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Comparisons of Noninvasive Methods Used to Assess Exercise Stroke Volume in HFpEF

Erik H. Van Iterson¹, Thomas P. Olson¹, Barry A. Borlaug¹, Bruce D. Johnson¹, and Eric M. Snyder²

¹Department of Cardiovascular Medicine, Mayo Clinic, Rochester, MN

²Department of Kinesiology, University of Minnesota, Minneapolis, MN

Abstract

Introduction—Cardiopulmonary exercise testing (CPET) plays an important role in properly phenotyping signs and symptoms of heart failure with preserved ejection fraction (HFpEF). The prognostic value of CPET is strengthened when accompanied by cardiac hemodynamic measurements. Although recognized as the ‘Gold’ standard, cardiac catheterization is impractical for routine CPET. Thus, advancing the scientific/methodologic understanding of noninvasive techniques for exercise cardiac hemodynamic assessment is clinically impactful in HFpEF. This study tested the concurrent validity of noninvasive acetylene gas (C_2H_2) uptake, echocardiography (ECHO), and oxygen pulse (O_2 pulse) for measuring/predicting exercise stroke volume (SV) in HFpEF.

Methods—18 Caucasian HFpEF and 18 age/sex matched healthy controls participated in upright CPET (ages, 69 ± 9 vs. 63 ± 9 years). At rest, 20-W, and peak exercise, SV was measured at steady-state via C_2H_2 rebreath (SV_{ACET}) and ECHO (SV_{ECHO}), whereas O_2 pulse was derived ($=V O_2/HR$).

Results—Resting relationships between SV_{ACET} and SV_{ECHO} , SV_{ECHO} and O_2 pulse, or SV_{ACET} and O_2 pulse were significant in HFpEF ($R^2=0.30, 0.36, 0.67$), but not controls ($R^2=0.07, 0.01, 0.09$), respectively. Resting relationships persisted to 20-W in HFpEF ($R^2=0.70, 0.53, 0.70$) and controls ($R^2=0.05, 0.07, 0.21$), respectively. Peak exercise relationships were significant in HFpEF ($R^2=0.62, 0.24, 0.64$), but only for SV_{ACET} vs. O_2 pulse in controls ($R^2=0.07, 0.04, 0.33$), respectively. Standardized standard error of estimate (Std.SEE) between techniques was strongest in HFpEF at 20-W: SV_{ACET} vs. $SV_{ECHO}=0.57 \pm 0.22$; SV_{ECHO} vs. O_2 pulse= 0.71 ± 0.28 ; SV_{ACET} vs. O_2 pulse= 0.56 ± 0.22 .

Conclusion—Constituting a clinically impactful step towards construct validation testing, these data suggest SV_{ACET} , SV_{ECHO} , and O_2 pulse demonstrate moderate-to-strong concurrent validity for measuring/predicting exercise SV in HFpEF.

Correspondence to: Erik H. Van Iterson, PhD, MS, MA, Department of Cardiovascular Medicine, Mayo Clinic, 200 First St SW, Rochester, MN 55905, vaniterson.erik@mayo.edu, Phone: 773-433-0787, Fax: 507-255-4861.

Conflict of interest

The authors declare no conflicts of interest. The results of the present study do not constitute endorsement by ACSM. The results of the study are presented clearly, honestly, and without fabrication, falsification, or inappropriate data manipulation.

Keywords

cardiac output; echocardiography; CPET; inert gas breathing; heart failure with preserved ejection fraction

INTRODUCTION

Cardiopulmonary exercise testing (CPET) plays an important role in properly phenotyping signs and symptoms of patients with heart failure with preserved ejection fraction (HFpEF) (6, 17, 27, 29, 34). Whether exercise intolerance linked to HFpEF manifests directly from pathophysiology of central (e.g., impaired inotropy and lusitropy (1, 5, 6, 24, 29)) and/or peripheral (e.g., skeletal muscle (21)) factors, there are immediately recognizable clinical implications associated with a reduced cardiac hemodynamic response during CPET in HFpEF.

The direct Fick approach requiring steady-state measures of pulmonary oxygen uptake ($\dot{V}O_2$) and invasive arterial and mixed venous O_2 samples for quantifying arteriovenous oxygen content difference (AVO_2) is the 'Gold' standard for measuring cardiac output (Q) and stroke volume (SV) (20). However, clinically impactful factors including increased patient risk (3) and required resources (e.g., specialists and costs) make implementing the direct Fick method during routine CPET impractical. Therefore, because of the prognostic power associated with evaluating exercise cardiac hemodynamics in HFpEF (1, 5, 6, 17, 24, 29), there is a high-priority need to advance the understanding of unique scientific and methodologic factors influential to assessing noninvasive exercise cardiac hemodynamics in generalizable laboratory settings in HFpEF.

Noninvasive imaging methods for assessing cardiac hemodynamics involving echocardiography (ECHO) have been validated in controlled environments (19, 32). However, primary limitations of ECHO, which are pronounced during exercise, are that image acquisition and post-processing are highly sensitive to patient factors (e.g., obesity, body position, etc.) and subjective interpretations of personnel (23, 25). By contrast, perfusion sensitive inert gas pulmonary uptake techniques involving for example, acetylene (C_2H_2) (Bunsen solubility coefficient in blood is 0.740, ~two times that of carbon dioxide [CO_2 , 0.470] or nitrous oxide [N_2O , 0.405] (8, 15, 16, 36)) have demonstrated utility for objectively measuring Q and SV at rest and/or exercise in adults across the health spectrum (2, 13, 16, 33). Nevertheless, while observations from validation studies support the usefulness of inert gas uptake methods for measuring Q and SV in patients demonstrating overt cardiac and/or pulmonary limitations (2, 13, 16, 33), we acknowledge that marked perturbations in specific factors (e.g., low lung diffusing capacity (17, 30)) affecting ventilation-perfusion ($\dot{V}_A/Q$) matching could render results from these approaches open to question. Likewise, any factor disturbing $\dot{V}_A/Q$ matching influential to inert gas uptake methods would affect interpretability of oxygen pulse (O_2 pulse), which has been validated for predicting exercise SV given invariability in AVO_2 and heart rate (HR) (37, 40).

