FUNCTION™ american physiological society

FUNCTION, 2022, 3(4): zqac029

https://doi.org/10.1093/function/zqac029
Advance Access Publication Date: 26 May 2022
Original Research

RESEARCH ARTICLE

The Link Between Childhood Adversity and Cardiovascular Disease Risk: Role of Cerebral and Systemic Vasculature

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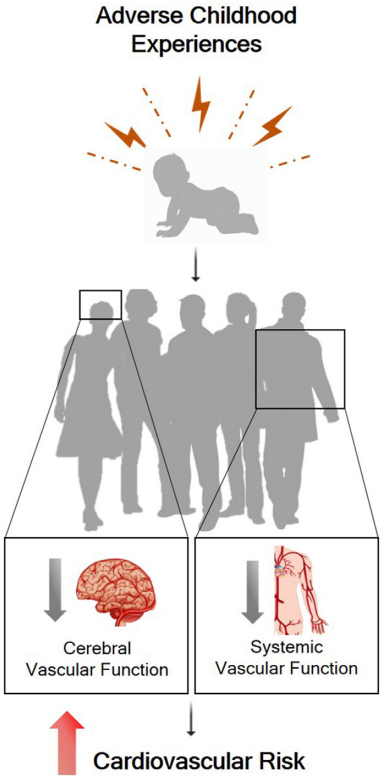
Abstract

Adverse childhood experiences (ACEs) are traumatic events during the first years of life that are associated with a higher risk of developing cardiovascular disease (CVD) during adulthood. The medial prefrontal cortex (mPFC) is a core region in the brain that modulates emotions and is directly involved in the cardiovascular response to stress by increasing vascular resistance. In the present study we examined the relationship between ACEs, mPFC and peripheral vascular function. Forty-five, adults (33±5 yrs.) participated in the present study to evaluate cerebral hemodynamics and peripheral vascular function. The impact of adverse experiences was evaluated through the ACE questionnaire. Among those that experienced ACEs (ACE group, n = 22), there was a significantly ($P < 0.001$) reduced activation of the mPFC as well as greater peripheral vascular resistance observed in the small ($P \leq 0.035$), conduit ($P \leq 0.042$) and large ($P \leq 0.001$) blood vessels, when compared to those that did not report ACEs (Control group, n = 23). In addition, relationships between the number of ACEs and mPFC activation ($r_s = -0.428$; $P = 0.003$) and peripheral vascular function ($r_s \leq -0.373$; $P \leq 0.009$) were observed. Findings from the present study support that adults who experienced ACEs exhibit a reduced activation of the mPFC along with systemic vascular dysfunction. In addition, individuals exposed to more childhood traumatic events exhibited a progressively greater inactivation of the mPFC and an increased peripheral vasoconstriction in a dose-dependent manner. These findings provide novel insights into the potential role that the brain and the peripheral vasculature may have in connecting adverse childhood events to the increased risk of CVD.

Submitted: 11 April 2022; **Revised:** 10 May 2022; **Accepted:** 13 May 2022



Key words: adverse childhood event; cerebral blood flow; vascular function; nitric oxide; cardiovacular

Introduction

Childhood adversity, including abuse, neglect, or household dysfunctions, are highly prevalent stressful experiences during the first years of life that have a crucial impact on the individual's psychological and physical health.[1,2] Compelling evidence supports that adverse childhood experiences (ACEs) increase the likelihood of developing cardiovascular diseases (CVD) later in life, including ischemic heart disease, hypertension, or atherosclerosis.[3–5] Vascular endothelial dysfunction, a well-established precursor of CVD,[6] is an important mediator in the ACE-induced increase in CVD risk.[7] Prevailing data in the literature supports that young adults exposed to ACE exhibit greater peripheral vascular resistance, augmented arterial stiffness, and reduced endothelial function,[8,9] all independent predictors of CVD events.

The brain is directly involved in controlling cardiovascular functions during stressful situations.[10] Specifically, the medial prefrontal cortex (mPFC), a core region of the brain that modulates emotions,[11] is directly engaged in the cardiovascular response to stress.[12] The mPFC closely interacts with subcortical regions, such as the amygdala,[13] activating the autonomous nervous system and promoting changes in cardiovascular reactivity that include exacerbated changes in blood pressure, heart rate, or peripheral vascular resistance.[10] Data supports that stress-mediated peripheral vasoconstriction is related to reduced activation of the mPFC[14] and associated hyperresponsiveness of the amygdala.[15] Importantly, this cascade of events is recognized in individuals with depression, anxiety, or post-traumatic stress disorders (PTSD).[16,17] However, to the best of our knowledge, no studies have examined these mechanisms as it relates to ACE-related stress.

Thus, the present study sought to test the hypothesis that adults who experienced ACEs had a reduced mPFC activation that was associated with a reduced peripheral vascular function when compared to adults that did not experience ACEs.

Material and Methods

Experimental Design

Volunteers presented to the Laboratory of Integrative Vascular and Exercise Physiology (LIVEP) on two separate occasions: a preliminary day and an experimental day. The preliminary day consisted of assessing body composition, blood pressure, medical history, and overall health status. For the experimental visit, participants reported to the LIVEP at 8:00 AM following an overnight fast and having abstained from tobacco, caffeine, and vigorous physical activity for 24 h and vitamin supplementation for 72 h. Cerebral hemodynamics and peripheral vascular function were evaluated.

