Energy for Myocardial Ca^{2+} Handling per Beat Increases with Heart Rate in Excised Cross-circulated Canine Heart

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Objective: Although tachycardia is well known to increase cardiac oxygen consumption (Vo_2) per min, the relationship between Vo_2 for excitation-contraction (E-C) coupling per beat and heart rate change over its full working range still remains controversial.
Methods: To elucidate this relationship, we varied heart rate over a reasonably wide range (60-180 beat/min) and studied the relationship between left ventricular (LV) Emax (load-independent contractility index), PVA (pressure-volume area)-independent Vo_2, and basal metabolic Vo_2 in nine excised, cross-circulated canine hearts.
Results: PVA-independent Vo_2 per min significantly increased linearly with increasing heart rate while Emax remained unchanged. Basal metabolic Vo_2 per min was measured under KCl arrest. E-C coupling Vo_2 per min obtained by subtracting the constant basal metabolic Vo_2 from the PVA-independent Vo_2 also significantly increased linearly with increasing heart rate. However, PVA-independent Vo2 per beat significantly decreased with increasing heart rate. In contrast, E-C coupling Vo_2 per beat, as well as that normalized to Emax, slightly but significantly increased with increasing heart rate.
Conclusion; The E-C coupling energy for myocardial Ca^{2+} handling increases with heart rate despite constant contractility in the left ventricle of the excised cross-circulated canine heart.

Key words: end-systolic pressure-volume relation, excitation-contraction coupling, myocardial oxygen consumption, ventricular energetics

INTRODUCTION

Although there have been many studies on the effects of heart rate on intra-myocardial Ca^{2+} handling for excitation-contraction (E-C) coupling in beating hearts, the relationship between heart rate and E-C coupling energetics per beat still remains controversial [1–5]. Increasing heart rate may either enhance or depress cardiac contractility (Bowditch or Woodworth phenomena, respectively) [6, 7]. These phenomena have been accounted for by augmented or reduced amounts of Ca^{2+} releasable from sarcoplasmic reticulum (SR) respectively [8–11]. Both the sarcoplasmic reticulum (SR) Ca^{2+} pump (SERCA2) and the sarcolemmal Na^+/Ca^{2+} exchanger (NCX) remove sarcoplasmic Ca^{2+} released from the SR in each beat by consuming ATP [8].

SERCA2 normally sequesters most of the Ca^{2+} in a stoichiometry of $2Ca^{2+}$: 1ATP [8, 12], whereas the NCX coupled with the Na^+-K^+ pump extrudes the remaining Ca^{2+} in a stoichiometry of $1Ca^{2+}$: 1ATP [8]. The fraction of Ca^{2+} removed internally by SERCA2 is the intra-myocardial Ca^{2+} recirculation fraction (RF) [13]. Although the sarcolemmal Ca^{2+} pump extrudes an auxiliary fraction of Ca^{2+}, its stoichiometry is the same as the NCX [14]. Therefore, this pathway can be included energetically in the NCX. When changes in heart rate vary the RF, the energy consumption for E-C coupling varies. We have already found that the RF increases with heart rate [4].

In this study, we found that increasing heart rate over the reasonably wide range from 60 to 180 beats/min did not affect Emax, but significantly increased E-C coupling Vo_2 per beat. This is the first finding of a significant dependence of the E-C coupling Vo_2 per beat on a wide range of heart rates in cardiac mechano-energetics. Therefore, E-C coupling energy for myocardial Ca^{2+} handling increases despite an absence of enhanced contractility in the blood-perfused canine heart. We discuss the possible mechanisms underlying this finding below.

METHODS

Surgical preparation

We used nine excised, cross-circulated canine heart preparations of the same type as that normally used in our cardiac mechano-energetic studies [4, 15–20]. Studies were conducted in conformity with the Guiding Principles for Research Involving Animals and Human Beings endorsed by the American

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Physiological Society. Materials and methods for setting up and maintaining the heart preparation have been described in detail elsewhere [4, 15–20].

Briefly, two adult mongrel dogs (10–18 kg) were anesthetized with pentobarbital sodium (25 mg/kg iv) and fentanyl (0.1 mg/h iv) after premedication with ketamine hydrochloride (25 mg/kg im). Dogs were intubated, air-ventilated, and heparinized (15,000 or 10,000 units for a larger or smaller dog respectively, iv). The larger dog was used as the metabolic supporter. Its bilateral common carotid arteries and the right external jugular vein were cannulated with the arterial and venous cross-circulation tubes, respectively. The smaller, heart donor dog was thoracotomized. Its left subclavian artery was cannulated with the arterial cross-circulation tube and the right atrium via the venous tube. All systemic and pulmonary vascular connections to the donor heart were ligated.