While separate lines of study have tested the utility of C_2H_2 uptake, ECHO, and O_2 pulse for measuring or predicting rest and/or exercise SV in a spectrum of adults (2, 16, 19, 20, 33,

37), those data cannot be immediately extrapolated to the unique population of HFpEF. There are immediate clinical implications linked to exercise cardiac hemodynamic testing in HFpEF (1, 5, 6, 17, 24, 29). Therefore, this study aimed to test concurrent validity across C_2H_2 uptake, ECHO, and O_2 pulse for noninvasively measuring or predicting exercise SV in patients with HFpEF.

METHODS

Participants—Eighteen Caucasian HFpEF patients and 18 Caucasian healthy control participants matched for age and sex participated in this study (demographics, Table 1). Inclusion criteria for HFpEF at initial study screening were (27–29, 34): left ventricular ejection fraction (LVEF) $\geq 50\%$ (within the previous 6 months), stable pharmacologic therapy ≥ 1 month, evidence of elevated left heart filling pressures (i.e., mitral E/e' >15 , or mitral E/e' >8 plus either enlarged left atrial volume ($>34\text{ mL/m}^2$), chronic loop diuretic use for control of symptoms, or elevated natriuretic peptides [BNP or NT-proBNP levels between 35–250 pg/mL or 125–300 pg/mL, respectively]), and absence of diagnosed lung disease (e.g., COPD, PH, etc.) secondary to HFpEF. Exclusion criteria for HFpEF were (within the previous 6 months): evidence of significant coronary artery disease (requiring for example, an angioplasty, coronary artery bypass grafting, etc.), valvular heart disease (any stenosis, $>$ mild regurgitation), hypertrophic or infiltrative cardiomyopathy, constrictive pericarditis, exercise-induced PH due to vascular disease (mean exercise pulmonary artery pressure $>30\text{ mm Hg}$ with pulmonary capillary wedge pressure $<15\text{ mm Hg}$), or radiographic pulmonary congestion were excluded. The Mayo Clinic Heart Failure Service and the Cardiovascular Health Clinic were used to recruit HFpEF. Controls were recruited through advertisement in the surrounding community. Controls had normal cardiac function without evidence of exercise-induced ischemia and were without history of hypertension, lung disease, or coronary artery disease. The present analysis represents a sub-study aim as well as subset of data from original work (7). However, the parts of these data that have been published previously do not relate to the primary aims of this manuscript. All aspects of this study were reviewed and approved by the Mayo Clinic Institutional Review Board, and all participants provided written informed consent prior to study participation.

Protocol—Upon arrival to the environmentally controlled physiology laboratory and after assessing body anthropometrics (e.g., height, weight, etc.), participants performed resting flow-volume loop spirometry (CPFS system spirometer, Medical Graphics, St. Paul, MN) while sitting upright according to ATS criteria for measures of forced vital capacity (FVC) and forced expiratory volume in 1 s (FEV_1) (11), whereas percent predicted FVC and FEV_1 were computed according to Crapo et al. (11). Participants were then fitted with a 12-lead electrocardiogram (Marquette Electronics, Milwaukee, WI) to monitor HR and rhythm and assumed an upright seated position on a cycle ergometer (Corival Lode B.V., Netherlands) where C_2H_2 uptake was performed for resting measures of $\dot{Q}_{\text{ACET}}$. Occurring at rest and continuing throughout CPET, breath-by-breath measures of ventilation and gas exchange were measured (CPX/D, Medical Graphics, St. Paul, MN) using a mid 5 of 7 filter, which included: $\dot{V}_{\text{O}_2}$, carbon dioxide output ($\dot{V}_{\text{CO}_2}$), respiratory exchange ratio (RER), respiratory rate (RR), tidal volume (V_T), minute ventilation ($\dot{V}_E$), and end-tidal CO_2 tension

($P_{ET}CO_2$). Ventilation and gas exchange data for these analyses were averaged over the final 30 seconds for each of the following time points: rest, 20 W, and the final fully completed exercise stage (i.e., peak exercise). The first three minutes of CPET were at 20 W, and increased thereafter by 20 W every three min at a pedal rate of 60–70 rpm until volitional fatigue.

Acetylene uptake—Measurement of $\dot{Q}_{ACET}$ was performed using standard rebreathe technique as previously described at rest as well as within the final 30 s of each CPET stage (36). Briefly, participants breathed into a non-rebreathing three-way pneumatic switching valve (Hans Rudolph, Kansas City, MO) connected to a pneumotachometer (Hans Rudolph, Kansas City, MO) and gas mass spectrometry (Perkin Elmer MGA-1100, Wesley, MA), which were integrated with custom analysis software for the determination of $\dot{Q}_{ACET}$ (36). The inspiratory port of the switching valve allowed for rapid switching for breathing room air or from a 5 L anesthesia rebreathe bag (Hans Rudolph, Kansas City, MO) containing 1 to 3 L of test gas [0.65 % C_2H_2 , 9 % helium [He], balance N_2 , and 35 % O_2] depending on initial V_T of the participant and the exercise intensity as previously described (36).

Gas concentrations were continuously measured beginning with the first end-expiration of the rebreathe period using gas mass spectrometry (36). Acetylene is not diffusion limited and does not readily bind to hemoglobin (Hb) while demonstrating high solubility in blood (8, 15, 16). Therefore, uptake of C_2H_2 in pulmonary blood demonstrates linearity with the rate of pulmonary capillary hemodynamics. As such, the rate of decrease in the end-tidal concentration of C_2H_2 with each breath to determine SV_{ACET} is calculated from the slope of the exponential end-tidal decrease in C_2H_2 concentration with respect to the blood-insoluble gas He (36).

According to the direct Fick equation and modifications of Grollman (15), Q_{ACET} , as well as SV_{ACET} using HR can be calculated using C_2H_2 as follows:

$$\dot{Q} = \frac{\dot{V} O_2 \cdot (C_2H_2)_{diff}}{(O_2)_{diff} \cdot \bar{x}C_2H_2 \cdot P_b - 47 \cdot 0.00974}, \text{ and } \times \frac{1}{HR} = SV$$

where $\dot{V}O_2$ is acquired using gas mass spectrometry, $(C_2H_2)_{diff}$ is the amount of C_2H_2 absorbed per L of blood during the time of sampling, $(O_2)_{diff}$ is the amount of O_2 absorbed during the sampling period, $\bar{x}C_2H_2$ is the average concentration of C_2H_2 during the sampling period, P_b is barometric pressure, 47 is the assumed vapor pressure of water in lungs (mm Hg), and 0.00974 is a numerical constant derived by combining the constants 760, 100, and 740, which represents the solubility coefficient for C_2H_2 in blood at body temperature (15).