Participants

A total of forty-five adult men and women, ages 18–41 yr were enrolled in the present study, following the principals of the Declaration of Helsinki and after approval by the Institutional Review Board at Augusta University. Participants were excluded if they (1) had a body mass index (BMI) greater than 40 kg/m^2 (Class III obesity), (2) were pregnant or postmenopausal women, (3) were diagnosed with any cardiovascular, pulmonary, renal, hepatic, cerebral, or metabolic disease, (3) were prescribed any vasoactive medications (i.e., nitrates, β-blockers, angiotensin-converting enzyme inhibitors, PDE-5 inhibitors, etc.), or (4) had

symptoms of uncontrolled hypertension. All participants were informed of the objectives, and possible risks of the investigation before written consent for participation was obtained.

Demographic Characteristics and Clinical Laboratory Values

Demographic characteristics were evaluated in all the participants during the preliminary visit. Volunteers completed a standard anthropometric assessment of height, weight, and calculated body mass index (BMI). Fasting concentrations of standard biochemical values for lipids (total cholesterol (TC), high-density lipoproteins (HDL), low-density lipoproteins (LDL), and triglycerides) and glucose levels were obtained using the Cholestech LDX analyzer (Alere, Providence, RI). Hemoglobin and hematocrit values were obtained using the HemoPoint H2 analyzer (Stanbio Laboratory, Boerne, TX). Concentrations of high-sensitivity C-reactive protein (hsCRP) were obtained from standard core laboratory techniques (Laboratory Corporation of America Holdings, Burlington, NC).

Adverse Childhood Experiences

ACEs information was collected in all the participants using the ACE questionnaire from the Behavioral Risk Factor Surveillance System adapted from the original CDC-Kaiser Study, specifically designed to gather information related to specific experiences during childhood. The ACE questionnaire contained detailed questions about childhood abuse, neglect, and household dysfunction divided into eleven items[18]. The ACE score (the number of categories of ACEs reported) was used to assess the cumulative effect of multiple ACEs, following the ACE scoring guidelines. Each exposure to a specific category of childhood adversity was coded as one ACE. Participants' ACE score composite indicated how many types of childhood adverse events they were exposed to.

Cerebral Hemodynamics

A Portalite near-infrared spectroscopy (NIRS) device (Artinis, Elst, The Netherlands) was used to non-invasively monitor cerebral hemodynamics in the mPFC. Changes in cerebral hemodynamics are strongly correlated, both spatially and temporally, with changes in neural activity.[19,20] NIRS was used as a continuous-wave tissue spectrometer that operates at wavelengths of 760 and 850 nm. The device is designed with three transmitters and one receiver that penetrate between 30 to 40 mm, to avoid signal interference from the scalp, skull, and cerebrospinal fluid. The Portalite device was secured to the subject's forehead and covered with a black cloth to avoid light interference while the participant laid supine in a low-light and temperature-controlled room. Resting assessments were recorded from each participant for 5 min to evaluate cerebral hemodynamics through a modification of the Lambert–Beer Law using individual age-related brain differential pathlength factor.[21] The changes in concentrations of oxyhemoglobin (O_2Hb) and deoxyhemoglobin (HHb), reflecting brain activity, were obtained. Total hemoglobin (tHb), representing the sum of O_2Hb and HHb, was evaluated as an indicator of cerebral blood volume (CBV) and therefore, cerebral hemodynamics.[20] Cerebral oxygen saturation (ScO_2) was calculated from the ratio of O_2Hb to tHb, considering the scattering coefficient of the brain. Estimated measurements of CBF were also computed using a derivation of the Fick equation.[22] Oxyhemoglobin concentration, blood concentration of hemoglobin (Hb), arterial saturation (SaO_2) measured non-invasively with a pulse oximeter, and a constant (k) related to the mass density of brain tissue were used as follows:

$$CBF = k \frac{\Delta [O_2Hb]}{Hb \int_0^{\Delta t} \Delta SaO_2 \ (t)\, dt}$$

Macrovascular Function

Brachial artery endothelial function was evaluated using the flow-mediated dilation (FMD) test. A detailed explanation of the technique has been described previously[23]. Briefly, using a 12-MHz linear transducer, simultaneous B-mode and blood velocity profiles of the brachial artery were evaluated through ultrasound imaging (Logiq 7, G.E. Medical Systems, Milwaukee, WI). After an initial baseline, a forearm occlusion cuff (D.E. Hokanson, Bellevue, WA) placed immediately distal to the medial epicondyle, was rapidly inflated to 250 mmHg for five minutes (E-20 rapid cuff inflator, D.E. Hokanson,) to induce arterial occlusion. Then, the pressure of the cuff was release inducing reactive hyperemia of the brachial artery[23]. R-wave gaiting (Accusync 72, Accusync Medical Research Corporation, Milford, CN) was used to capture end-diastolic arterial diameters and used for automated offline analysis of brachial artery vasodilation (Medical Imaging Applications, Coralville, IA). Peak diameter was determined by the highest five second average following cuff release. FMD is expressed as the % increase in peak diameter from baseline diameter. Cumulative shear rate (area under the curve, AUC) was determined every 4 s for the first 20 s, and every 5 s thereafter for the remainder 2-min data collection period using the trapezoidal rule. FMD was normalized by shear rate and presented as FMD/shear, as previously was reported.[23]