The cross-circulated heart was excised from the chest without stopping the coronary circulation. The left atrium of the excised heart was opened and all the LV chordae tendineae were cut. To decrease the minimal heart rate as much as possible, a complete atrio-ventricular block was induced by focal injection of 0.2–0.5 ml 36% formaldehyde. Para-Hisian pacing was performed with a bipolar electrode to minimize pacing-induced ventricular asynergy. A flabby balloon with an unstretched volume of 50 ml was fitted into the LV chamber. The balloon was connected to our custom-made volume servo pump (AR-Brown, Tokyo, Japan), and both the balloon and the water housing of the pump were primed with water [4, 15–20].

The servo pump enabled us to accurately measure and precisely control LV volume (LVV). LV pressure (LVP) was measured with a miniature pressure gauge (model P-7, Konigsberg Instruments, Pasadena, CA, USA) placed within the apical end of the balloon. Temperature of the heart was maintained at 37ºC. LV epicardial electrocardiogram (ECG) was recorded with a pair of screw-in electrodes. Coronary arteriovenous O_2 content difference was measured with a custom made analyzer (PWA-200S, Shoei-Technica, Tokyo, Japan). Cardiac Vo_2 per min was calculated as the product of coronary flow and coronary arteriovenous O_2 content difference. Its LV component was calculated by subtracting the right ventricular Vo_2 from the cardiac Vo_2 [15, 16, 19]. To this end, we weighed the LV including the septum and the right ventricular (RV) free wall after the experiment. LV and RV weights were 81.7 ± 10.2 g and 24.7 ± 5.6 g respectively.

Experimental protocol

All experiments were carried out using isovolumic contractions. We used Emax (end-systolic maximal elastance) as a load-independent ventricular contractility index (Fig. 1A). We obtained Emax as the slope of the linear regression line fitted to the steady-state peak isovolumic P-V data points at 4 - 5 different LVVs at one randomly selected heart rate (HR) out of 60, 80, 120 and 180 beats/min and confirmed the linearity of the end-systolic P-V relation. We then set the LVV at V_0 where the end-systolic pressure was zero, to obtain steady-state unloaded contractions. We repeated the procedure to obtain Emax and steady-state unloaded contractions at the three other heart rates, except that we selected 2 or 3 of the 4 to 5 LVVs to obtain Emax without causing deterioration of the heart. The RV was always unloaded by draining the coronary venous return.

We then measured cardiac Vo_2 of steady-state unloaded contractions at V_0. This Vo_2 comprised the PVA-independent Vo_2 consisting of the E-C coupling Vo_2 and basal metabolic Vo_2 of the whole heart (i.e., the LV, RV and atria). We neglected the contribution of atrial Vo_2 to the cardiac Vo_2 simply because of the low weight of the atria. We assumed the PVA-independent Vo_2, E-C coupling Vo_2, and basal metabolic Vo_2 to be shared by the LV and RV in proportion to their weights. Therefore, LV PVA-independent Vo_2, E-C coupling Vo_2, and basal metabolic Vo_2 were obtained by dividing their cardiac Vo_2 values by LV/ (LV + RV) (Fig. 1B). We did not measure Vo_2 of loaded contractions, which was beyond the scope of the present study.

These Emax and Vo_2 measurements were repeated at each of the four heart rates. The heart was finally arrested to obtain basal metabolic Vo_2 by continuously infusing 0.17 M KCl solution at a rate of 1.0 - 1.5 ml/min after its 15 to 20 ml bolus infusion into the coronary artery [15]. E-C coupling Vo_2 was calculated by subtracting basal metabolic Vo_2 from the PVA-independent Vo_2.

The systemic arterial blood pressure of the support dog served as the coronary perfusion pressure of the excised donor heart. We successfully prevented the tendency for development of hypotension and maintained mean systemic arterial blood pressure of the support dog at approximately 90 mmHg by electrically stimulating the Neiguan (PC-6) acupoint in the bilateral forearms, as is usual in our laboratory [21, 22]. Briefly, we inserted one stainless steel needle vertically into the acupoint 3 cm above the transverse crease of each wrist between the tendons of the long palmar muscle and the radial flexor muscle. Stimulation parameters were 5 V-40 Hz biphasic pulses with 5 msec pulse width.

Statistics

Emax and Vo_2 values were normalized to 100 g LV. The data were expressed as means ± SD. A one-way analysis of variance (ANOVA), followed by the Bonferroni method, was used in the statistical analysis. We considered $p < 0.05$ to indicate statistical significance.

RESULTS

Fig. 2 shows that Emax remained unchanged despite increases in heart rate from 60 to 80, 120, and 180 beats/min. Panel A shows mean ± SD of raw values of Emax in all 9 hearts. Panel B shows mean ± SD of Emax normalized to the Emax at 60 beats/min in each of the 9 hearts. There was no significant change in Emax among the different heart rates.

