1 2	Associations between non-invasive upper- and lower-limb vascular function assessments: Extending the evidence to
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Peripheral Vascular Function in Young Women

# 41 ABSTRACT

42 Brachial artery (BA) flow-mediated dilation (FMD) is a well-established measure of peripheral vascular function prognostic of future cardiovascular events. The vasodilatory response to FMD 43 (FMD%) reflects upper-limb conduit artery function, while reactive hyperemia (RH) following 44 45 cuff-occlusion release reflects upper-limb resistance artery function. Comparatively, passive leg movement (PLM) is a newer, increasingly utilized assessment of lower-limb resistance artery 46 function. To increase its clinical utility, PLM-induced leg blood flow (LBF) responses have been 47 compared to hemodynamic responses to FMD, but only in men. Therefore, the purpose of this 48 study was to retrospectively compare LBF responses to FMD% and RH responses in women. 49 We hypothesized that LBF responses would be positively associated with both FMD% and RH, 50 but to a greater extent with RH. FMD and PLM were performed on 72 women (23±4 years). 51 52 Arterial diameter and blood velocity were assessed using Doppler ultrasound. Pearson 53 correlation coefficients were used to evaluate associations. Measures of resistance artery function were weakly positively associated: change in BA blood flow  $\triangle BABF$  and  $\triangle LBF$  (r=0.33, 54 p < 0.01), BABF area under the curve (BABF AUC) and LBF AUC (r = 0.33, p < 0.01), and BABF<sub>neak</sub> 55 and LBF<sub>peak</sub> (r=0.37, p<0.01). However, FMD% was not associated with any index of PLM (all 56 57 p>0.30). In women, indices of resistance artery function in the upper- and lower-limbs were positively associated. However, contrary to the previous work in men, upper-limb conduit artery 58 59 function was not associated with lower-limb resistance artery function suggesting these assessments capture different aspects of vascular function and should not be used 60 interchangeably in women. 61

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Key words: flow-mediated dilation, passive leg movement, vascular function, conduit artery
 function, resistance artery function

Peripheral Vascular Function in Young Women

# 65 New and Noteworthy

Upper- and lower-limb indices of resistance artery function are positively associated in young women when assessed by reactive hyperemia following brachial artery flow-mediated dilation (FMD) cuff-occlusion release and leg blood flow responses to passive leg movement (PLM), respectively. However, despite previous data demonstrating a positive association between upper-limb conduit artery function assessed by FMD and lower-limb resistance artery function assessed by PLM in young men, these measures do not appear to be related in young women.

Peripheral Vascular Function in Young Women

# 72 INTRODUCTION

The vascular system plays a critical role in the delivery and control of blood flow via 73 alterations in vascular tone. More specifically, the vascular endothelium is a single cell layer that 74 lines the entire circulatory system and is responsible for the vasomotor properties of the 75 76 vasculature via the controlled release of vasodilator and vasoconstrictor substances (1). Wellestablished, non-invasive laboratory methods are used to indirectly assess endothelial-mediated 77 peripheral vascular function in both large conduit arteries and smaller resistance arteries. 78 Importantly, these laboratory methods assessing vascular endothelial function can provide 79 valuable insight into cardiovascular disease (CVD) risk and progression, even in young, 80 otherwise healthy individuals. 81

Brachial artery flow-mediated dilation (FMD) is a non-invasive assessment of upper-limb 82 83 conduit artery function. The vasodilatory response to FMD, expressed as the percent change in 84 brachial artery diameter following cuff occlusion-release (FMD%), is traditionally used to evaluate peripheral macrovascular function and has been shown to predict future cardiovascular 85 events in a variety of populations (2-5). Recently, FMD% performed following current guidelines 86 87 (2, 6) was reported to be strongly positively associated with the more invasive assessment of coronary artery endothelial function with an r value of 0.77, highlighting the clinical relevance of 88 FMD% (7). Although the FMD test is commonly utilized to assess macrovascular function, 89 90 reactive hyperemia (RH) following FMD cuff-occlusion release can be quantified via Doppler ultrasound and be used as a proxy to assess downstream resistance artery, or microvascular, 91 function (8, 9). Like FMD%, the RH response following FMD cuff-occlusion release has also 92 been shown to predict future cardiovascular events in both apparently healthy and diseased 93 populations (10, 11). Of note, disturbances in microvascular function generally precede that of 94 95 the macrovasculature, therefore, microvascular assessments may be particularly useful in providing insight into CVD disease risk in younger populations free of CVD (11-13). 96