Microvascular Function

Peripheral vascular function in the cutaneous microvasculature was evaluated using laser Doppler imaging (MoorVMS-LDF, Moor Instruments, DE, USA) of the forearm. Imaging was combined with three reactivity tests to assess microvascular function: (1) local thermal hyperemia (LTH) to determine microvascular maximal dilation primarily mediated by nitric oxide and endothelial-derived hyperpolarization factors (EDHF), (2) post-occlusive reactive hyperemia (PORH) to evaluate microvascular shear-stress response primarily mediated by sensory nerves and EDHF, and (3) iontophoresis of acetylcholine (Ach) to assess microvascular endothelial-dependent vasodilation through nitric oxide, EDHF, and prostaglandins. A detailed explanation of each test has been described elsewhere.[24]

For this study, baseline (BL) flux was determined prior to every test by calculating a 30 s average. Brownian movement of macromolecules in the cutaneous interstitial space was calculated when a forearm cuff was inflated, reported as biological zero (B_0), and subtracted from both baseline and peak responses. Cutaneous blood flow was evaluated as red blood cell flux (RBF) in perfusion units (PU) and as cutaneous vascular conductance (CVC) when mean arterial blood pressure was considered (PU/mmHg). Results are presented as (1) maximal hyperemic response (Peak), (2) area under the curve (Area), and (3) time-to-peak (TTP), which represents the time from the start of the stimuli to the maximal hyperemic response. Skin resistance (SR) was also evaluated for every individual using Ohm's law.

Arterial Stiffness

Peripheral vascular function in the central arteries was determined non-invasively through applanation tonometry using the SphygmoCor Xcel System (AtCor Medical, Sydney, Australia). Augmentation index (AIx) was determined in duplicate and normalized to a heart rate of 75 beats per minute (AIx75).[25] Carotid–femoral pulse wave velocity (cfPWV), another index of arterial stiffness, was determined in duplicate by sequentially recording electrocardiographic-gated carotid and femoral artery waveforms using applanation tonometry with the SphygmoCor Xcel system and controlled by blood pressure.[26]

Statistical Analysis

The data were analyzed using SPSS version 27 (SPSS Inc., Chicago, IL) and expressed as mean ± standard error of mean (SEM), unless otherwise noted. An initial power calculation was performed based on the anticipated effect size estimated for the primary outcomes. The initial proposed sample size yielded a power > 0.89 in the primary outcome for the present study (macrovascular function). A power analysis and sample size calculation were performed before initiating the study, considering that under most circumstances, an $\alpha = 0.05$ and a statistical power ≥ 0.85 is well accepted.

For all statistical analyses, significance was set at $P<0.05$. Based on the ACE score obtained in the questionnaire and for analysis purposes, subjects were divided into two groups: those who reported an ACE score equal or greater to 1 (ACE group) and those who reported an ACE score of 0 (Control group), following a similar stratification than for other traumatic-related conditions such as PTSD. The Shapiro-Wilk test was used to analyze the normality of the measurement distribution. When normality was met, independent group t-tests were performed to identify group differences between participants from the ACE and the Control group. If normality was not met, Mann-Whitney U tests were completed. Results are illustrated with box-and-whisker plots with minimum and maximum values. To compare the effects of ACE on cerebral hemodynamics and vascular function, a one-way ANOVA was completed with post hoc comparison using the Tukey's Honest Significant Difference test and group based on ACE scores (0, 1–4, 5–7, and > 8). Relationships between ACEs, cerebral hemodynamics, and peripheral vascular function assessments were evaluated using Pearson's correlation coefficients (r) or Spearman's correlation (r_s) when the assumption of normality was not met. Effect size calculations using Cohen's d were reported for primary outcomes to represent small (Cohen's $d = 0.2$), medium (Cohen's $d = 0.5$), and large (Cohen's $d = 0.8$) effect sizes[27,28].

Results

Participant Characteristics and Clinical Laboratory Values

Demographic characteristics and laboratory values for participants from the ACE and the Control groups are presented in Table 1. As expected, the only significant ($P < 0.001$) difference among both groups was the number of ACEs reported. No differences in subject demographics and body composition were observed between participants from both groups. Similarly, no differences were identified in the clinical laboratory values between both groups.

Table 1. Participant Characteristics and Clinical Laboratory Values

Variable	ACE	Control	P value
N	22	23	–
ACE score	4 ± 2	0 ± 0	**<0.001**
Sex (M/F)	14/8	10/13	0.113
Race (C/AA)	9/13	11/12	0.291
Age (yrs)	34 ± 3	31 ± 5	0.135
Height (cm)	174 ± 9	164 ± 8	0.194
Weight (kg)	88 ± 17	79 ± 20	0.134
BMI (kg/m^2)	29.5 ± 3.7	26.9 ± 4.5	0.186
Waist/hip ratio	0.9 ± 0.1	0.9 ± 0.1	0.321
Heart rate (bpm)	63 ± 17	59 ± 11	0.299
SBP (mmHg)	121 ± 9	112 ± 8	0.110
DBP (mmHg)	75 ± 6	72 ± 6	0.148
MAP (mmHg)	90 ± 6	85 ± 7	0.101
TC (mg/dL)	175 ± 23	162 ± 28	0.201
HDL (mg/dL)	51 ± 12	56 ± 18	0.201
LDL (mg/dL)	95 ± 29	84 ± 28	0.183
TRIG (mg/dL)	101 ± 29	82 ± 32	0.106
TC/HDL ratio	3.5 ± 0.9	3.0 ± 1.1	0.109
GLU (mg/dL)	90 ± 8	87 ± 8	0.106
HbA_{1C} (%)	5.5 ± 0.3	5.4 ± 0.4	0.103
Hb (g/dL)	14.7 ± 1.6	14.1 ± 1.4	0.112
Hct (%)	44 ± 4	42 ± 4	0.791
hs-CRP (mg/L)	2.3 ± 1.5	1.7 ± 1.5	0.190

Values are mean ± standard deviation (S.D.). Boldfaced value indicates statistical significance. ACE: Adverse childhood event; M: male; F: female; C: Caucasian; AA.: African American; BMI: body mass index; SBP: systolic blood pressure; DBP: diastolic blood pressure; MAP: mean arterial blood pressure; TC: total cholesterol; HDL: high density lipoproteins; LDL: low density lipoproteins; TRIG: triglycerides; GLU: glucose; HbA1c: hemoglobin A1c; Hb: hemoglobin; Hct: hematocrit; hs-CRP: high sensitivity C-reactive protein.