Echocardiography—While image acquisition for SV via ECHO (SV_{ECHO}) took approximately 1.5 min in length (longer than C_2H_2 uptake periods), all measurements of SV_{ECHO} were acquired within the final 1.5 min of each CPET stage by the same trained research cardiac sonographer using standard 2-dimensional (2D) and pulsed wave tissue Doppler imaging (PW-TDI) using the Vivid 7® ECHO system (GE Medical Systems, Milwaukee, WI). Images were stored off-line for analyses using EchoPac® (v.5.0, GE

Medical Systems, Milwaukee, WI). Echocardiographic image analyses were blinded to participant condition (HFpEF vs. control) and activity state (rest vs. 20 W vs. peak exercise). The same sonographer as well as ECHO reader were used for all studies (both individuals, >10 years working with stress ECHO data acquisition and interpretation).

Measurements of SV_{ECHO} were obtained according to standards of the American Society of Echocardiography using the left ventricular outflow tract (LVOT) method, which included the apical- five- chamber (A5C) and parasternal- long- axis (PLAX) views (23). Briefly, in the PLAX view, 5 to 10 consecutive cardiac cycles of the LVOT at the aortic annulus was captured using gray-scale 2D imaging. Cross-sectional area (CSA) of the LVOT—proximal to the aortic valve points of insertion of the LVOT was measured at the time point of maximal separation of aortic valve leaflets near end-systole. The CSA value used for analysis was the average of five cardiac cycles and calculated as: $CSA = \pi \times r^2$, where r is the radius of the LVOT.

In the A5C view using PW-TDI imaging, with the pulsed-wave beam focused at the depth of the LVOT—proximal to the aortic valve points of insertion of the LVOT where the velocity-time integral (VTI) of the flow entering the LVOT during systole was measured using GE EchoPac®. The average VTI of five cardiac cycles was used for the calculation of SV as:

$$SV \text{ (cm}^3\text{)} = CSA \text{ (cm}^2\text{)} \times VTI \text{ (cm)}$$

The following additional calculations were performed: $O_2\text{pulse}$ ($\dot{V}O_2/\text{HR}$) and AVO_2 ($V_{O_2}/Q_{ACET}/10$ as well as $\dot{V}O_2/\dot{Q}_{ECHO}/10$) (37, 40). For statistical tests involving measures/predictions of SV, SV_{ECHO} , SV_{ACET} , and $O_2\text{pulse}$ were indexed to body surface area ($BSA = \sqrt{[(\text{height} \cdot \text{weight})/3600]}$). Rate of perceived exertion (RPE, Borg scale, 6 to 20) was assessed near the end of rest as well as each fully completed CPET stage.

Statistical Analyses

Parametric data are presented as mean $\pm$ SD. D'Agostino & Pearson omnibus testing confirmed normality of data, whereas Levene's testing confirmed homoscedasticity of data. In a progressive manner to test the validity (18) of techniques across SV_{ECHO} , SV_{ACET} , and $O_2\text{pulse}$, 1) univariate linear models via ordinary least squares regression (coefficient of determination, R^2) were performed separately for each group for the time periods of rest, 20 W, and peak exercise; 2) the goodness of fit equation (i.e., $y = mx + b$) and R^2 from each univariate regression model was used as the basis to derive the validity index (18) termed standard error of estimate [$SEE = SD \cdot \sqrt{(1-R^2)}$] for each technique in both groups at each of the above time points; and 3) as a subjective means of complementing the interpretive value of SEE tests (18), residuals and predicted values of SV derived from combined linear regression and SEE modeling were plotted for each time point of interest across techniques. Between group differences in R^2 were compared using Fisher's Z-test. Power of regression models and standard interpretation of R^2 were based on thresholds of Cohen (10): modest = 0.02, moderate = 0.15, and strong = 0.25; power = 0.80. Analysis of variance models with repeated measures were used to test between and within group differences for dependent variables of interest during CPET (group-by-time interaction terms set in all models). In the

event of significant interaction terms, Tukey post-hoc testing was performed to identify between-within pairwise differences. Two-tailed significance was determined using an alpha level set at 0.05. All computations were performed using SAS statistical software (v.9.4.) (SAS Institute, Cary, North Carolina).

RESULTS

Although there were no differences between controls and HFpEF for basic demographics (Table 1), HFpEF demonstrated a lower % predicted peak $\dot{V}O_2$ compared to controls, which was consistent with 89% of HFpEF classified as Weber-Janicki class C or D. While no HFpEF demonstrated a formal diagnosis of a pulmonary related comorbidity secondary to HFpEF, reduced resting airway function was present in HFpEF (Table 1). However, decreased resting airway function in HFpEF suggested for example, by low absolute FVC or FEV₁, did not explain the variance associated with SV_{ECHO}, SV_{ACET}, or O₂pulse at either 20 W (FVC, $R^2 = 0.03$, 0.00, or 0.00, respectively, all $P > 0.50$; and FEV₁, $R^2 = 0.00$, 0.02, or 0.01, respectively, all $P > 0.60$) or peak exercise (FVC, $R^2 = 0.01$, 0.02, or 0.05, respectively, all $P > 0.30$; and FEV₁, $R^2 = 0.00$, 0.02, or 0.05, respectively, all $P > 0.30$) in HFpEF.

In addition to the proportion of women and presence of hypertension in HFpEF demonstrating consistency with published literature (27, 29, 34, 39), all criteria used to classify patients as HFpEF for this study including LVEF (range 50 to 62 %), blood biochemistry, resting ECHO (e.g., $\uparrow$ left atrial volume index, $\uparrow$ mitral E/e' ratio, etc.), and pharmacologic therapy (>60% of HFpEF were on β -blockers or diuretics, whereas approximately one-third to two-fifths were on ACE inhibitors, ARBs, aspirin, or nitrates) were generally met in accordance with consensus statements and recommendations for the diagnosis of HFpEF (27, 29, 34, 39). All participants completed testing without adverse events. Routine pharmacologic therapy for HFpEF patients was maintained during testing.

Rest and CPET responses

While group-by-time interaction terms were significant for all dependent variables of interest presented in Table 2 (all $P < 0.05$), following post-hoc Tukey testing, there were no between group differences at rest for metrics descriptive of symptoms, ventilation, gas exchange, or cardiovascular function. Similarities between groups persisted at matched 20 W of exercise except for HFpEF demonstrating a higher RER compared to controls. At peak exercise, however, W were lower in HFpEF compared to controls despite similar RPE and RER (Table 2). Differences in peak workload were mirrored by blunted increases in $\dot{V}O_2$ (both absolute and weight indexed), $\dot{V}CO_2$, $\dot{V}_E$, and V_T at peak exercise in HFpEF compared to controls; whereas RR and P_{ET}CO₂ did not differ (Table 2).