Fig. 3 Panel A shows that the PVA-independent Vo_2 per min, namely, the Vo_2 for the unloaded contractions at Vo, significantly increased linearly with increasing heart rate with a correlation coefficient of 0.918 ($p < 0.001$). Average PVA-independent Vo_2 increased 2.3 ± 0.2 times from 1.70 ± 0.16 ml O_2/100 g LV per min

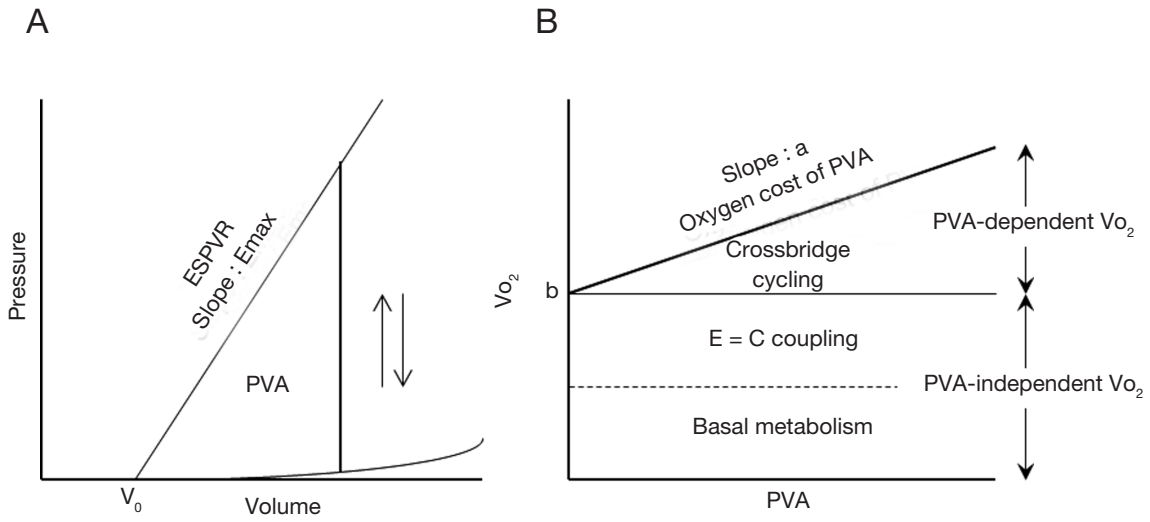

Fig. 1 Mechano-energetic framework of the left ventricle (LV) used in the present study. Panel A: Emax (left ventricular contractility index measured as the maximum or end-systolic pressure-volume ratio) and PVA (left ventricular total mechanical energy measured as systolic pressure-volume area) in the pressure-volume (P-V) diagram. Slope of the end-systolic P-V relation (ESPVR) line is defined as Emax. PVA is the area surrounded by the ESPVR line, the end-diastolic P-V relation curve, and the systolic P-V trajectory. PVA consists of the potential energy alone in an isovolumic contraction. V_0, volume at which LV peak isovolumic pressure is 0. Panel B: The Vo_2-PVA relation of volume-loaded contractions in baseline contractile state (solid diagonal line). The slope **a** of this relation indicates the oxygen cost of PVA. Vo_2 consists of 2 components: PVA-dependent and PVA-independent Vo_2 fractions. The former indicates crossbridge cycling and is zero in the unloaded contractions with zero PVA. The latter is equal to Vo_2 intercept **b**, which is the sum of Vo_2 for excitation-contraction (E-C) coupling and basal metabolism.