Table 2. Cerebral Hemodynamics

Variable	ACE	Control	P value
ScO_2 (%)	65 ± 1	64 ± 1	0.420
O_2Hb (μM)	34 ± 2	45 ± 2	**<0.001**
HHb (μM)	22 ± 2	28 ± 2	**0.013**
tHb (μM)	56 ± 3	71 ± 3	**<0.001**

Values are mean ± standard error of mean (SEM). Boldfaced value indicates statistical significance. ACE: Adverse childhood event; ScO_2: cerebral oxygen saturation; O_2Hb: oxyhemoglobin; HHb: deoxyhemoglobin; tHb: total hemoglobin.

Cerebral Hemodynamics

Values related to cerebral hemodynamics in both the ACE and the Control groups are presented in Table 2. Overall, both groups showed similar ($P = 0.420$) cerebral oxygen saturation, as expected from individuals that are considered apparently healthy. Nonetheless, participants in the ACE group exhibited a significantly lower oxyhemoglobin ($P < 0.001$; Cohen's $d = 1.27$) and CBV ($P < 0.001$; Cohen's $d = 0.99$, Figure 1A) when compared to the Control group. Similarly, the balance between O_2 supply and O_2 demand in the cerebral tissue was also significantly lower in the ACE group when compared to the control (ACE: 12±1 μM *vs*. Control: 17±2 μM, $P = 0.033$; Cohen's $d = 0.69$). Cerebral blood flow was also significantly ($P < 0.001$; Cohen's $d = 1.76$) reduced in the ACE group than in the Control group (36±2 ml/100g/min *vs*. 51±2 ml/100g/min, respectively).

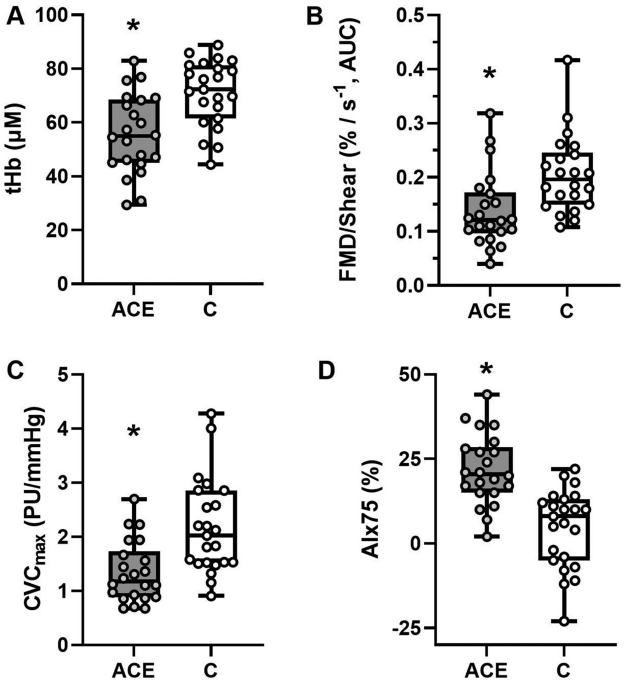


Figure 1. Individual data illustrated as Box-and-Whisker plots with minimum and maximum values for (A) cerebral hemodynamics (total hemoglobin, tHb), (B) macrovascular function (flow mediated dilation normalized for shear rate, FMD/shear), (C) microvascular function (maximal cutaneous vascular conductance, CVC_{max}), and (D) arterial stiffness (augmentation index normalized for a heart rate of 75 bpm, AIx75). Group differences were determined by independent group *t* tests or Mann-Whitney U-tests and denoted by $*$ when $P < 0.05$ vs. Control group. μM: micromolar; AUC: Area under the curve; PU: perfusion units.

Macrovascular Function

Table 3 presents vascular function in the brachial artery in participants from the ACE and the Control groups. Baseline diameter was similar ($P = 0.101$) between groups. The FMD response was significantly ($P = 0.042$; Cohen's $d = 0.70$) lower in the ACE group compared to the Control group. No differences ($P = 0.625$) in shear rate were identified between both groups; howbeit, FMD normalized for shear rate was significantly ($P = 0.030$; Cohen's $d = 0.96$, Figure 1B) lower in the participants from the ACE group compared to the Control. No differences ($P = 0.354$) were identified between both groups in the time-to-peak vasodilation.

Microvascular Function

Data for participants in the ACE and Control groups for red blood flux (RBF) and cutaneous vascular conductance (CVC) is presented in Table 4. Baseline flux and conductance were similar ($P \geq 0.340$) between groups for all the reactivity tests completed. Brownian movement of macromolecules in the cutaneous interstitial space was also similar between participants from both groups (B_0, ACE: 1.4 ± 0.1 PU *vs.* Control: 1.8 ± 0.1 PU, $P = 0.121$). No differences ($P = 0.274$) in skin resistance were identified between groups (ACE: 176 419 ± 13 094 Ω vs. Control: 159 181 ± 9063 Ω).