Resembling the lack of between group ventilation and gas exchange differences at rest and at 20 W, there were no between group differences for any cardiovascular based metric in Table 2. However, peak exercise HR, SBP, MAP, and O₂pulse were lower in HFpEF compared to controls, whereas DBP, SV_{ECHO}, SV_{ACET}, AVO₂ (both ECHO and C₂H₂), and SaO₂ did not differ between groups.

R² and SEE between techniques for measurement or estimation of stroke volume

Illustrated in Figure 1(A to C) are modest strength univariate regressions at rest for controls when setting SV_{ECHO} to explain the variance in SV_{ACET} (R^2 did not change when flipping SV_{ACET} to predict SV_{ECHO} at any time point for controls or HFpEF; see Figure, Supplemental Digital Content 1, which demonstrates flipped regression plots), or when setting O_2 pulse to explain the variance in SV_{ECHO} or SV_{ACET} . By comparison, the same relationship testing performed in Figure 1, panels D to F, in HFpEF were strong at rest while differing in R^2 between groups for panels C vs. F ($P=0.02$), but not for A vs. D ($P=0.33$) or B vs. E ($P=0.11$). Likewise, the smaller standardized SEE output in Table 3 along with the more favorable data spread for each residual vs. predicted SV plot within HFpEF in Figure 1(G to I, which are commensurate with regressions in panels A to C or D to F in controls or HFpEF, respectively) were consistent with larger R^2 in HFpEF compared to controls.

Similar to rest, relationship testing performed at matched 20 W resulted in R^2 in controls (Figure 2, A to C) markedly weaker compared to the same relationships tested in HFpEF (Figure 2, D to F, respectively), which lead to differences between panels A vs. D ($P<0.01$) as well as C vs. F ($P=0.04$), but trending for D vs. E ($P=0.07$). Moreover, consistent with larger, but not significant increases in R^2 from rest to 20 W within HFpEF ($P=0.10, 0.52$, and 0.88 for panels D, E, and F, respectively, between Figure 1 and 2), which were accompanied by smaller SEE in HFpEF (Table 3), the splay of data in residual vs. predicted SV plots in panels G to I of Figure 2 appeared improved from rest to 20 W, whilst also remaining stronger compared to controls.

Lastly, persisting from 20 W, Figure 3(A to C) illustrates SV_{ECHO} could not explain the variance in SV_{ACET} , nor could O_2 pulse explain the variance in SV_{ECHO} in controls, whereas linearity between O_2 pulse and SV_{ACET} increased modestly from 20 W ($P=0.68$). Alternatively, while strength of R^2 presented in Figure 3, panels D and F, carried over from 20 W to peak exercise within HFpEF, less of the variance in SV_{ECHO} appeared to be explained by O_2 pulse in Figure 3E vs. 2E ($P=0.28$). Nevertheless, smaller standardized SEE across SV_{ECHO} , SV_{ACET} , and O_2 pulse at both rest and 20 W in HFpEF vs. controls was maintained at peak exercise, to which residual vs. predicted SV plots in Figure 3(G to I) appeared to remain consistent with the more favorable spread of data at 20 W in HFpEF compared to controls.

DISCUSSION

As an important clinical diagnostic tool, the prognostic power of CPET is strengthened when accompanied by objective measurements of exercise cardiac hemodynamics in patients with HFpEF (1, 5, 6, 17, 24, 29, 34). Therefore, this study aimed to address a clinically impactful gap in knowledge by testing in a generalizable laboratory setting the (quasi)concurrent validity of noninvasive measures or estimates of exercise SV using a more recognized ECHO approach against lesser understood C_2H_2 uptake and O_2 pulse methods in patients with HFpEF.

These clinically translational data suggest exercise SV can be measured or predicted with similar levels of precision using SV_{ECHO} , SV_{ACET} , or O_2 pulse in patients with HFpEF. This

study did not employ invasive cardiac hemodynamic techniques to establish true construct validity of SV_{ECHO} , SV_{ACET} , or O_2 pulse as noninvasive approaches to measure or estimate exercise SV in HFpEF. However, it is worthwhile noting that in various studies across the adult health spectrum (e.g., pulmonary hypertension, reduced ejection fraction HF, etc.), all three noninvasive approaches studied at present have been tested, while demonstrating validity with respect to ‘Gold’ standard methodology at rest and/or exercise conditions (2, 16, 19, 20, 32, 33, 37, 40). Nevertheless, although we appreciate the physiologic-based constructs supporting the utility of the present techniques in patients with pulmonary and/or cardiac limitations, we recognize the direct translation from previous reports suggesting robust validity of SV_{ECHO} , SV_{ACET} , or O_2 pulse cannot at this time be fully recognized in HFpEF. Therefore, in a translational and clinically impactful manner, while also a necessary first step towards proper construct validation of the present methods in HFpEF, these data lend immediate support for the high-priority need to confirm the present observations in the setting of a more complex exercise catheterization study in patients with HFpEF.

While the direct Fick method involving catheterization is the ‘Gold’ standard for measuring cardiac hemodynamics, there are limitations associated with this approach that aid in providing firm reasoning for the aims tested in this study as well as in previous reports (2, 16, 19, 20, 32, 33, 37, 40). For example, although highly trained specialists are tasked with performing catheterization procedures, patients are inherently exposed to increased risks associated with these techniques. This includes modest, but meaningful susceptibility to post-operative bleeding, cardiac events (e.g., myocardial infarction), and in worst cases, patient death (3). Moreover, and of secondary importance beyond increased patient risk, because of complexities involved with cardiac catheterizations, resources that must be dedicated to performing these types of procedures are costly to the patient as well as medical center (e.g., personnel, facilities, etc.), which contributes to the lack of routine availability of cardiac catheterization across institutions. Lastly, while it is recognized that interpretability of cardiac, pulmonary, and vascular responses during CPET is most robust and, hence, clinically preferred while performed in the upright position (e.g., treadmill or ergometry), it is not standard for centers that have the resources and willingness to perform cardiac catheterization during CPET that treadmill testing or upright ergometry are performed as opposed to supine ergometry. With this, although we are unaware of any study to date that specifically compares the clinical diagnostic strength of supine versus upright exercise concomitant with invasive cardiac hemodynamic testing in HFpEF, it should be considered when interpreting supine CPET that both hemodynamics and ventilation may be altered based on effects of body position (e.g., upward diaphragm shift, central to peripheral blood flow distribution, etc.) independent of pathologic state (22), whereby the full-extent of cardiovascular impairment may be falsely lessened.