## Version of record at: https://doi.org/10.1152/japplphysiol.00177.2022

#### Peripheral Vascular Function in Young Women

In comparison to FMD, passive leg movement (PLM) is a newer, methodologically 97 98 simpler, non-invasive assessment indicative of lower-limb microvascular function measured at the femoral artery (14). PLM is similar to RH given that the leg blood flow (LBF) responses and 99 100 brachial artery blood flow (BABF) responses respectively, are largely dependent on downstream 101 resistance artery function (15). PLM has also been shown to be reliable and reproducible in both men and women (14, 16); however, the clinical utility of PLM in predicting future CVD risk 102 103 remains to be determined. Therefore, in an effort to increase its clinical relevance, PLM has been previously compared to FMD in a population of men (17). Those findings suggest that 104 PLM may provide a similar prognostic index of cardiovascular health as a strong positive 105 106 correlation was found between upright seated PLM and brachial artery FMD (17), however, it is unknown if these findings are generalizable to women. Thus, elucidating the relations between 107 108 FMD%, RH, and PLM in young women may expand the efficacy of PLM and allow for a better 109 interpretation of each vascular measure in terms of whole-body vascular health.

Therefore, the purpose of this retrospective study was to compare LBF responses to PLM to both brachial artery FMD% and brachial artery RH in healthy young women. We hypothesized that LBF responses to PLM would be moderately positively associated with FMD% (r = 0.40 - 0.69) and strongly positively associated with RH responses (r > 0.70) in this population of healthy young women.

#### Peripheral Vascular Function in Young Women

# 115 METHODS

## 116 Study Participants and Protocol

The data presented in this study are from a retrospective analysis of vascular function 117 data collected between 2016 and 2021 within the same vascular research laboratory at the 118 119 University of Delaware. Protocols were approved by the Institutional Review Board at the University of Delaware (IRB study #'s 941369 and 1288814) and were conducted in accordance 120 121 with the Declaration of Helsinki. Written informed consent was obtained from all participants prior to participation. This study included apparently healthy premenopausal women between 122 the ages of 18 and 35, recruited from the University of Delaware and the surrounding Newark, 123 DE region. Participants were non-hypertensive (blood pressure <140/90 mmHg), non-obese 124 (body mass index <30kg/m<sup>2</sup>), non-tobacco users (<1 cigarette in the past month), free of any 125 126 chronic diseases, and did not take any medications or supplements that might interfere with 127 vascular function. Following consenting procedures, participants self-identified their race/ethnicity, completed a review of medical history, body mass index was calculated, and 128 body fat percentage was assessed using bioelectrical impedance (Tanita TBF-300A, Arlington 129 Heights, IL). 130

131 All participants completed the vascular assessments during the same visit and, when appropriate, during the early follicular phase of their menstrual cycle to control for the potential 132 influence of sex hormones on our findings (18). Specifically, the vascular visit was scheduled 133 134 within 7 days of the onset of menstruation for naturally cycling participants, during the placebo/no pill phase for participants taking oral contraceptives, and for participants using other 135 136 forms of hormonal contraception, this visit was scheduled during menstruation, or at the participant's convenience if they did not menstruate as a result of their contraception (n=2). All 137 assessments were performed during the morning hours (i.e., between 7-11 AM) in a 138 139 temperature-controlled laboratory (~23°C). Participants were instructed to report to the laboratory fasted for  $\geq 6$  hours, without caffeine, alcohol, and exercise  $\geq 24$  hours, and without 140

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Peripheral Vascular Function in Young Women

over-the-counter medications or supplements for ≥24 hours prior to the visit. Upon arrival to the laboratory, resting blood pressure was assessed (Omron 5 Series, BP7200) and participants underwent intravenous blood sampling for the clinical analysis of fasting blood glucose and a lipid panel (Quest Diagnostics, Inc., Philadelphia, PA). Participants rested quietly for a minimum of 20 minutes prior to FMD and a minimum of 10 minutes between the transition from supine FMD to upright-seated PLM.