For local thermal hyperemia, the overall microvascular response was lower in the ACE group compared to the Control group. Specifically, during the initial peak in response to the thermal provocation, the ACE group exhibited a significantly lower response than the Control group (RBF, ACE: 95±7 PU *vs.* Control: 126±12 PU, $P = 0.056$; CVC, ACE: 1.1 ± 0.1 PU/mmHg *vs.* Control 1.5 ± 0.2 PU/mmHg, $P = 0.026$). Similarly, maximal dilation achieved during the plateau phase was significantly ($P \leq 0.020$) lower in the ACE group than in the Control for both RBF ($P = 0.020$; Cohen's $d = 0.91$) and CVC ($P < 0.001$; Cohen's $d = 1.46$, Figure 1C).

For the post-occlusive reactive hyperemia test, both RBF and CVC baseline values were similar between groups ($P \geq 0.491$). During this reactivity test, the ACE group showed a lower RBF hyperemic response (compared to the Control group ($P = 0.096$), findings that persisted when controlling for changes in blood pressure ($P = 0.005$; Cohen's $d = 0.90$).

During iontophoresis with Ach, both groups showed similar baseline response ($P \geq 0.340$) and a progressive increase in perfusion in response to the delivery of Ach. However, the ACE group showed a significantly reduced maximum response compared to the Control group, both for RBF ($P = 0.035$; Cohen's $d = 0.80$) and CVC ($P = 0.002$; Cohen's $d = 1.30$).

Arterial Stiffness

Parameters of arterial stiffness in both the ACE and Control groups are presented in Table 5. Significant differences ($P \leq 0.001$, Cohen's $d = 1.51$) were identified between both groups in the arterial tonometry assessments for both AIx and adjusted AIx75 (Figure 1D). Similarly, significant ($P < 0.001$, Cohen's $d = 1.40$) differences were observed in cfPWV assessment between the ACE and control group.

Relationships Between ACEs, Cerebral Hemodynamics, and Peripheral Vascular Function

Associations between ACE scores and markers of cerebral and peripheral vascular function are illustrated in Figure 2. Significant differences between groups were identified in cerebral hemodynamics (tHb, $F_{(3,44)} = 4.59$, $P = 0.007$; Figure 2A), with

Table 3. Macrovascular Function

Variable	ACE	Control	*P* value
Baseline Diameter (mm)	3.37 ± 0.2	3.33 ± 0.2	0.101
Peak Diameter (mm)	3.86 ± 0.2	3.73 ± 0.2	0.379
FMD (%)	6.3 ± 0.7	8.6 ± 0.8	**0.042**
Shear (s^{-1}, AUC)	44,606 ± 5261	41,517 ± 3632	0.625
FMD (%)/Shear (s^{-1}, AUC)	0.139 ± 0.02	0.206 ± 0.02	**0.030**
Time to Peak (s)	45 ± 3	51 ± 5	0.354

Values are mean ± standard error of men (SEM). Boldfaced value indicates statistical significance. ACE: Adverse childhood event; FMD: flow-mediated dilation; AUC: area under the curve.

Table 4. Microvascular function

Variable	ACE	Control	P value
Local Thermal Hyperemia			
$Baseline_{LTH}$ (PU)	9 ± 1	9 ± 1	0.923
$Peak_{LTH}$ (PU)	125 ± 12	174 ± 16	**0.020**
$Area_{LTH}$ (PU s^{-1})	151 980 ± 16 706	211 190 ± 20 042	**0.031**
$Baseline_{LTH}$ (PU/mm Hg)	0.10 ± 0.01	0.10 ± 0.02	0.370
$Peak_{LTH}$ (PU/mm Hg)	1.34 ± 0.13	2.17 ± 0.18	**<0.001**
TTP_{LTH} (s)	1056 ± 55	1128 ± 19	0.192
Post-Occlusive Reactive Hyperemia			
$Baseline_{PORH}$ (PU)	8 ± 1	8 ± 1	0.939
$Peak_{PORH}$ (PU)	39 ± 5	50 ± 4	0.096
$Area_{PORH}$ (PU s^{-1})	3048 ± 324	4067 ± 310	**0.029**
$Baseline_{PORH}$ (PU/mm Hg)	0.09 ± 0.01	0.10 ± 0.01	0.491
$Peak_{PORH}$ (PU/mm Hg)	0.43 ± 0.05	0.64 ± 0.05	**0.005**
TTP_{PORH} (s)	35 ± 4	33 ± 4	0.715
Iontophoresis with Acetylcholine			
$Baseline_{ACH}$ (PU)	8 ± 1	7 ± 2	0.340
$Peak_{ACH}$ (PU)	98 ± 8	128 ± 11	**0.035**
$Area_{ACH}$ (PU s^{-1})	40 998 ± 4484	52 673 ± 4577	**0.047**
$Baseline_{ACH}$ (PU/mm Hg)	0.09 ± 0.01	0.08 ± 0.03	0.728
$Peak_{ACH}$ (PU/mm Hg)	1.11 ± 0.12	1.64 ± 0.50	**0.002**
$TTP_{ACH,}$ (s)	18 ± 3	18 ± 2	0.933

Values are mean ± standard error of men (SEM). Boldfaced value indicates statistical significance. ACE: Adverse childhood event; LTH: Local thermal Hyperemia; PORH: Post occlusive reactive hyperemia; Ach: Acetylcholine; TTP: Time to peak; PU: perfusion units.

