Gender influences coronary L-type Ca\(^{2+}\) current and adaptation to exercise training in miniature swine

D. K. BOWLES
Department of Veterinary Biomedical Sciences, College of Veterinary Medicine and Dalton Cardiovascular Research Center, University of Missouri, Columbia, Missouri 65211

Received 1 June 2001; accepted in final form 14 August 2001

Bowles, D. K. Gender influences coronary L-type Ca\(^{2+}\) current and adaptation to exercise training in miniature swine. J Appl Physiol 91: 2503–2510, 2001.—Endurance exercise training increases smooth muscle L-type Ca\(^{2+}\) current density in both resistance and proximal coronary arteries of female miniature swine. The purpose of the present study was to determine 1) whether gender differences exist in coronary smooth muscle (CSM) L-type Ca\(^{2+}\) current density and 2) whether endurance training in males would demonstrate a similar adaptive response as females. Proximal, conduit (~1.0 mm), and resistance (~200 μm (internal diameter)) coronary arteries were obtained from sedentary and treadmill-trained swine of both sexes. CSM were isolated by enzymatic digestion (collagenase plus elastase), and voltage-gated Ca\(^{2+}\)-channel current (I\(_{\text{Ca}}\)) was determined by using whole cell voltage clamp during superfusion with 75 mM tetraethylammonium chloride and 10 mM BaCl\(_2\). Current-voltage relationships were obtained at test potentials from −60 to 70 mV from a holding potential of −80 mV, and I\(_{\text{Ca}}\) was normalized to cell capacitance (pA/pF). Endurance treadmill training resulted in similar increases in heart weight-to-body weight ratio, endurance time, and skeletal muscle citrate synthase activity in male and female swine. I\(_{\text{Ca}}\) density was significantly greater in males compared with females in both conduit (−7.57 ± 0.58 vs. −4.14 ± 0.47 pA/pF) and resistance arteries (−11.25 ± 0.74 vs. −6.49 ± 0.87 pA/pF, respectively). In addition, voltage-dependent activation of I\(_{\text{Ca}}\) in resistance arteries was shifted to more negative membrane potentials in males. Exercise training significantly increased I\(_{\text{Ca}}\) density in both conduit and resistance arteries in females (−7.01 ± 0.47 and −9.73 ± 1.13 pA/pF, respectively) but had no effect in males (−8.61 ± 0.50 and −12.04 ± 1.07 pA/pF, respectively). Thus gender plays a significant role in determining both the magnitude and voltage dependence of I\(_{\text{Ca}}\) in CSM and the adaptive response of I\(_{\text{Ca}}\) to endurance training.

electrophysiology; vascular smooth muscle; microcirculation; voltage-gated calcium channels

GENDER EXERTS SIGNIFICANT INFLUENCE on coronary vascular physiology and pathophysiology. For example, whereas coronary heart disease (CHD) is a leading cause of mortality in both men and women, men show a greater prevalence of CHD compared with premenopausal women (1). Similarly, the incidence of hypertension is greater in men and postmenopausal women compared with premenopausal women (20). Mechanisms underlying gender differences in the development of complex cardiovascular disease are likely multifaceted. However, gender differences in several candidate mechanisms impacting cardiovascular disease have been reported, including endothelial nitric oxide synthase (NOS) content, NO production (25), smooth muscle proliferation (11), and smooth muscle responsiveness to agonists (30), e.g., endothelin (29). Recently, functional studies have provided indirect evidence that gender differences in dihydropyridine-sensitive, voltage-gated Ca\(^{2+}\) channels (VGCC) may also contribute to differences in vascular function between males and females (12). Crews et al. (12) found that depolarization-induced contraction and Ca\(^{2+}\) influx is greater in thoracic aorta of male rats compared with female rats, suggesting a gender difference in plasmalemmal VGCC activity. VGCC activity plays a central role in regulation of vascular resistance and, therefore, total blood flow and distribution (32). In addition, VGCCs have been proposed to contribute to the incidence and severity of both acute and long-term cardiovascular events, such as vasospasm and CHD (18, 28). If present, gender differences in VGCC activity in coronary smooth muscle (CSM) could play a significant role in determining gender-related differences in vascular function in both health and disease.