Echocardiography in HFpEF—In acknowledging ECHO for the noninvasive assessment of cardiac hemodynamics is likely the most utilized noninvasive technique in clinical and research settings, the presumptive link between prevalence of technique use and invariability in accuracy, precision, and reliability are not mutually exclusive to ECHO (9, 23, 25, 32). While a comprehensive review of the strengths and limitations of ECHO are beyond the scope of this report, in brief, it has been suggested that despite validation studies and

established guidelines (9, 19, 23, 25, 28, 32), variability associated with the process of image acquisition and post-processing calculation of Q and SV is contributed to by confounding degrees of human factors (e.g., sonographer experience), test setting (e.g., rest vs. exercise, or supine vs. upright exercise), patient factors (e.g., chest adiposity), acquisition protocols, and technology platforms (9, 23, 25, 28, 32). Nevertheless, despite proposed documented limitations, which may influence the utility of ECHO for exercise SV measurement in HFpEF, confounding sources of variability may be lessened when techniques are conducted by experienced sonographers in combination with skilled ECHO readers (9, 19, 23, 25, 28, 32).

Acetylene uptake in HFpEF—In light of recent observations in HFpEF, in addition to previous work in reduced ejection fraction HF, we acknowledge that the potential impact changes in lung diffusing capacity and pulmonary capillary volume might have on measures of gas uptake within lungs cannot be overlooked (17, 30, 35). For example, the presence of sub-clinical or overt pulmonary edema could theoretically influence the accuracy of SV_{ACET} since the perfusion sensitive inert gas uptake technique is dependent on free diffusion of C_2H_2 across the alveolar–capillary membrane. However, despite low lung diffusing capacity in HFpEF (17, 30) similar to that observed in reduced ejection fraction HF (35), these data suggest that the relative precision of SV measured using C_2H_2 is linearly related to SV_{ECHO} (the latter of which is not directly influenced by low lung diffusing capacity) while also appearing closely related to O_2 pulse during CPET in HFpEF.

Although we support with tests of concurrent validity (i.e., R^2 , SEE, etc. (18)) encouraging data comparing SV_{ACET} , SV_{ECHO} , and O_2 pulse during CPET in HFpEF, we recognize that cross-technique validity in the setting of our paradigm may have a higher likelihood of being influenced by changes in any physiologic construct underpinning the relevant method as well as the time period of measurement (e.g., rest, 20 W, or peak exercise). For example, there appeared to be modest inconsistencies in the magnitude of explained variance between O_2 pulse and SV_{ECHO} versus comparisons of O_2 pulse and SV_{ACET} or SV_{ECHO} and SV_{ACET} when transitioning from rest through to peak exercise in HFpEF. As such, while only theoretical based on these data, it could be reasoned that because of intrinsic physiologic and anatomic adjustments of the pulmonary system that are provoked by exercise, which may include for example, increases in pulmonary capillary recruitment, collateral recruitment of airways, increased blood flow to lung apices, and alveoli unfolding; favorable changes in alveoli—pulmonary capillary interactions similarly affected SV_{ACET} and O_2 pulse without having any direct effect on SV_{ECHO} . These global pulmonary interactions may have led to more consistency in the rise between SV_{ACET} and O_2 pulse observed in HFpEF. By contrast, because of major differences in gas properties between C_2H_2 and O_2 , such as the lesser Bunsen solubility coefficient in blood demonstrated by O_2 (<30 times that of C_2H_2) (8, 31), it may be that lower lung diffusing capacity (30) while also considering the potential for maldistribution of pulmonary capillary blood volume is more influential to $V O_2$ and O_2 pulse dynamics than what would be expected by C_2H_2 at peak exercise in HFpEF. Nevertheless, while these hypotheses aimed to explain some of the modest variability with respect to interactions between O_2 pulse and SV_{ECHO} cannot be confirmed at present, we are encouraged by the consistency of SV_{ACET} in relation to both O_2 pulse and SV_{ECHO} as a tool

to measure SV during CPET in HFpEF. It follows, therefore, that with advanced construct validation testing, SV_{ACET} may fulfill an immediate need-based gap for the routine clinical evaluation of exercise cardiac hemodynamics in HFpEF.

Comparison of inert gas uptake studies—In reaffirming our appreciation for previous studies describing various high blood soluble inert gas uptake techniques for measuring cardiac hemodynamics, we suggest these data are consistent with Saur et al. (33) who demonstrated across patients with pulmonary disease that lung diffusing capacity does not significantly influence measures of Q via N_2O uptake (physiologically relevant because of effects on perfusion sensitivity; compared to C_2H_2 , N_2O demonstrates a ~two-fold lower Bunsen solubility coefficient in blood (15)). Likewise, the present observations in HFpEF support and extend the work of others who have reported that C_2H_2 (16, 20, 36), N_2O (2, 26), and/or CO_2 (13) techniques can be used to measure Q and SV in conditions of rest as well as exercise in various conditions of adult health or disease.

O₂pulse in HFpEF—With acknowledgement that there are studies demonstrating strong linearity between O₂pulse and SV during exercise in adults (4, 12, 37, 40), it is also noteworthy that with marked variability in AVO_2 and/or HR, it is unlikely O₂pulse can be used to predict SV. Intriguingly, however, while it is suggested that AVO_2 and/or HR responses may be abnormal in individuals such as patients with HF who are strong candidates to demonstrate pathology of both central and peripheral organ systems (1, 21), it is remarkable that Taylor et al. (37) demonstrated robust linearity between O₂pulse and SV during exercise in reduced ejection fraction HF. Likewise, assuming O₂pulse is grounded in meeting assumptions of the direct Fick equation where Q aligns with $V O_2$ as intact AVO_2 function persists, it could be interpreted that observations of Abudiab et al. (1) imply there is linearity between SV and O₂pulse in HFpEF. In using cardiac catheterization techniques, they observed absolute increases in Q and $V O_2$ paralleled one another from rest to peak exercise in the presence of an adaptable AVO_2 response to exercise (1). Thus, the present observations offer support for the need to confirm via advanced exercise cardiac catheterization studies that there is utility in using O₂pulse to predict exercise SV in HFpEF.