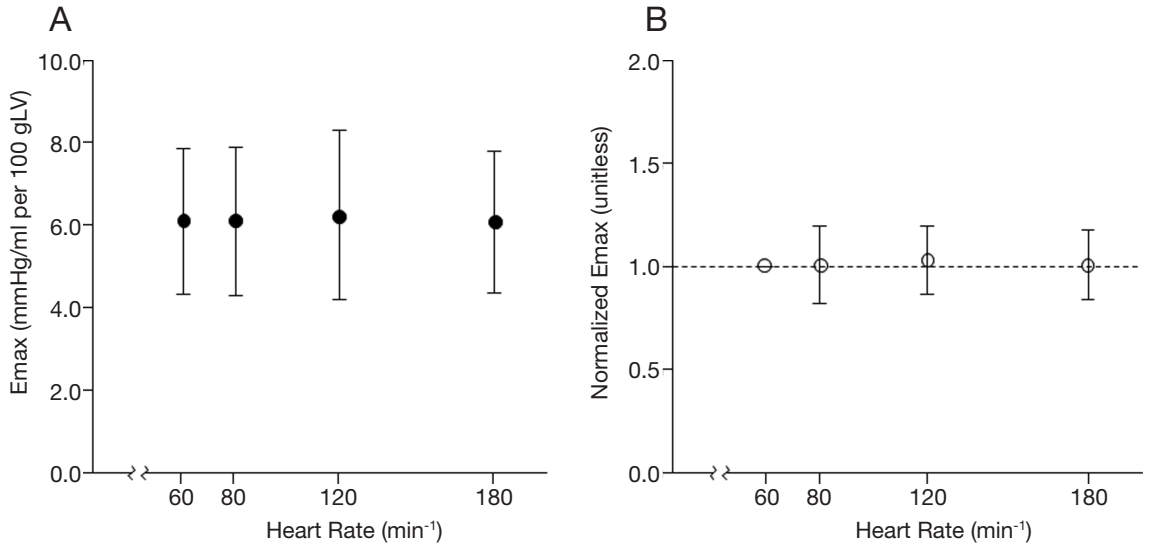

Fig. 2 Heart rate-independence of Emax. Panel A: The relation between Emax and heart rates of 9 hearts. There were no significant changes in Emax over the heart rate range of 60-180 beats/min. All values are mean ± SD. Panel B: Emax at HR of 80, 120 and 180 beats/min were normalized relative to that at HR of 60 beats/min in each heart.

at HR of 60 beats/min to 3.91 ± 0.35 ml O_2/100g LV per min at HR of 180 beats/min.

Basal metabolic Vo_2 measured under KCl arrest was 0.95 ± 0.29 ml O_2/100 g LV per min. We assumed that the same basal metabolic Vo_2 existed in the beating hearts as in our previous studies [15]. Panel A simply plots the same mean ± SD at the different heart rates.

Fig. 3 Panel B shows that the E-C coupling Vo_2 per min, namely, the PVA-independent Vo_2 per min minus the basal metabolic Vo_2 per min, significantly increased linearly with increasing heart rate with a correlation coefficient of 0.918 ($p < 0.001$). This correlation coefficient is the same as that for the PVA-independent Vo_2 in Panel A because the E-C coupling Vo_2 relationship in Panel B is merely a parallel downward shift of the PVA-independent Vo_2 relation in Panel A. The E-C coupling Vo_2 per min increased 4.2 ± 1.1 times from 0.754 ± 0.289 ml O_2/100 g/min at HR of 60 beats/min to 2.93 ± 0.54 ml O_2/100 g/min at HR of 180

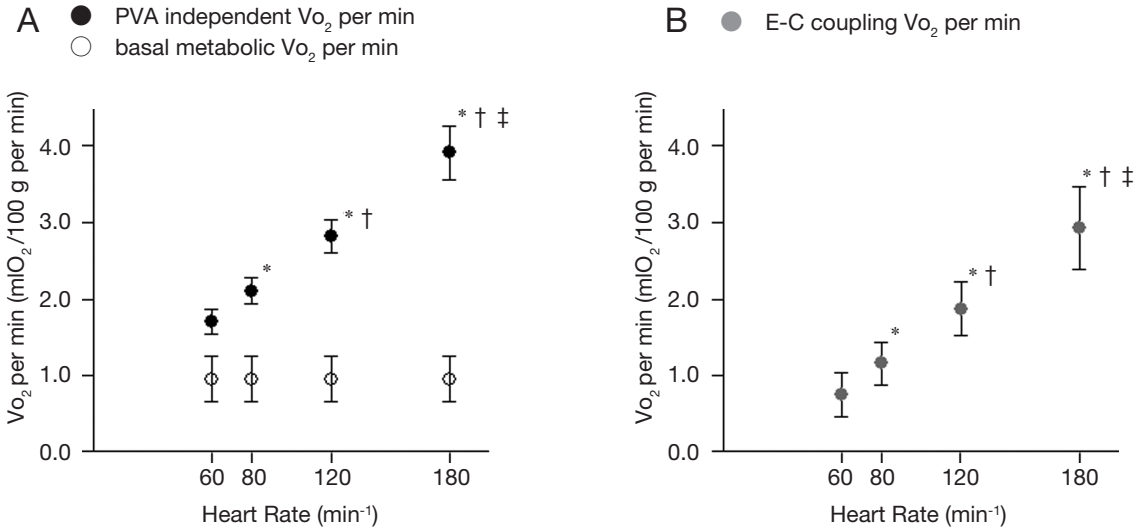

Fig. 3 PVA-independent Vo_2, basal metabolic Vo_2, and E-C coupling Vo_2, all per min, as a function of heart rate increases from 60 to 180 beats/min. Panel A: Graph showing the heart rate-dependence of PVA-independent Vo_2 per min (solid circle) in all hearts. The same basal metabolic Vo_2 per min (open circle) measured under KCl arrest is plotted for all heart rates. Panel B: Graph showing the heart rate-dependence of E-C coupling Vo_2 per min. All values are mean ± SD. Vo_2: oxygen consumption. E-C coupling: excitation-contraction coupling.
*: $P < 0.05$ vs. HR of 60 beats/min. † : $P < 0.05$ vs. HR of 80 beats/min. ‡ : $P < 0.05$ vs. HR of 120 beats/min.

beats/min.