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#### 148 Flow-Mediated Dilation Protocol

FMD was performed in accordance with current guidelines (6). In the supine position 149 with the right arm fully extended, a narrow rapidly inflating cuff (E20 Rapid Cuff Inflation System, 150 Hokanson, WA) was placed immediately proximal to the elbow and distal to the imaging site. 151 152 Brachial artery diameter and blood velocity were recorded proximal to the cuff and distal to the 153 shoulder joint via duplex ultrasound imaging (Logig e, General Electric Medical Systems, Milwaukee, WI) using a linear array ultrasound probe (12 Hz) and a transducer with a Doppler 154 frequency of 5 MHz, with the probe appropriately positioned to maintain an insonation angle of 155 156 60° or less. A 3-lead electrocardiogram (ECG100C, BIOPAC Systems Inc., CA) was used to 157 record end diastolic R-wave gated images throughout baseline and post-cuff occlusion. Following 1-minute of baseline measures, the cuff was rapidly inflated to 250 mmHg for 5 158 159 minutes. Brachial artery diameter and blood velocity were measured continuously throughout baseline and 2 minutes immediately following cuff deflation. Images were collected from the 160 video output of the Logiq e for offline analysis of brachial artery diameter conducted using an 161 automated edge-detection software (Brachial Analyzer for Research, Medical Imaging 162 Applications, Coralville, IA). 163

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#### 165 Passive Leg Movement Protocol

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#### Peripheral Vascular Function in Young Women

PLM was performed as previously described (15, 19-21) and in accordance with current 166 167 guidelines (14). Briefly, in the upright seated position, the protocol consisted of 1 minute of baseline measurements immediately followed by a 1-minute bout of passive leg flexion and 168 extension at the knee joint. Participants were instructed to remain passive and relaxed 169 170 throughout the duration of the protocol. Femoral artery diameter and blood velocity were recorded in the right common femoral artery (CFA) distal to the inguinal crease and proximal to 171 the femoral artery bifurcation via duplex ultrasound imaging (as described above). Passive 172 movement was achieved by a member of the research team moving the participant's lower leg 173 at the knee joint through a 90° to 180° range of motion at a frequency of 1 Hz, while movement 174 cadence was maintained by a metronome. Femoral artery diameter was measured during 175 baseline, while blood velocity was measured throughout the protocol. Throughout the duration 176 177 of the protocol, the unaffected leg remained extended and fully supported.

178

# 179 Blood Flow Calculations and Analyses

Arterial blood velocity for both tests was evaluated using the Logig e ultrasound system 180 (as described above). The sample volume was maximized according to vessel size and 181 182 centered within the vessel based on real-time ultrasound visualization. Arterial diameter was determined at a perpendicular angle along the central axis of the scanned area and mean 183 velocity (V<sub>mean</sub>) values [angle-corrected and intensity-weighted area under the curve (AUC)] 184 were automatically calculated by the Doppler ultrasound system. Using arterial diameter 185 arterial blood flow was mathematically calculated 186 and  $V_{\text{mean}}$ , as follows: blood flow =  $V_{\text{mean}} \pi$  (vessel diameter/2)<sup>2</sup> × 60, where blood flow is in milliliters per minute, in custom 187 Excel spreadsheets designed for FMD or PLM analyses. 188

For FMD, end-diastolic ECG R-wave gated images were collected from the video output of the Logiq e for offline analysis of brachial artery diameter using automated edge-detection software (Brachial Analyzer for Research, Medical Imaging Applications, Coralville, IA). FMD%

Peripheral Vascular Function in Young Women

192 was calculated as the maximal percent change in brachial artery diameter from baseline to post 193 occlusion-cuff release. Indices of RH included ΔBABF, BABF AUC, and BABF<sub>peak</sub>. ΔBABF was 194 calculated as peak blood flow - baseline blood flow. BABF AUC was calculated as the sum of brachial artery blood flow above baseline for 2-minutes following cuff release, according to the 195 196 trapezoidal rule and using the equation as follows:  $\Sigma(y_i(x_{(i+1)} - x_i))$ +  $(1/2)(y_{(i+1)} - y_i)(x_{(i+1)} - x_i))$ . BABF<sub>peak</sub> was identified as the highest blood flow achieved during 197 the 2 minutes following occlusion-cuff release. 198