Clinical Implications—In the absence of reported adverse events directly associated with the ECHO technique as implemented in this study, it is also relevant that it has been demonstrated that inert gas uptake (i.e., C_2H_2) at gas mix concentrations used in the study is safe in humans (38). Therefore, with the intent of this study aimed at providing an advanced scientific and methodologic examination of noninvasive techniques for immediately measuring or estimating exercise SV in HFpEF, these data suggest that relative to one another, SV_{ACET} , SV_{ECHO} , and O₂pulse can be used with similar levels of precision during CPET to measure or predict SV in patients with HFpEF. In addition to being cost-effective, noninvasive, and more widely available to patients and practitioners, these data with support from sound scientific reasoning, suggest it is feasible (via approaches tested presently) that noninvasive assessment of exercise cardiac hemodynamics may be routinely available to quickly grade syndrome severity and response to therapy in HFpEF.

The need to demonstrate generalizable lab-based efficacy across the present noninvasive techniques all tested within the same study of HFpEF is well-supported as previous work

from our group and others demonstrate there is indeed an important role for cardiac-centric contributors to exercise intolerance in HFpEF (1, 5, 6, 24, 29). Again, while we appreciate hypotheses generating suggestions that there are important contributions from pathology of the periphery to exercise intolerance in HFpEF (21), we cannot emphasize to a greater extent that these data have an immediate high-priority translational impact with respect to strengthening the clinical decision making process linked to CPET in HFpEF. Likewise, additional availability to cardiac hemodynamic responses to CPET will serve to improve our understanding of the role that impaired cardiac hemodynamics play in the pathophysiology exercise intolerance in HFpEF.

Limitations—In addition to previously discussed technique specific limitations, we recognize that in all cases exercise-related interstitial pulmonary edema and $\dot{V}_A/Q$ mismatch may occur in HFpEF patients. While abnormal shifts in perfusion such as extra-pulmonary R-to-L shunt may be detectable via ECHO (e.g., obvious cases of septal defects), the presence of pulmonary edema and/or inhomogeneous ventilation has the potential to not only affect ventilation-based techniques, but also the mirrored projection and reflection of PW beams associated with PW-TDI ECHO and, subsequently, the quality of image acquisition associated with ECHO (14, 19, 23, 25, 32). Nevertheless, while cardiac catheterization studies were not performed at present, we are confident that we identified and recruited patients for this study who did not demonstrate overt signs or symptoms to suggest the presence of these or other cardiac and pulmonary limitations, which could lead to marked technique influencing $\dot{V}_A/Q$ mismatch (see Figure, Supplemental Digital Content 2, which demonstrates surrogate estimates of whole lung $\dot{V}_A/\dot{Q}$ matching cannot explain variance associated with SV_{ACET} , SV_{ECHO} , and O_2 pulse at any time point in HFpEF).

Moreover, it could be interpreted from studies of lung diffusing capacity, whilst being supported by validation studies of Q via inert gas uptake (2, 13, 16, 26, 33), that in the absence of severe impediment (e.g., presence of marked exercise provocation of accumulated interstitial pulmonary edema, parenchymal disease, etc.) of the free diffusion of high blood soluble inert gas across the alveolar—capillary membrane barrier, it is not likely that low lung diffusing capacity would be influential to the perfusion sensitive inert gas uptake technique for the measurement of exercise SV in HFpEF (8, 16, 26, 33).

Additionally, as previously discussed, a ‘Gold’ standard criterion technique (e.g., direct Fick) that was in addition to our three noninvasive techniques was not included in this study and, therefore, construct validity across SV_{ECHO} , SV_{ACET} , and O_2 pulse in this experimental paradigm cannot be confirmed with these data. Again, however, while the exact translation to this study based on previous patient centered studies of noninvasive approaches to assess cardiac hemodynamics is not clear, we note that individual studies of each of the techniques employed in this study have been tested for validity against true criterion techniques, whilst accompanied by favorable observations (2, 13, 16, 19, 26, 33, 37).

Lastly, relevant to the discussion of O_2 pulse, it must be acknowledged that effects of rate-limiting therapy (i.e., β -blockers) on a blunted rise in HR may in-part account for some unexplained variance associated with O_2 pulse during CPET in HFpEF. Although we would not expect to observe marked increases in SV_{ECHO} or SV_{ACET} via an improved Frank-Starling effect given the afterload dependence of HF, a β -blocker effect may have plausibly

led to an over-estimation of SV by O₂pulse as opposed to our interpretations suggesting under-estimation. Nevertheless, while a deeper understanding of whether the influence of pharmacologic therapy markedly confounded the present observations is of certain intrigue at a mechanistic level, it is of greater applicability to the larger clinical implications of these data that real-world HFpEF patients typically perform CPET in an optimally medicated state, which suggests these data may be more generalizable to a classical clinical setting.

CONCLUSIONS

By implementing physiologic-based scientifically rigorous and sound methodologic testing of SV_{ACET}, SV_{ECHO}, and O₂pulse for the measurement or prediction of SV during CPET in patients with HFpEF, these data suggest each noninvasive technique can be used at similar levels of precision in a test setting that is generalizable and immediately clinically relevant to patients with HFpEF. Therefore, because CPET accompanied by objective assessment of cardiac hemodynamics has immediate clinical translational implications for its key role in assisting in the clinical classification of HFpEF (1, 5, 6, 17, 24, 29), the scientific aims tested in this study constitute a primary original step in lending physiologic-based support for the need to perform construct validation studies of each of the present methods while involving ‘Gold’ standard techniques during CPET in HFpEF.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Figure 1.

Ordinary least squares univariate linear regressions between noninvasive techniques for measuring or predicting stroke volume (SV) at rest in healthy controls (CTL) or patients with heart failure with preserved ejection fraction (HFpEF). Solid line represents the goodness of fit line with 95% confidence limits as dotted lines. Goodness of model fit expressed as coefficient of determination (R^2) from Cohen (10); modest = 0.02, moderate = 0.15, and strong = 0.25. For panels **A to F**, the variable set on the abscissa was considered the practical method, whereas the variable set on the ordinate was considered the quasi-criterion method. ECHO = echocardiography; C₂H₂ = acetylene uptake; O₂pulse = oxygen pulse. Panels **G to I** align vertically with panels **A to C** or **D to F** for CTL or HFpEF, respectively. Predicted (abscissa), represents predicted SV using the respective goodness of fit equation ($y = mx + b$) from univariate regressions in panels **A to F**, whereas residuals (ordinate), represent the difference between observed SV (via either ECHO or acetylene) and predicted SV. In panel **G**, for corresponding regression plots where ECHO was set as the criterion see the Figure in Supplemental Digital Content 1.