Fig. 4 shows the same Vo_2 data shown in Fig. 3 after converting the per min values to per beat values. Panel A shows both the PVA-independent and basal metabolic Vo_2 per beat at the four heart rates. Both Vo_2 values per beat significantly decreased curvilinearly with increasing heart rate. The PVA-independent Vo_2 per beat decreased 0.76 ± 0.07 times from 0.0284 ± 0.0027 ml O_2/100 g/beat at HR of 60 beats/min to 0.0215 ± 0.0019 ml O_2/100 g/beat at HR of 180 beats/min. Since the basal metabolic Vo_2 per beat was obtained by dividing the KCl-arrest basal metabolic Vo_2 per min by heart rate, the basal metabolic Vo_2 per beat decreased 0.33 times, namely, by a HR ratio of 60/180 = 1/3 = 0.33, from 1.58 ± 0.49 ml O_2/100 g/beat at HR of 60 beats/min to 0.53 ± 0.16 ml O_2/100 g/beat at HR of 180 beats/min.

Fig. 4 Panel B shows that the E-C coupling Vo_2 per beat, namely, the PVA-independent Vo_2 per beat minus the basal metabolic Vo_2 per beat, still significantly increased curvilinearly with increasing heart rate with a correlation coefficient of 0.328 ($p = 0.05$). The E-C coupling Vo_2 per beat increased 1.4 ± 0.4 times from 1.26 ± 0.48 ml O_2/100 g/min at HR of 60 beats/min to 1.62 ± 0.27 ml O_2/100 g/min at HR of 180 beats/min. The E-C coupling Vo_2 per beat at any HR of 80, 120, and 160 beats was significantly greater than that at HR of 60 beats/min. However, the E-C coupling Vo_2 at either 120 or 180 beats/min was no more significantly greater than that at 80 beats/min.

Fig. 5 shows that the ratio of the E-C coupling Vo_2 per beat to the corresponding Emax, namely, the E-C coupling Vo_2 normalized to the Emax, still significantly increased curvilinearly with increasing heart rate. The ratio increased 1.4 ± 0.4 times from 0.00207 ± 0.00053 (ml O_2/100 g/beat)/ (mmHg/ml/100 g LV) at HR of 60 beats/min to 0.00281 ± 0.00062 (ml O_2/100 g/beat)/ (mmHg/ml/100 g LV) at HR of 180 beats/min. The normalized E-C coupling Vo_2 per beat at any HR of 80, 120, and 160 beats was significantly greater than that at HR of 60 beats/min. The normalized E-C coupling Vo_2 at 180 beats/min was still significantly greater than that at 80 beats/min. However, the former was no more significantly greater than that at 120 beats/min.

DISCUSSION

This is the first study to reveal the relationship between Emax, E-C coupling Vo_2, and heart rate over a reasonably wide physiological range (60−180 beats/min) in excised cross-circulated canine hearts. The present results have shown a significant increase in E-C coupling Vo_2 per beat with increasing heart rate despite absence of a significant increase in Emax (Figs. 2 and 4B). As the significance tests show (Figs. 4B and 5), the significant increase in E-C coupling Vo_2 per beat at HR of 180 beats/min could not be validated if the lowest HR chosen was 80 instead of 60 beats/min. Similarly, the significant increase in E-C coupling Vo_2 per beat normalized to Emax at HR of 180 beats/min could not be validated if the lowest HR chosen was 120 instead of 60 or 80 beats/min. Therefore, unless we chose the minimal HR of 60 beats/min, we could not have obtained significant HR-dependent increases in E-C coupling Vo_2 per beat as well as that normalized to Emax.

Although increasing the heart rate from 120 to 190 beats/min had not changed PVA-independent Vo_2 per beat [19], E-C coupling Vo_2 had not been investigated in that study. Other studies showed an increased Vo_2 under a fixed peak wall tension with heart rate increases from 120 to 180 beats/min [5] and a slight decrease in Vo_2 under a fixed peak isovolumic pressure with heart rate increases from 100 to 200 beats/min