For PLM, V mean was calculated as anterograde – retrograde blood velocities using 199 continuous ultrasound Doppler imaging of the CFA. LBF was then calculated in ml/min as V mean 200  $\pi$  (vessel diameter/2)<sup>2</sup>) × 60. Baseline LBF was calculated as a 60-second average of V mean. 201 while second-by-second analysis of anterograde and retrograde blood velocities were used to 202 determine V<sub>mean</sub> and LBF flow during the movement phase of PLM, using the blood flow 203 204 equation previously described. Indices of PLM included the overall change in LBF from baseline to peak (ALBF), LBF AUC, and LBF<sub>peak</sub>. ALBF was calculated as LBF<sub>peak</sub> – baseline LBF. LBF 205 AUC was calculated as the sum of femoral artery blood flow above baseline for each second 206 during the 60-second movement phase of PLM, according to the trapezoidal rule and using the 207 208 equation as follows:  $\Sigma(y_i(x_{(i+1)} - x_i) + (1/2)(y_{(i+1)} - y_i)(x_{(i+1)} - x_i))$ . LBF<sub>peak</sub> was calculated as 209 the maximal blood flow achieved during the first 30-second movement phase of PLM.

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# 211 Statistical Analyses

Pearson correlation coefficients (Pearson's *r*) were used to evaluate associations between LBF responses to PLM and FMD% and RH variables and 95% confidence intervals were calculated. The strength of the associations were interpreted based on the following criteria: weak (r = 0.10 - 0.39), moderate (r = 0.40 - 0.69), or strong (r = 0.70 - 0.89) (22).  $\Delta$ LBF was compared with  $\Delta$ BABF, LBF AUC was compared with BABF AUC, LBF<sub>peak</sub> was compared with BABF<sub>peak</sub>, and all LBF variables were compared with FMD%. Analyses were

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Peripheral Vascular Function in Young Women

- 218 performed using GraphPad Software (Version 9.3, San Diego, CA). An a priori power analysis
- was conducted using the software package G\*Power 3.1.9.4 (23) for sample size estimation. To
- detect a medium effect with significance set at  $\alpha \le 0.05$  and power = 0.80, the minimum sample
- size needed is 69, thus our obtained sample size of 72 is adequate to test the study hypothesis.
- Statistical significance was accepted at  $p \le 0.05$  and participant characteristics and continuous
- 223 data are presented as mean ± standard deviation (SD).

Peripheral Vascular Function in Young Women

# 224 **RESULTS**

225 Participant characteristics are presented in **Table 1**. A total of 72 premenopausal women (41 White women, 15 Black women, 8 Hispanic women, and 8 Asian women; 23±4 years) 226 completed both upper- and lower-limb vascular assessments. Fifty-eight (81%) women in the 227 228 present study were not using any form of hormonal birth control, 8 were taking oral contraceptive pills, and 6 were using other forms of hormonal contraception including progestin 229 230 intrauterine device (n=3), Nexplanon (n=2), and Depo Provera (n=1). All anthropometric variables, resting blood pressure, and basic clinical blood values fell within normal expected 231 ranges for healthy young women. Group averages for FMD and PLM variables are displayed in 232 Table 2. 233

234

# 235 Associations Between FMD and PLM

236 There were significant positive associations between indices of lower-limb resistance artery function assessed by LBF responses to PLM and indices of upper-limb resistance artery 237 function assessed by RH following FMD cuff-occlusion release (**Figure 1**). Specifically,  $\Delta LBF$ 238 239 was positively associated with ΔBABF (Figure 1A), LBF AUC was positively associated with 240 BABF AUC (Figure 1B), and BABF<sub>peak</sub> was positively associated with LBF<sub>peak</sub> (Figure 1C). However, none of the indices of lower-limb resistance artery function as assessed by LBF 241 242 responses to PLM were associated with upper-limb conduit artery function assessed by FMD% (Table 3). 243

Peripheral Vascular Function in Young Women

# 244 DISCUSSION

245 This study sought to compare the hyperemic responses to PLM to the hemodynamic responses to the FMD assessment in healthy young women. We found that upper-limb conduit 246 artery (macrovascular) function assessed by FMD% was not associated with any measure of 247 248 lower-limb resistance artery function as assessed by LBF responses to PLM. However, upperand lower-limb resistance artery (microvascular) function assessed by brachial artery RH and 249 250 PLM respectively, were weakly positively associated, suggesting some overlap between microvascular physiological responses in the upper and lower limbs in young women. 251 Importantly, brachial artery RH following FMD cuff-occlusion release has been shown to be a 252 strong predictor of future cardiovascular events in both healthy and diseased populations; 253 therefore, findings from the present study help to advance the clinical utility of PLM, particularly 254 255 in young women who represent a generally understudied population in vascular research.