Table 5. Arterial Stiffness

Variable	ACE	Control	P value
AIx (%)	26 ± 2	13 ± 2	**<0.001**
AIx75 (%)	22 ± 2	5 ± 3	**<0.001**
cfPWV (m/s)	6.5 ± 0.3	5.5 ± 0.1	**<0.001**

Values are mean ± standard error of men (SEM). Boldfaced value indicates statistical significance. ACE: Adverse childhood event; AIx: Augmentation Index; AIx75: Augmentation index normalized for a heart rate of 75 beats per minute; cfPWV: carotid–femoral pulse wave velocity.

negative associations identified between ACE scores and markers of cerebral hemodynamics including tHb ($r_s = -0.428$; $P = 0.003$, Figure 2A), O_2Hb ($r_s = -0.442$; $P = 0.002$) and CBF ($r_s = -0.522$; $P < 0.001$). Similarly, macrovascular function was significantly difference among ACE groups (FMD: $F_{(3,43)} = 2.29$, $P = 0.009$; FMD/Shear: $F_{(3,43)} = 2.32$, $P = 0.009$) and ACE scores were negatively associated with FMD ($r_s = -0.335$; $P = 0.019$) and FMD/Shear ($r_s = -0.423$; $P = 0.003$, Figure 2B). Likewise, significant differences in microvascular function were also identified among ACE groups ($F_{(3,44)} = 3.22$, $P = 0.032$, Figure 2C) and ACE scores negatively associated with maximal dilation (CVC_{max}: $r_s = -0.373$; $P = 0.009$, Figure 2C). Significant differences were also identified among ACE groups in arterial stiffness (AIx75: $F_{(3,44)} = 6.59$, $P < 0.001$, Figure 2D; PWV: $F_{(3,44)} = 13.53$, $P < 0.001$, Figure 2F) and ACE scores were negatively associated with AIx ($r_s = 0.518$; $P < 0.001$), AIx75 ($r_s = 0.545$; $P < 0.001$, Figure 2D), and PWV ($r_s = 0.525$; $P = 0.001$, Figure 2F).

Relationships between cerebral hemodynamics and markers of peripheral vascular function were also evaluated and are illustrated in Figure 3. Specifically, cerebral hemodynamics were significantly associated with markers of peripheral vascular function including FMD ($r = 0.335$; $P = 0.026$) and FMD/Shear ($r = 0.413$; $P = 0.005$, Figure 3A), microvascular function through maximal dilation ($r = 0.381$; $P = 0.027$, Figure 3B) and arterial stiffness ($r = -0.477$; $P = 0.012$, Figure 3C).

Discussion

The experience of traumatic events during the first years of life has tremendous implications for the individual's psychological and physical health. Cumulative evidence supports a link between childhood adversity and the early development of multiple conditions later in life, such as ischemic heart disease, hypertension, or atherosclerosis. The present investigation has expanded prevailing information and has examined whether adversity early in life is associated with dysfunctions on the mPFC region, an area of the brain centrally involved in emotional regulation, as well as reduced peripheral vascular function. Findings of the present investigation also identified that different physiological mechanisms that are involved in central, conduit, and microvascular responses are diminished in individuals exposed to ACEs supporting the notion of multisystemic vascular dysfunction. In addition, present results support that individuals exposed to larger number of childhood traumatic events exhibited a progressively greater inactivation of the mPFC and increased peripheral vasoconstriction in a dose-dependent manner. Thus, the current study provides new and important insights into the link between ACE, mPFC, and peripheral vascular function.

Adversity occurring during the first years of life triggers behavioral and emotional stress that mediate physiological changes and precipitate an adaptive response to environmental stressors.[1,3] Acute and intense mental stress leads to structural changes in the mPFC, a critical region of the brain that regulates emotions.[29] Changes are detected by the amygdala, which responds to threats and activates physiological mechanisms related to stress.[30] Compelling data supports that in conditions such as anxiety or PTSD, there is decreased activity of the mPFC[29,31,32] and an increase in amygdala function.[33,34] Prevailing data in the literature have also reported that children affected by low socioeconomic status, a known ACE, have a reduced mPFC activity[35] and associated greater amygdala volume.[36] Findings from the present study are in good agreement