Figure 2.

Ordinary least squares univariate linear regressions between noninvasive techniques for measuring or predicting stroke volume (SV) at matched 20 W exercise in healthy controls (CTL) or patients with heart failure with preserved ejection fraction (HFpEF). Solid line represents the goodness of fit line with 95% confidence limits as dotted lines. Goodness of model fit expressed as coefficient of determination (R^2) from Cohen (10); modest = 0.02, moderate = 0.15, and strong = 0.25. For panels **A to F**, the variable set on the abscissa was considered the practical method, whereas the variable set on the ordinate was considered the quasi-criterion method. ECHO = echocardiography; C₂H₂ = acetylene uptake; O₂pulse = oxygen pulse. Panels **G to I** align vertically with panels **A to C** or **D to F** for CTL or HFpEF, respectively. Predicted (abscissa), represents predicted SV using the respective goodness of fit equation ($y = mx + b$) from univariate regressions in panels **A to F**, whereas residuals (ordinate), represent the difference between observed SV (via either ECHO or acetylene) and predicted SV. In panel **G**, for corresponding regression plots where ECHO was set as the criterion see the Figure in Supplemental Digital Content 1.

Figure 3.

Ordinary least squares univariate linear regressions between noninvasive techniques for measuring or predicting stroke volume (SV) at peak exercise in healthy controls (CTL) or patients with heart failure with preserved ejection fraction (HFpEF). Solid line represents the goodness of fit line with 95% confidence limits as dotted lines. Goodness of model fit expressed as coefficient of determination (R^2) from Cohen (10); modest = 0.02, moderate = 0.15, and strong = 0.25. For panels **A to F**, the variable set on the abscissa was considered the practical method, whereas the variable set on the ordinate was considered the quasi-criterion method. ECHO = echocardiography; C₂H₂ = acetylene uptake; O₂pulse = oxygen pulse. Panels **G to I** align vertically with panels **A to C** or **D to F** for CTL or HFpEF, respectively. Predicted (abscissa), represents predicted SV using the respective goodness of fit equation ($y = mx + b$) from univariate regressions in panels **A to F**, whereas residuals (ordinate), represent the difference between observed SV (via either ECHO or acetylene) and predicted SV. In panel **G**, for corresponding regression plots where ECHO was set as the criterion see the Figure in Supplemental Digital Content 1.

Table 1**Participant characteristics**

	Controls	HFpEF	P-value
N	18	18	
Sex, male/female (%)	5/13 (72)	5/13 (72)	0.99
Age, years	63 ± 9	69 ± 9	0.09
Height, m	1.68 ± 0.11	1.65 ± 0.09	0.48
Weight, kg	84 ± 10	93 ± 10	0.20
BMI, kg·m ⁻²	30 ± 3	34 ± 3	0.07
BSA, m ²	1.97 ± 0.26	2.06 ± 0.26	0.33
Peak V O ₂ , % of predicted	92 ± 9	58 ± 9	<0.01
Left ventricular ejection fraction (%)		62 ± 3	
Hypertension, N (%)		15 (83)	
Plasma BNP, pg·mL ⁻¹		125 ± 79	
Creatinine, mg·dL ⁻¹		1.41 ± 0.40	
eGFR, mL·min ⁻¹ per 1.73 m ²		51 ± 11	
Hemoglobin, g·dL ⁻¹		12.94 ± 0.61	
Medications			
ACE inhibitor		8 (44)	
Angiotensin II receptor blockers		6 (33)	
β-blocker		12 (66)	
Ca ²⁺ channel blocker		4 (22)	
Digitalis		0 (0)	
Aspirin		7 (39)	
Nitrates		5 (28)	
Diuretics		14 (78)	
Resting Echocardiography			
Left atrial volume index, mL·m ⁻²		41 ± 16	
Mitral E/A ratio		1.84 ± 1.22	
Mitral E/e' ratio		18.9 ± 7.9	
Interventricular septum thickness, mm		10.88 ± 2.42	
Posterior wall thickness, mm		10.44 ± 2.04	
Resting Spirometry			
FVC, L	3.47 ± 0.90	2.36 ± 0.63	<0.01
FVC, % predicted	100 ± 15	73 ± 15	<0.01
FEV ₁ , L	2.79 ± 0.68	1.90 ± 0.50	<0.01
FEV ₁ , % predicted	101 ± 12	75 ± 17	<0.01
FEV ₁ /FVC	0.81 ± 0.06	0.81 ± 0.08	0.83
FEV ₁ /FVC, % predicted	101 ± 7	102 ± 9	0.51

Data presented as mean ± SD or count (N) and percentage (%). BMI = body mass index; BNP = B-type natriuretic peptide; BSA = body surface area; eGFR = estimated glomerular filtration rate; FVC = forced vital capacity; FEV₁ = forced expiratory volume in 1 s; V O₂ = pulmonary oxygen uptake.

Table 2
Rest, 20 W, and peak exercise subjective and objective responses in controls or heart failure with preserved ejection fraction