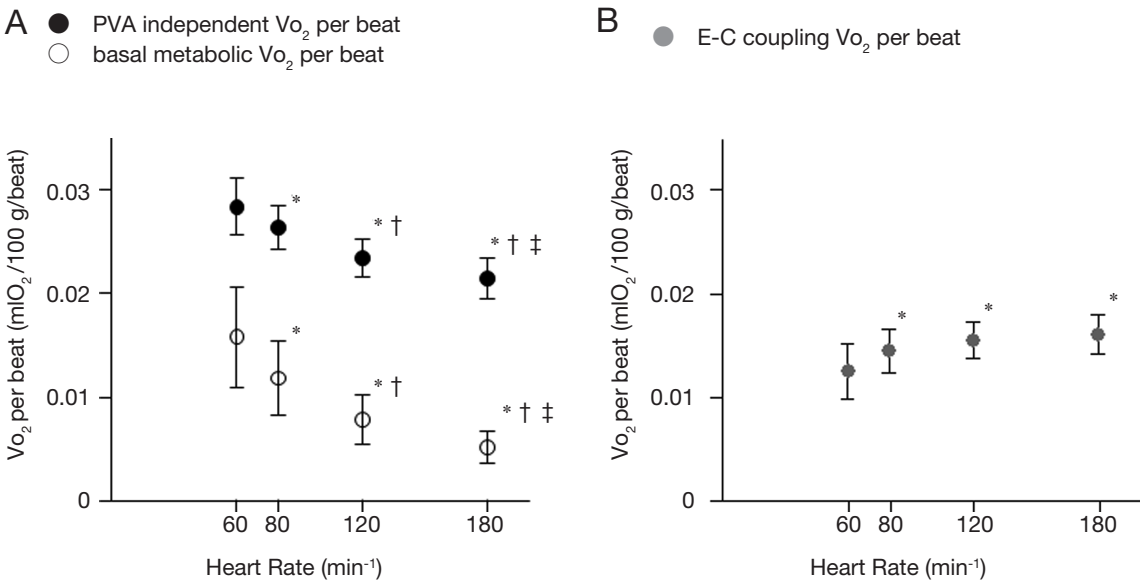

Fig. 4 PVA-independent Vo_2, basal metabolic Vo_2, and E-C coupling Vo_2, all per beat, as a function of heart rate increases from 60 to 180 beats/min. Panel A: Graph showing the heart rate-dependence of PVA-independent Vo_2 per beat (solid circle) and basal metabolic Vo_2 per beat (open circle) in all hearts. Basal metabolic Vo_2 per beat was obtained by dividing the same basal metabolic Vo_2 per min measured under KCl arrest by the respective heart rates. Panel B: Graph showing the heart rate-dependence of E-C coupling Vo_2 per beat. All values are mean ± SD. Vo_2: oxygen consumption. E-C coupling: excitation-contraction coupling.
*: $P < 0.05$ vs. HR of 60 beats/min. † : $P < 0.05$ vs. HR of 80 beats/min. ‡ : $P < 0.05$ vs. HR of 120 beats/min.

[1]. However, the lowest heart rates (100−120 beats/min) in both these studies [1, 5] were much higher than that (60 beats/min) in the present study. In these studies, no E-C coupling Vo_2 had been investigated.

A previous study showed that increasing the heart rate from 50 to 250 beats/min, a wider range than in the present study, did not change Emax, but decreased PVA-independent Vo_2 per beat hyperbolically [2]. This finding may appear consistent with our present results (Figs. 2 and 4A). However, these authors did not measure basal metabolic Vo_2 and hence did not obtain the E-C coupling Vo_2. From the unchanged Emax, they rather speculated that E-C coupling Vo_2 would be constant with decreasing PVA-independent Vo_2. In this respect, our present study is the first to clearly show that E-C coupling Vo_2 per beat increases with heart rate over a reasonably wide heart rate range, though slightly narrower than 50−250 beats/min [2].

From the significantly increased E-C coupling Vo_2 per beat with increasing heart rate (Fig. 4B), one could simply expect an increased intra-myocardial total Ca^{2+} handling. However, the unchanged Emax would negate this possibility. The alternative possibility would be increased energy wastage during total Ca^{2+} handling in E-C coupling, with increasing heart rate.

To discuss this aspect further, consideration of the intra-myocardial Ca^{2+} recirculation fraction (RF) is helpful [17]. RF refers to the fraction of total Ca^{2+} handled during E-C coupling that recirculates intra-myocardially via the SERCA2, SR, and the ryanodine-sensitive Ca^{2+} release channel without being extruded across the sarcolemma via the NCX [8, 13, 17]. We have already shown that the RF increases with increasing heart rate using the same experimental system employed in the present study [4]. Increasing RF with heart rate could be accounted for by shortening of the action potential, greater shortening of the electrical diastolic phase with increasing heart rate and the resultant accumulation of intracellular Ca^{2+} [4, 9, 11, 14, 23].

However, this mechanism alone would accumulate intra-myocardial Ca^{2+}, augment total Ca^{2+} handling, and then increase Ca^{2+} release at a higher heart rate. This in turn would contradict the constant value for Emax obtained despite increasing heart rate (Fig. 2). An additional possible mechanism would involve time-dependent SERCA2 inactivation associated with the inactivating process mediated by the Ca^{2+}/calmodulin-dependent protein kinase-II (CaMKII) [10]. Thus, SERCA2 is activated quickly on excitation and then slowly inactivated [10]. This leads to an incomplete inactivation of SERCA2 within a short beat interval and then in the next beat a rapid re-sequestration of part of the Ca^{2+} released from the SR before it can bind to the troponin C for crossbridge cycling (Fig. 6 Panel A). In contrast, a complete inactivation of SERCA2 at the end of a long beat interval allows all the released Ca^{2+} to contribute to the contraction (Fig. 6 Panel B). This mechanism would appear to account for the unchanged Emax despite increasing heart rate.