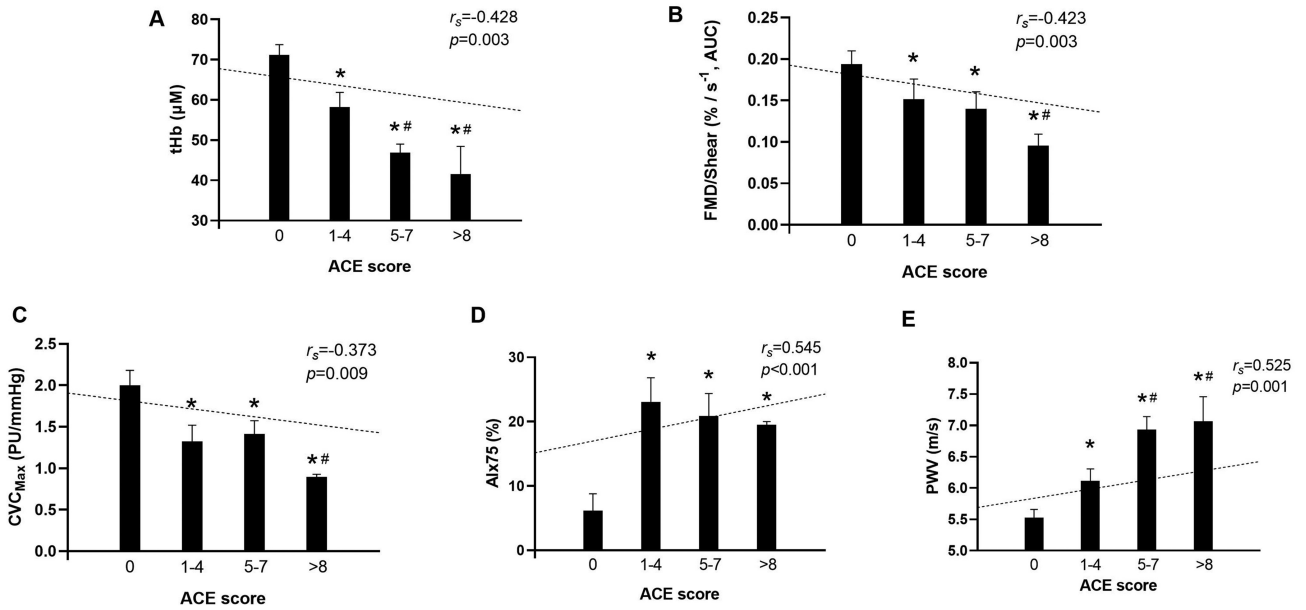


Figure 2. Relationships between ACE scores and (A) cerebral hemodynamics (tHb); (B) macrovascular function (FMD/Shear); (C) microvascular function (CVC_{max}); (D) arterial stiffness (AIx75); (E) pulse-wave velocity (PWV). Differences between groups determined by a one-way ANOVA based on ACE score (0, 1 to 4, 5, to 7 and a score greater than 8) and denoted by $* P < 0.05$ *vs.* Control group, $\# P < 0.05$ *vs.* 1–4 group. Relationships between individual ACE scores and each measurement were evaluated using Spearman's correlation coefficients and denoted by a dotted line.

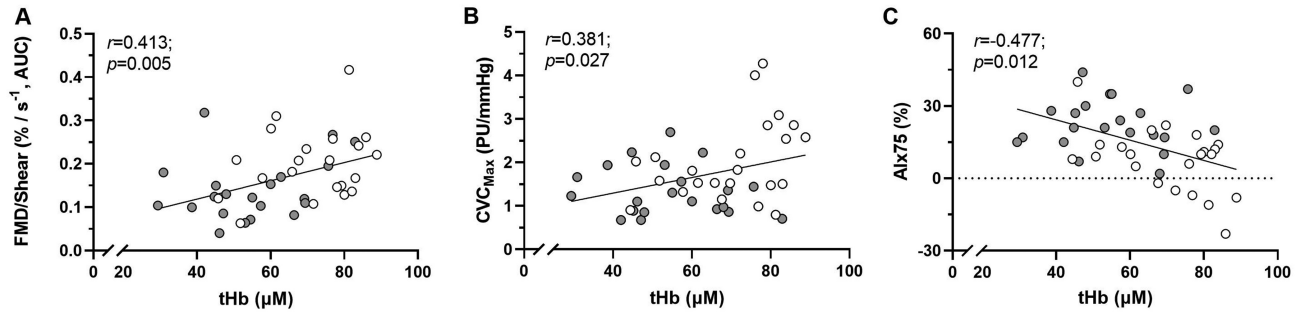


Figure 3. Relationships between mPFC hemodynamics and markers of peripheral vascular including (A) macrovascular function (FMD/Shear), (B) microvascular function (CVC_{max}) and (C) arterial stiffness (AIx75). Relationships were identified using Pearson's correlation coefficients. ACE group, grey circles. Control group, white circles.

with prior data since adults that were exposed to adversity early in life presented with reduced activation of the mPFC. Interestingly, the present results also revealed an inverse relationship between mPFC activity and the number of ACEs suggesting that those individuals who were exposed to a greater number of stressors during childhood exhibited lower activation of the mPFC during adulthood. Considering that both animal and human data support that early exposure to chronic stress produces lasting neurobiological changes in the immature brain[37] and the mPFC matures primarily during adolescence,[38] it is not surprising that repeated childhood stressors can progressively impact the plasticity and functionality of the mPFC.[37]

Apart from emotional recognition and processing, the mPFC plays a central role in modulating the cardiovascular response to stress and fear.[39] Emotional stress triggers the mPFC-amygdala circuit that prompts a hyperactivation of the sympathetic nervous system.[40,41] In response, the body secretes hormones to cope with stress through rapid changes in blood pressure, heart rate, disruption of normal endothelial function, abnormal vasoconstriction, and peripheral vascular resistance.[42,43] Inactivation of the mPFC has also been related to reductions in vagal activity and, therefore, exacerbate the cardiovascular response.[44] It is important to note that endothelial dysfunction is considered one of the earliest precursors in the development and progression of nearly all types of CVD.[45] In this line, data from animal models using maternal separation as an early life stressor, support the presence of vascular endothelial dysfunction in adult mice.[46] Similar outcomes have been recently confirmed in young adults exposed to ACEs with comparable associations identified between macrovascular function and the reported number of ACEs.[9] Current results extend previous findings and identify that individuals who experienced adversity during childhood also present with a significantly diminished vascular endothelial function both in the macro- and the microvasculature. Endothelial cells, and therefore endothelial dysfunction, differ in structure and phenotype, depending on the vessel type, and macro and microvessels respond differently to insults.[47] Particularly, endothelial dysfunction in the macrovasculature is frequently associated with a reduced nitric oxide bioavailability,[48] while prostaglandins and EDHF play an important role in the microcirculation.[49] In the present study, parallel dysfunctions in both the micro and macrovascular response were observed in the ACE cohort, supporting that different physiological mechanisms that govern vascular health are impacted