Endurance exercise training also influences ion-channel activity and Ca\(^{2+}\) regulation in CSM (6, 8, 9, 14, 17). In female miniature swine, L-type Ca\(^{2+}\)-channel current (I\(_{\text{Ca}}\)) was increased in both conduit and resistance arterial smooth muscle after treadmill training (8, 14). Given the potential influence of gender on vascular smooth muscle Ca\(^{2+}\) regulation, the purpose of the present study was to determine 1) whether gender differences exist in CSM L-type Ca\(^{2+}\)-current density and 2) whether males demonstrate a similar adaptive response to exercise training as females.

http://www.jap.org 8750-7587/01 $5.00 Copyright © 2001 the American Physiological Society

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.
MATERIALS AND METHODS

Animals. Sexually mature, adult male and female miniature swine weighing 25–40 kg were obtained from the breeder (Charles River) and housed in pens at the College of Veterinary Medicine. All pigs included in this study were familiarized with treadmill exercise over a 1- to 2-wk period. Treadmill performance tests were administered to each animal. Pigs of each sex were then randomly divided into two groups. One group (Ex; n = 16, 6 females, 10 males) underwent a progressive treadmill training program used previously in our laboratory (7, 8, 14). The second group of pigs was restricted to their pens (6 × 12 ft) for 16–20 wk and served as sedentary controls (Sed; n = 15, 7 females, 8 males). Animal protocols were approved by the University of Missouri Animal Care and Use Committee in accordance with the “Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research, and Training.”

Training procedures and treadmill performance tests. Ex pigs underwent a 16- to 20-wk treadmill endurance program followed by treadmill performance tests as described previously (8, 9, 14, 24). During the final 4–8 wk of training, a typical training session consisted of the following 85-min workout: 1) 5-min warm-up run at 2.5 miles/h (mph), 2) 15-min sprint at speeds of 5–8 mph, 3) 60-min endurance run at 4–5 mph, and 4) 5-min cool-down run at 2 mph. Treadmill performance tests were administered before and at the completion of the training (Ex) or pen confinement (Sed).

Skeletal muscle oxidative enzyme activity. At the time of death, muscle samples were taken from the medial and lateral heads of the triceps brachii and deltoid, frozen in liquid nitrogen, and stored until processed. Citrate synthase activity was measured spectrophotometrically from whole muscle homogenates (34).

Preparation of coronary arteries. Pigs were anesthetized with ketamine (35 mg/kg im), Rompun (2.25 mg/kg im), and pentothal sodium (10 mg/kg iv), followed by administration of heparin (1,000 U/kg iv). Hearts were removed and placed in iced (4°C) Krebs bicarbonate solution during vessel isolation. Main right conduit arteries (1.0-mm ID) were dissected free from the heart beginning ~2 cm distal to the ostia and proceeding to the origin of the posterior descending artery and cleaned of connective and adipose tissue at 4°C in physiological saline solution (PSS) containing (in mM) 2 CaCl₂, 138 NaCl, 1 MgCl₂, 5 KCl, 10 HEPES, and 10 glucose, pH 7.4. Similarly, epicardial resistance vessels 175–225 μm ID were obtained from the apex region of the anterior left ventricular free wall at 4°C in PSS.

Cell dispersion. All experiments were performed on freshly dispersed cells by using methods modified from those described previously (35–37, 39). Arteries were incubated in dispersion equation, 1/{1 exp[(V0.5 – V)/k]}).

RESULTS

Efficacy of exercise training. Male swine tended to have greater body and heart weights compared with females; however, heart weight-to-body weight ratio, treadmill endurance time, and skeletal muscle citrate synthase activity levels were similar in sedentary male and female pigs (Table 1). Consistent with previous reports, the treadmill-trained miniature swine model (24), Ex animals of both sexes demonstrated overall similar adaptations to endurance training, including an increase in citrate synthase activity in the medial and lateral heads of the triceps brachii, in-
creased treadmill endurance time, and increased heart weight-to-body weight ratio (Table 1).

Gender and cell size. In both conduit and resistance arteries, CSM cells from male animals were significantly larger than cells from female animals, as indicated by a greater cell membrane surface area (i.e., cell capacitance, pF; Table 2). Cell size was unaffected by exercise training with the exception of a smaller mean cell size in resistance arterioles of Ex compared with Sed males. Whether the smaller cell size in resistance arterioles of Ex males is an effect of training or due to unintentional bias is unclear; however, the latter is unlikely as the experimenter was blinded to the treatment of the animal. Furthermore, a similar trend was noted for Ex females in this and a previous study (8). Therefore, it is likely that this is a true response to exercise training and may indicate vascular remodeling. Due to variations among groups in cell membrane surface area, all ion currents were normalized to cell capacitance and presented as current density (pA/pF).

Series resistance during voltage clamp was minimal and similar in all groups (data not shown). Due to variations among groups in cell membrane surface area, all ion currents were normalized to cell

Table 1. Efficacy of training in male and female pigs

<table>
<thead>
<tr>
<th>BW, kg</th>
<th>HW, g</th>
<th>HW/BW, y/kg</th>
<th>Endurance, min</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Female</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Sed</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>33 ± 2</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>156 ± 7</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>4.8 ± 0.1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>25 ± 1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>15 ± 1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>13 ± 1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>18 ± 2</td>
</tr>
</tbody>
</table>

Values are means ± SE for sedentary (Sed) female (n = 7), Sed male (n = 8), exercise-trained (Ex) female (n = 6), and Ex male (n = 10) pigs. BW, body weight; HW, heart weight; HW/BW, heart weight-to-body weight ratio; endurance, endurance time on treadmill test; medial, medial head of triceps; lateral, lateral head of triceps. *P < 0.05 for Ex vs. Sed. †P < 0.05 for male vs. female.