There are several limitations to this study. Firstly, we obtained the value for E-C coupling Vo_2 by subtracting basal metabolic Vo_2 measured under KCl arrest from PVA-independent or unloaded Vo_2. Basal metabolic Vo_2 cannot be directly measured within the respective PVA-independent Vo_2 in a beating heart. No method is yet available to measure both basal metabolic Vo_2 and E-C coupling Vo_2 simultaneously in a beating

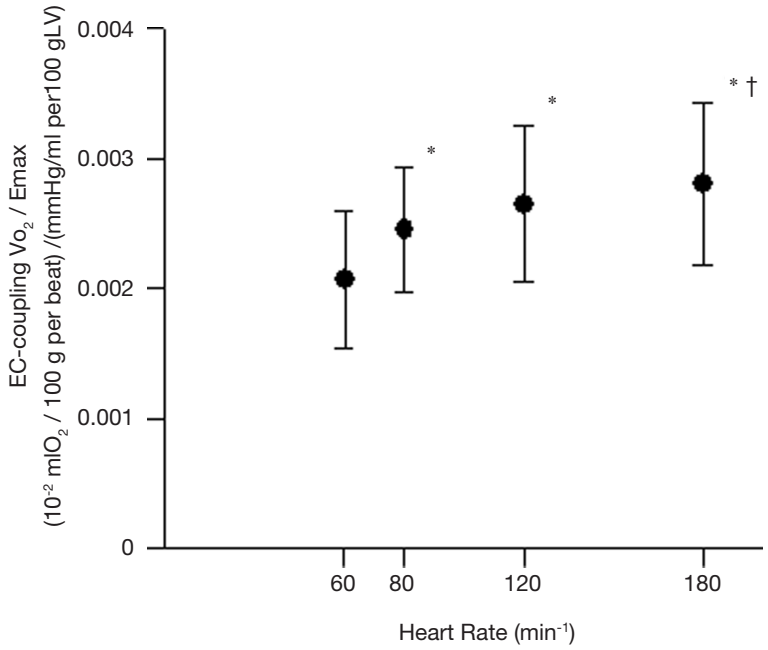

Fig. 5 Graph showing the heart rate-dependence of E-C coupling Vo_2 per beat normalized to Emax in all hearts. All values are mean ± SD. Vo_2: oxygen consumption. E-C coupling: excitation-contraction coupling. Emax: end-systolic maximal elastance.
*: $P < 0.05$ vs. HR of 60 beats/min. † : $P < 0.05$ vs. HR of 80 beats/min

heart. Other interventions such as 2, 3-butanedione monoxime can be used to eliminate crossbridge cycling for direct measurement of PVA-independent Vo_2 [24–26]. Ca^{2+} channel blockers and SERCA2 blockade with cyclopiazonic acid may be used to eliminate intramyocardial Ca^{2+} handling for direct measurement of basal metabolic Vo_2 [24–26]. However, these interventions also weaken the heart and hemodynamics of the support dog for the cross-circulation so adversely as to disrupt the experiment itself. Therefore, our present method is the only one available in the present type of heart preparation.

A second limitation is our assumption that residual crossbridge cycling in the unloaded contraction with zero PVA does not contribute to PVA-independent and hence E-C coupling Vo_2. Although this residual crossbridge cycling appears to contribute considerably to the unloaded Vo_2 in rabbit hearts [27], we have obtained evidence supporting our contention that the residual crossbridge cycling energy, if any, is negligibly small in E-C coupling Vo_2 of canine blood-perfused hearts [27, 28].

A third limitation is that we neglected a role for other trans-sarcolemmal Ca^{2+} transporters such as the T-type Ca^{2+} channel and the reverse mode NCX in accounting for part of the E-C coupling Vo_2 since they also consume some ATP directly or indirectly. These processes have been identified by the reductionist approach in isolated, but not in situ, mammalian cardiomyocytes [29–31]. For example, involvement of the reverse mode NCX in Ca^{2+} handling is known in rabbit hearts and possibly in failing, but not normal canine heart [32, 33]. In addition, even in isolated cardiomyocytes, the net Ca^{2+} current carried by these processes compared to the L-type Ca^{2+} channel, the forward mode NCX and SERCA2 is quite small [8, 34]. Thus, our consideration of the L-type Ca^{2+} channels, the forward mode NCX, and SERCA2 seems to be reasonable enough in the context of the present study.