in these individuals. It is also worth noting that 1% reduction in vasodilation during the FMD test has been associated with 8% greater risk of future cardiovascular events.[50] Accordingly, based on the FMD data, the ACE group in the present study may present with a ≈ 18% greater risk to develop future cardiovascular outcomes compared to the control group. These results are especially noteworthy since the cohort investigated is still young and all classic clinical cardiovascular markers (i.e., blood pressure, lipid profile, CRP, diabetes) evaluated demonstrate that the participants from both groups are relatively healthy.[51]

If the trauma is severe and/or repeated, the vasodilatory response of the endothelium is progressively reduced over time, leading to vasoconstriction, increasing blood flow resistance, and stiffening large arteries. Thus, it is not surprising that individuals in the ACE cohort also presented with significantly stiffer arteries compared with those from the control group. Similar results have been obtained from large epidemiological studies supporting that children who face adversity are more likely to exhibit higher blood pressure and augmented arterial stiffness[8] during early adulthood,[52] thus contributing to a greater risk to develop ischemic heart disease, hypertension or atherosclerosis.[3–5] Deficiencies in different mechanisms have been associated with greater aortic stiffness including reduced NO, increased endothelin-1 (ET-1) and/or altered glycosaminoglycans, among others. Prevailing data have already identified elevated concentrations of circulating ET-1 in adolescents exposed to ACEs, along with enhanced arterial stiffness.[8] Bearing in mind the role that NO plays in the functionality of blood vessel function, it is possible that a reduced systemic NO bioavailability may contribute, at least in part, to the observed systemic vascular dysfunction identified in the present study. However, further investigations of the role of NO in ACE-mediated vascular dysfunction are needed to confirm that hypothesis. Nonetheless, findings from the present study expand current knowledge, and demonstrate that different physiological mechanisms of endothelial dysfunction, one of the earliest markers of CVD,[45] are associated with childhood adversity. These results also emphasize the importance of investigating earlier markers of CVD risk in order to identify premature vascular dysfunction in otherwise healthy individuals.

To our knowledge, few studies have explored the association between stress, brain, and cardiovascular health. Data from a large cohort reported that low socioeconomic status was associated with higher resting amygdala activity and a greater risk of developing cardiovascular events.[53,54] Similarly, a relationship between stress-mediated peripheral vasoconstriction and reduced activation of the mPFC was found in patients with coronary artery disease.[14] Results from the present study expand previous observations and identify that individuals exposed to childhood adversity exhibited a reduced activation of the mPFC and a progressively greater vascular dysfunction of the small, conduit, and large vessels. Bearing in mind that 23% of children in the U.S. have experienced two or more ACEs,[55] these results reinforce the notion that early traumatic experiences represent a threat to cardiovascular health and should be considered an emerging public health concern. Certainly, these findings gain more significance considering that physical and, especially, emotionally stressful events tend to be underrecognized in routine clinical care. Conditions such as stress cardiomyopathy exemplified the limited attention paid to emotional stressors into one's health until severe symptomatology appears. Present findings also highlight that other cardiovascular conditions, where endothelial dysfunction is a primary mechanism,[45] may also be impacted by ACEs. These cardiovascular events carry a high risk of a fatal outcome, so increasing clinical and public health awareness of the risks related to repetitive childhood adversity is critical and deserves further attention.

The high prevalence of ACEs and their association with a wide variety of negative outcomes support the need to prioritize ACEs recognition and prevention as early as possible. Primary prevention of childhood adversity through detailed screening and assessment, as well as early identification and intervention, are crucial in order to mitigate the ACE-related negative health consequences.[56] The present results also open the door to potential novel interventions targeting psychological wellbeing that could exert benefits on the neuro- and cardio- vascular systems. Interventions aiming at reducing stress and the associated neurobiological changes could also be effective therapies to mitigate childhood stress-related cardiovascular morbidity and mortality prior to the development of severe outcomes.[57]

Conclusion

In conclusion, for the first time, results from the present study demonstrate that adults who experienced adversity during their childhood exhibit a reduced activation of the mPFC and a concomitant systemic vascular dysfunction that impacts small, conduit, and large vessels. Findings also identified that individuals that were exposed to a greater number of ACEs exhibit a progressively greater inactivation of the mPFC and an associated decreased peripheral vasodilation in a dose-dependent manner. Results from the present study emphasize the importance of early life adversities as a modifiable risk factor for the development of premature CVD. Additionally, these findings identify the brain, and especially the mPFC region, as a potential therapeutic target that can mitigate the CVD risk associated with ACEs. Future studies should expand these results in larger cohorts and evaluate interventions that target stress-induced changes in the brain as well as systemic vascular dysfunction, to further reduce the burden of cardiovascular disease.

Acknowledgments

The authors want to thank all the volunteers that participated in this study, including those that were part of the Augusta Heart Study, for their commitment to this research investigation.

Funding

This work was supported in part by the National Institute of Health NIH/NHLBI P01HL06999 (JSP, RAH) and the American Heart Association 16POST31080031 (PRM).

Conflict of Interest Statement

None declared

Data Availability

The data underlying this article will be shared on reasonable request to the corresponding author.

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