

SUPPLEMENTAL METHODS

Cardiopulmonary Exercise Testing

Cardiopulmonary Exercise Testing (CPET) was conducted using an electronically braked upright cycle ergometer to maximal tolerance incorporating an individualized work rate. An experienced blinded investigator carried out all exercise testing. Breath-by-breath gas exchange analysis (VIASYS; MasterScreen CPX, Hoechberg, Germany) was performed. Before each test, the equipment was calibrated using standard reference gases and a 3-L syringe. Care was taken to ensure that each study patient understood the maximal exercise test protocol. This included explanation of the anticipated early symptoms of lactic acid–associated leg fatigue or discomfort that must not lead to premature cessation of pedaling or incremental loading. Each patient rested for 3 minutes, followed by 3 minutes of unloaded pedaling prior to workload increments, and a continuous 12-lead electrocardiogram was recorded. The test was terminated at maximal exhaustion, accompanied by attainment of a respiratory exchange ratio (RER; ratio of CO₂ production to O₂ consumption) of ≥ 1.15 .

Individualized predicted VO₂Max was calculated according to the formula proposed by Wasserman et al.³³ The VO₂ at the point of anaerobic threshold (AT) was determined by the V-slope method in conjunction with analyses of the ventilatory equivalents (VE/VO₂ and VE/VCO₂) and end-tidal gas tension (PETO₂ and PETCO₂) plots.³³ The corresponding milliliters of VO₂ per minute at AT was expressed as a percentage of predicted VO₂Max.^{23, 37} The latter adjusts for age, sex, weight, and height.

Echocardiography

Two-dimensional Doppler and tissue Doppler transthoracic echocardiography were performed using the Vivid 7 (GE Healthcare) ultrasound system according to a standardized study protocol. Calculations included LV ejection fraction according to quantitative biplane Simpson method, LV end-diastolic volume (LVEDV), LV mass, and left atrial volume. Mass and volume measures were indexed to body surface area. Tissue Doppler imaging of the mitral annulus sequentially at the

lateral and septal annular sites were obtained from the apical 4-chamber view. The ratio of early transmitral flow velocity to averaged annular (septal and lateral) mitral velocity ($E/\text{mean } e'$) was taken as an estimate of LV filling pressure. All measurements were undertaken according to the American Society of Echocardiography guidelines³⁶ and analyzed offline (EchoPAC; GE Healthcare) by a blinded investigator.