Clinical implications

Beneficial effects of beta-blockers on morbidity and mortality in patients with chronic heart failure (CHF) are well established [35, 36]. Although numerous possible mechanisms for beta-blocker therapy exist (upregulation of β receptors, improvement of β adrenergic signaling, protection from catecholamine myocyte toxicity, inhibition of renin-angiotensin-aldosterone system), β-blocker-mediated heart rate reduction is considered to be one of central mechanism, relating to the increament of coronary blood flow and the decreased myocardial oxygen demand [35, 36]. The present study showed that lower heart rates are energetically advantageous in Emax- PVA frame work. The present results may be a reasonable explanation for the efficacy of beta-blocker therapy in patients with CHF.

In conclusion, we were for the first time able to evaluate successfully the effect of heart rates of 60–180 beats/min on E-C coupling Vo_2 per beat. We found that E-C coupling Vo_2 per beat as well as that normalized to Emax increased slightly but significantly with increasing heart rate. This result seems to be attributable to the increased contribution of SERCA2 accompanied by its incomplete inactivation during the beat interval which decreases with increasing heart rate. This finding indicates that higher heart rates are energetically disadvantageous because of wasting of the excitation-contraction coupling energy as heart rates rise.

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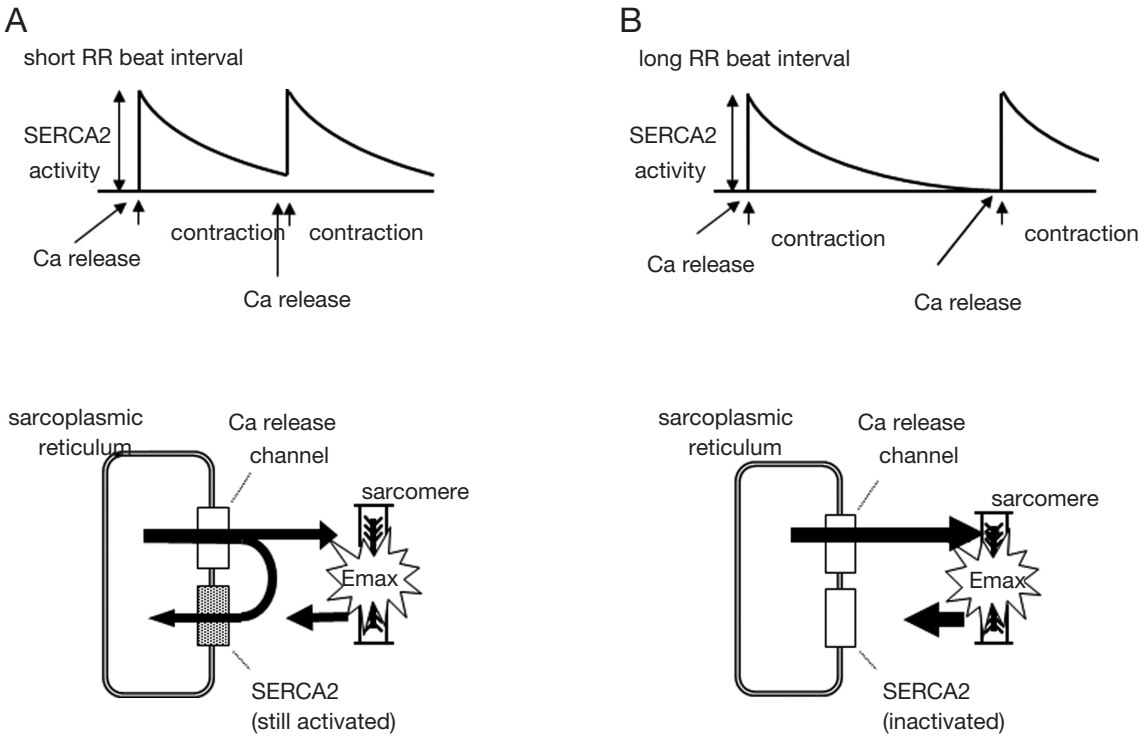

Fig. 6 Schematic diagram of the differences in Ca^{2+} handling between short and long RR beat intervals. Panel A: The shorter RR beat interval (higher HR) allows less complete inactivation of SERCA2. Therefore, the larger amount of Ca^{2+} is handled without contributing to the contraction. Such a state may require larger Ca^{2+} handling per unit of Emax. Panel B: The longer RR beat interval (lower HR) allows more complete inactivation of SERCA2. Thus, all of the released Ca^{2+} can contribute to the contraction. Such a state can reduce the total Ca^{2+} handling normalized to Emax. RR: beat interval between two adjacent R waves of ECG. HR: heart rate. Emax: end systolic maximal elastance. SERCA2: sarcoplasmic reticulum Ca^{2+}-ATPase.

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