#### 256 FMD% is not associated with LBF responses to PLM

Studies comparing macrovascular and microvascular function in non-diseased 257 populations are limited, despite the knowledge that impairments in microvascular function may 258 be more readily detected than impairments in macrovascular function in young, otherwise 259 260 healthy populations (24-26). Associations between FMD% and LBF responses to PLM have 261 been previously studied in healthy young and older men and demonstrated a strong positive association between FMD% and LBF responses to PLM (17). For instance, FMD% was found to 262 263 be significantly correlated with both  $\Delta$ LBF and LBF AUC, with r values for young men reaching 0.73 and 0.55, respectively (17). Alternatively, the present study does not indicate any 264 association between FMD% and LBF responses to PLM in young women and this discrepancy 265 in findings may be explained by sex differences in macrovascular and microvascular function 266 267 that have been identified across the lifespan.

### Version of record at: https://doi.org/10.1152/japplphysiol.00177.2022

## Peripheral Vascular Function in Young Women

268 Deteriorations in macrovascular function that occur with aging have been shown to be significantly delayed in women compared to men (27). As such, investigating microvascular 269 function in young women may be more sensitive and insightful regarding CVD risk, especially 270 given that macrovascular function seems to be preserved in this population. For example, in 271 272 response to prolonged sitting, a condition known to impair vascular function (28), young men experienced reductions in both lower-limb macrovascular and microvascular function as 273 274 assessed by popliteal artery FMD and hyperemic blood flow AUC following popliteal artery FMD, respectively (29). In contrast, young women only experienced reductions in microvascular 275 function, while macrovascular function remained unchanged in response to prolonged sitting 276 277 (29). There are also data demonstrating that young adult women have similar vascular function compared to young adult men in the upper extremities as measured by brachial artery FMD% 278 279 normalized for shear rate AUC, but appear to have reduced vascular function normalized for 280 shear rate AUC in the lower extremities measured at the popliteal artery (30). Therefore, the absence of an association between our measures of microvascular and macrovascular function 281 among young women in the present study may be driven by preserved macrovascular function, 282 283 particularly in the upper limbs, and an earlier decline in microvascular function in women as compared to age-matched men (31). Indeed, the presence of the female sex hormone estrogen 284 and its cardioprotective effects in young premenopausal women (32) likely help to explain, at 285 least in part, the discrepancy between our findings and the prior FMD% and PLM comparison in 286 287 young men (17).

Similar to findings from the present study, several previous studies have also reported no association between FMD% to other non-invasive assessments of vascular function. FMD% was not found to be associated with superficial femoral artery FMD nor popliteal artery FMD performed in the lower-limbs of young healthy adults (33), nor upper-limb microvascular function assessed by cutaneous reactive hyperemia following upper arm ischemia in adults aged 19 to 68 (34). It is important to consider that brachial artery FMD is performed in the upper-limb

# Version of record at: https://doi.org/10.1152/japplphysiol.00177.2022

Peripheral Vascular Function in Young Women

294 macrovasculature while PLM is performed in the lower-limb microvasculature, which may, in 295 part, be responsible for the null findings in the present study limbs (30, 35, 36). Moreover, FMD% and PLM are governed by potentially different mechanisms; PLM is mostly NO-mediated 296 (15) while FMD% is only partly mediated by shear stress-induced NO release when performed 297 298 following current guidelines (2) and other contributing vasodilators have been identified including prostaglandins (PG) and endothelium-derived hyperpolarizing factor (EDHF) (37, 38). 299 300 Therefore, findings from the present study are in agreement with previous work demonstrating that, despite its clinical importance and predictive abilities of overall cardiac disease incidence 301 (4), FMD% may not reflect other measures of peripheral vascular function (e.g., it does not 302 appear to be related to lower limb microvascular function in young women). 303

