants undergo each of the negative emotion provocation tasks and the neutral condition in randomized order. Our decision not to use a within-subjects design was based on our experience in our prior unpublished studies in which we found considerable asymmetric order effects, particularly when participants were randomized to the anger recall task first. For the secondary hypotheses, we made the a priori assumption that the effect sizes for the anxiety and sadness conditions are comparable to the effect size for anger condition. It is possible that the study was not sufficiently powered to examine smaller effects of anxiety or sadness. We also

did not include the induction of positive emotions, joy or laughter, which can transiently improve EDV,[54–56] as we were interested in negative emotions shown to be associated with increased CVD events rather than factors that may be protective. In future studies, there may be merit to examining whether the adverse effects of negative emotion provocation on EDV can be blunted by positive emotions. During recovery, the participants sat quietly in a chair, which may not reflect the real-world conditions an individual may be in after experiencing a negative emotion. Our study was designed to standardize the recovery phase across all conditions and to isolate the effects of negative emotion induction task on EC health. This study did not examine other EC health measures including soluble adhesion molecules, and inflammatory cytokines. Finally, this study did not examine the chronic effects of exposure to repeated provoked negative emotion tasks nor did the study examine ecologic momentary feelings of anger, anxiety, and sadness outside the laboratory setting. It would be advantageous to test these effects on RHI score as well as EMPs and EPCs, which were not observed to be adversely affected with a single brief provocation of each negative emotion.

CONCLUSIONS

In conclusion, feelings of anger, anxiety, and sadness are common experiences. In the current study, the provocation of anger adversely affected EC health by impairing EDV, whereas the provocation of anxiety and sadness did not. An implication of these findings is that the contribution of a biological mechanism that increases CVD risk may differ across core negative emotions (Figure 3). Therefore, all negative emotions should not be grouped together as the same when looking through the lens of CVD pathophysiology. Investigation into the deeper mechanisms underlying the links between anger, anxiety, and endothelial dysfunction may help identify effective intervention targets for a large proportion of individuals at increased risk for CVD.

ARTICLE INFORMATION

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Disclosures

None.

Supplemental Material

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