	Rest			20 W			Peak Exercise		
	Controls	HFpEF	P	Controls	HFpEF	P	Controls	HFpEF	P
Workload, W	-	-	-	-	-	-	100±10	52±11	<0.01
RPE, Borg 6 to 20	6±0	6±0	0.99	8±1	11±1	<0.01	16±1 [*]	16±1 [*]	0.80
Ventilation and gas exchange									
V O ₂ , L·min ⁻¹	0.29±0.03	0.30±0.04	0.99	0.80±0.10	0.81±0.15	0.99	1.60±0.24 [*]	1.11±0.14 [*]	<0.01
V O ₂ , mL·kg ⁻¹ ·min ⁻¹	3.55±0.46	3.29±0.82	0.86	9.51±1.15	8.86±1.42	0.66	19.18±1.73 [*]	12.69±3.23 [*]	<0.01
V CO ₂ , L·min ⁻¹	0.22±0.04	0.24±0.05	0.92	0.60±0.14	0.68±0.17	0.61	1.74±0.24 [*]	1.19±0.17 [*]	<0.01
RER	0.76±0.05	0.81±0.04	0.43	0.76±0.05	0.83±0.13	0.02	1.09±0.04 [‡]	1.07±0.04 [‡]	0.34
V E, L·min ⁻¹	8.5±0.4	9.5±0.6	0.71	20.0±2.9	24.0±5.3	0.20	58.6±6.4 [*]	42.9±6.3 [*]	<0.01
V T, L·min ⁻¹	0.59±0.14	0.66±0.13	0.89	0.87±0.12	0.91±0.15	0.99	1.63±0.10 [*]	1.21±0.14 [*]	0.03
RR, breaths·min ⁻¹	15±1	15±2	0.99	23±4	28±8	0.28	37±3 [*]	36±4 [*]	0.99
P _{ET} CO ₂ , mm Hg	36±2	35±1	0.15	38±3	36±4	0.58	36±2 [‡]	34±2	0.89
Cardiovascular responses									
HR, beats·min ⁻¹	68±5	68±13	0.99	93±14	94±15	0.99	147±10 [*]	119±11 [*]	<0.01
SBP, mm Hg	131±8	129±9	0.99	137±18	135±22	0.99	197±11 [‡]	158±13 [‡]	<0.01
DBP, mm Hg	77±4	69±4	0.15	76±11	70±10	0.76	87±9 [‡]	76±6	0.32
MAP, mm Hg	95±5	89±4	0.59	96±12	92±11	0.89	124±10 [‡]	103±7 [‡]	<0.01
SV (ECHO), mL·m ⁻²	32±7	36±10	0.73	42±8	38±10	0.53	48±8 [#]	39±13	0.07
SV (C ₂ H ₂), mL·m ⁻²	32±6	31±10	0.76	43±10	40±11	0.32	46±9 [#]	41±11	0.24
O ₂ pulse, mL·m ⁻² ·beat ⁻¹	2.22±0.34	2.18±0.50	0.77	4.39±0.64	4.24±0.70	0.49	5.56±0.61 [*]	4.79±0.92 [*]	0.02
AVO ₂ (ECHO), mL·dL ⁻¹	7.09±0.94	6.20±0.79	0.08	10.32±1.19	11.45±1.28	0.61	12.30±1.50 [*]	12.94±2.13 [*]	0.55
AVO ₂ (C ₂ H ₂), mL·dL ⁻¹	7.24±0.88	7.28±0.67	0.93	10.03±1.74	10.73±1.72	0.83	12.95±1.08 [*]	12.24±1.04 [*]	0.32
SaO ₂ , %	98±2	98±2	0.99	97±2	97±2	0.74	97±2	95±6	0.73

Raw data are presented as mean ± SD. Controls (N=18); HFpEF, heart failure with preserved ejection fraction (N=18). RPE=rate of perceived exertion; V O₂=oxygen uptake; V CO₂=carbon dioxide output; RER=respiratory exchange ratio; V E=minute ventilation; VT=tidal volume; RR= respiratory rate; PETCO₂=end-tidal partial-pressure carbon dioxide; HR=heart rate; SBP=systolic blood pressure; DBP=diastolic blood pressure; MAP=mean arterial pressure; SV =stroke volume; C₂H₂=acetylene gas; O₂pulse=oxygen pulse; AVO₂=arterial-mixed venous oxygen difference; SpO₂=oxygen saturation.

* P<0.05, all differed within group;

‡ P<0.05, within group, peak vs. 20 W and rest;

‡ P<0.05, within group, peak vs. 20 W;

P<0.05, within group, peak vs. rest.

Table P-values and symbols reflect Tukey post-hoc correction.

Table 3

Standard error of estimate (95% confidence limits) for measured or predicted stroke volume at rest, 20 W, or peak exercise.

Criterion (Y) vs. Practical (X)	Rest		20 W		Peak exercise	
	SEE	Std.SEE	SEE	Std.SEE	SEE	Std.SEE
C₂H₂ vs. ECHO						
Controls	6.47 (4.82, 9.84)	1.00 (0.74, 1.52)	9.74 (7.26, 14.83)	1.01 (0.75, 1.53)	9.43 (7.03, 14.36)	0.99 (0.74, 1.51)
HFpEF	8.40 (6.25, 12.78)	0.86 (0.64, 1.31)	6.20 (4.62, 9.44)	0.57 (0.42, 0.87)	7.01 (5.22, 10.67)	0.64 (0.48, 0.97)
ECHO vs. C₂H₂						
Controls	6.68 (4.98, 10.17)	1.00 (0.74, 1.52)	8.27 (6.16, 12.59)	1.01 (0.75, 1.53)	8.19 (6.10, 12.46)	0.99 (0.74, 1.51)
HFpEF	9.02 (6.71, 13.72)	0.86 (0.64, 1.31)	5.75 (4.28, 8.74)	0.57 (0.42, 0.87)	8.07 (6.01, 12.28)	0.64 (0.48, 0.97)
ECHO vs. O₂pulse						
Controls	6.89 (5.13, 10.49)	1.03 (0.77, 1.56)	8.18 (6.09, 12.45)	0.99 (0.74, 1.51)	8.30 (6.18, 12.64)	1.01 (0.75, 1.53)
HFpEF	8.60 (6.40, 13.09)	0.82 (0.61, 1.25)	7.14 (5.32, 10.87)	0.71 (0.53, 1.08)	11.39 (8.48, 17.43)	0.90 (0.67, 1.37)
C₂H₂ vs. O₂pulse						
Controls	6.39 (4.76, 9.73)	0.99 (0.73, 1.50)	8.88 (6.61, 13.51)	0.92 (0.68, 1.39)	7.99 (5.95, 12.16)	0.84 (0.63, 1.28)
HFpEF	5.79 (4.31, 8.82)	0.60 (0.44, 0.91)	6.12 (4.56, 9.32)	0.56 (0.42, 0.85)	6.77 (5.04, 10.30)	0.62 (0.46, 0.94)

Data presented as absolute or standardized (Std) standard error of estimate (SEE) with (95% confidence limits). Controls (N = 18). HFpEF = heart failure with preserved ejection fraction (N = 18). C₂H₂ = acetylene uptake; ECHO = echocardiography; O₂pulse = oxygen pulse. The SEE can be interpreted as given a predicted measurement of stroke volume (SV) using a practical method relative to a set quasi-criterion method (i.e., C₂H₂ or ECHO), it can be assumed that the predicted SV is likely to vary within plus or minus the SEE value on the SV measured using the quasi-criterion method with 68% confidence.