## 304 RH following cuff occlusion is positively associated with LBF responses to PLM

305 Both brachial artery RH and the LBF responses to PLM assess microvascular function, 306 as the hyperemic responses to both tests are largely dependent on down-stream resistance artery vasodilation (14). In the present study, LBF responses to PLM were positively associated 307 with RH in young women such that ΔLBF was positively associated with ΔBABF, LBF AUC was 308 309 positively associated with BABF AUC, and LBF<sub>peak</sub> was positively associated with BABF<sub>peak</sub> 310 suggesting overlap in microvascular function in the upper and lower limbs. However, the 311 associations between these parameters were relatively weak, which may be partly explained by the relative contributions of various vasodilators i.e., NO, PG, and EDHF, to each test. LBF 312 responses to PLM are ~80% NO mediated (15), and it has been recently determined that PG 313 314 and EDHF do not play a role in the hyperemic response to PLM in healthy young men (39). In contrast, the contribution of NO to BABF AUC is only ~30% (40) whereas the contribution of PG 315 is far greater, especially in controlling the BABF<sub>peak</sub> response (41). Moreover, RH following FMD 316 cuff release is also known to be mediated by combined activation of inwardly rectifying 317 318 potassium channels (KIR) and Na(+)/K(+)-ATPase which function to hyperpolarize

#### Version of record at: https://doi.org/10.1152/japplphysiol.00177.2022

#### Peripheral Vascular Function in Young Women

319 microvascular smooth muscle cells and induce resistance artery vasodilation (37, 42, 43). There 320 are also clear limb-specific vasomotor responses to shear and vasoactive substances as 321 demonstrated by distinct blood flow and vasodilatory responses to similar amounts of exerciseinduced shear between the brachial artery, the common femoral artery, and the deep femoral 322 323 artery (36). Therefore, although both assessments commonly evaluate resistance artery function, the heterogeneity of the vascular bed in which each test is performed (upper versus 324 lower limb), the relative contribution of vasodilators, and the differing mechanisms of each test 325 may be limiting the strength of this relation identified in these young women. 326

#### 327 Experimental Considerations

328 Brachial artery FMD% and RH have both been shown to be predictive of CVD risk, which justifies their widespread use as well as the clinical utility of these vascular assessments. 329 330 In comparison, PLM is a newer non-invasive vascular function assessment and may present as a methodologically simpler test than the traditionally performed brachial artery FMD. However, 331 the predictive ability of PLM in terms of CVD risk still remains largely unknown. Comparing PLM 332 to FMD may provide insight into the clinical utility of PLM, however it is still important to consider 333 various aspects of these tests that make them distinct from each other i.e., performed in an 334 335 upper extremity versus a lower extremity (36), macrovascular (conduit artery function) 336 assessment versus microvascular (resistance artery function) assessment (24, 34), the distinct mechanisms of each test, the observed sex differences in the vascular responses to each test 337 338 (29, 30), as well as certain disease states that more strongly influence the outcomes of one test over another (44). In order to establish PLM as a clinically relevant assessment of vascular 339 health, PLM should continue to be compared with other assessments of vascular function in a 340 341 variety of populations (45). In addition, future research may consider quantifying the contribution of NO and/or other vasodilators to each test when comparing vascular 342 343 assessments, as well as include both men and women across the lifespan.

Peripheral Vascular Function in Young Women

# 344 Conclusions

345 Our main findings suggest that RH following FMD cuff-occlusion release and LBF responses to PLM reflect some of the same microvascular physiological responses in the upper 346 347 and lower limbs in healthy premenopausal women. However, upper-limb conduit artery function and lower-limb resistance artery function, assessed by FMD% and PLM respectively, likely 348 capture different aspects of vascular function and should not be used interchangeably. FMD% is 349 350 a measure of upper-limb macrovascular function that is highly predictive of future cardiovascular 351 events, while in contrast, PLM is a measure of lower-limb microvascular function that is methodologically quicker and easier to perform and is predominantly NO mediated. Therefore, 352 performing both FMD% and PLM in combination may give the greatest insight into whole-body 353 354 vascular health, as clearly demonstrated by the unique benefits of each test.

Peripheral Vascular Function in Young Women

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362

# 363 DISCLOSURE STATEMENT

364 No conflicts of interest, financial or otherwise, are reported by the authors.

Peripheral Vascular Function in Young Women

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Peripheral Vascular Function in Young Women

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Peripheral Vascular Function in Young Women

496 Figure 1. Associations between indices of upper and lower limb resistance artery function as assessed by RH following FMD occlusion-cuff release and leg blood flow responses to PLM, 497 respectively (n=72). The association between the change in brachial artery blood flow (ΔBABF) 498 499 and the change in leg blood flow from baseline to peak ( $\Delta LBF$ ) (A). The association between brachial artery blood flow area under the curve (BABF AUC) and leg blood flow area under the 500 curve (LBF AUC) (B). The association between peak brachial artery blood flow (BABF<sub>peak</sub>) and 501 peak leg blood flow (LBF<sub>peak</sub>) (C). BABF, brachial artery blood flow; LBF, leg blood flow; AUC, 502 area under the curve; r, Pearson correlation coefficient; CI, confidence interval. P-values in **bold** 503 504 font indicate significance.

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# Table 1. Participant Characteristics

<b>N</b> (W/B/H/A)	<b>72</b> (41/15/8/8)
Age (years)	23 ± 4
Body Mass Index (kg/m²)	23.6 ± 2.4
Body Fat (%)	28.1 ± 5.4
Systolic Blood Pressure (mmHg)	111 ± 7
Diastolic Blood Pressure (mmHg)	69 ± 6
<sup>†</sup> Total Cholesterol (mg/dL)	157 ± 26
<sup>†</sup> HDL Cholesterol (mg/dL)	62 ± 14
<sup>†</sup> LDL Cholesterol (mg/dL)	81 ± 20
<sup>†</sup> Triglycerides (mg/dL)	67 ± 29
<sup>†</sup> Fasting Glucose (mg/dL)	85 ± 6

<sup>†</sup>Clinical blood values obtained in a subset of participants (n=67). Data displayed as mean ± standard deviation. BMI, body mass index; BP, blood pressure; HDL, high density lipoprotein; LDL, low density lipoprotein; W, white women; B, black women; H, Hispanic women; A, Asian women.

# Table 2. FMD and PLM variables

Baseline BA diameter (mm)	3.35 ± 0.43
Baseline BABF (ml/min)	43 ± 19
ΔBABF (ml/min)	461 ± 135
BABF AUC (ml)	347 ± 125
BABF <sub>peak</sub> (ml/min)	504 ± 146
FMD (%)	7.3 ± 3.2
Baseline CFA diameter (cm)	0.78 ± 0.09
Baseline LBF (ml/min)	224 ± 70
ΔLBF (ml/min)	397 ± 257
LBF AUC (ml)	129 ± 111
LBF <sub>peak</sub> (ml/min)	620 ± 283

Data displayed as mean  $\pm$  standard deviation (n = 72). FMD, flow-mediated dilation; PLM, passive leg movement; BA, brachial artery; BABF, brachial artery blood flow; AUC, area under the curve; CFA, common femoral artery; LBF, leg blood flow.

	FMD%		
	r	95% CI	p
∆LBF (ml/min)	0.05	(-0.17, 0.29)	0.70
LBF AUC (ml)	0.11	(-0.11, 0.34)	0.36
LBF <sub>Peak</sub> (ml/min)	0.05	(-0.16, 0.30)	0.65

**Table 3.** Associations between upper limb conduit artery function and lower limb resistance artery function as assessed by FMD and PLM, respectively.

Pearson correlation coefficients were used to evaluate relations between FMD and PLM variables (n=72). FMD, flow-mediated dilation; PLM, passive leg movement; LBF, leg blood flow; AUC, area under the curve; *r*, Pearson's correlation coefficient; CI, confidence interval.

# Associations between non-invasive upper- and lower-limb vascular function assessments: Extending the evidence to young women METHODS OUTCOMES



**CONCLUSIONS** Indices of resistance artery function in the upper- and lower-limbs were positively associated. However, upper-limb conduit artery function was not associated with lower-limb resistance artery function, suggesting these assessments capture different aspects of vascular function and should not be used interchangeably in women.