Table 2. Effect of gender and training on coronary smooth muscle cell size

<table>
<thead>
<tr>
<th></th>
<th>Female</th>
<th>Male</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Sed</td>
<td>Ex</td>
</tr>
<tr>
<td>Conduit</td>
<td>19.8 ± 0.9</td>
<td>21.1 ± 0.9</td>
</tr>
<tr>
<td>Resistance</td>
<td>15.1 ± 1.1</td>
<td>12.1 ± 1.2</td>
</tr>
</tbody>
</table>

Values are means ± SE in pF; n values are in parentheses. *P < 0.05 vs. Sed. †P < 0.05 vs. females.

Fig. 1. Voltage-dependent Ca²⁺ current (I_{Ca}) in coronary smooth muscle (CSM). Representative current tracings from conduit (A) and resistance (B) arteries of a sedentary male swine. Depolarization steps produced characteristic voltage- and time-dependent inward currents with slow inactivation. Only traces from sedentary male swine CSM, presented as currents, were similar in Ex males. Representative current tracings depicting effect of nifedipine (nif; 2 μM) on inward current (20-mV test potential) in conduit (C) and resistance (D) CSM. Nifedipine essentially abolished peak and sustained inward currents (>90% and ~100%, respectively) and was equally effective in sedentary (Sed) and exercise-trained (Ex) male swine. Similar findings in Sed and Ex female swine have been reported (8), indicating that L-type voltage-gated Ca²⁺ channels (VGCCs) contribute predominantly to whole-cell current in CSM from conduit and resistance arteries and that this is unaffected by gender or training. Scale bars indicate 100 ms (horizontal) and 5 pA/pF (vertical).
channels require greater depolarization for activation and inactivate slowly (5). As shown previously for female porcine CSM (7, 8), inward currents in CSM in the present study were completely inhibited by nifedipine (Fig. 1, C and D) and showed characteristic L-type voltage dependence of activation and slow inactivation (3, 32, 38). Together, these data indicate that L-type Ca\textsuperscript{2+} channels are the predominant channel type in CSM from both male and female porcine coronary conduit and resistance arteries. Furthermore, in agreement with data shown previously for females (8), endurance training in males does not alter this L-type Ca\textsuperscript{2+}-current predominance.

**Gender, exercise training, and I\textsubscript{Ca} I-V relationship.**

The effect of gender and exercise training on Ca\textsuperscript{2+} I-V relationships for conduit and resistance arteries are shown in Fig. 2. Exercise training, gender, and arterial size all exert significant effects on the I-V relationship. With regard to gender, I\textsubscript{Ca} density was greater in males compared with females in CSM from both conduit and resistance arteries. As described previously (7), we observed a greater I\textsubscript{Ca} density in CSM of resistance arteries compared with conduit arteries, confirming the significant effect of vessel size on CSM I\textsubscript{Ca} density. Exercise training significantly increased I\textsubscript{Ca} density in both conduit and resistance arteries of females; however, this effect was gender specific, as exercise training had no effect on I\textsubscript{Ca} in either artery from male swine. Figure 3 shows the maximum peak I\textsubscript{Ca} obtained, irrespective of membrane potential. Similar to effects on the I-V relationship, peak I\textsubscript{Ca} density in males was greater than in females in both conduit and resistance coronary arteries. Exercise training significantly increased peak I\textsubscript{Ca} density in both conduit and resistance arteries from females, with no effect in males. In addition, peak I\textsubscript{Ca} density was greater in resistance vessels compared with conduit arteries in both sedentary and exercise trained groups of both sexes.

**Voltage dependence of I\textsubscript{Ca}.** VGCCs switch between conducting (open) and nonconducting (closed) states.
primarily in response to changes in membrane potential (32). Whereas the open probability ($P_o$) of VGCCs is increased by membrane depolarization, steady-state depolarization also results in channel inactivation. The relative number of open vs. inactivated channels determines steady-state Ca$^{2+}$ conductance at a given membrane potential. Thus changes in the voltage dependence of channel activation or inactivation can significantly impact the steady-state Ca$^{2+}$ influx in the cell. Figure 4 depicts the effect of gender, exercise, and arterial size on voltage-dependent activation of $I_{Ca}$ in CSM. Exercise training had no effect on voltage-dependent activation of $I_{Ca}$ in arteries of either female or male swine. However, both gender and arterial size influenced voltage-dependent activation. Half-maximal activation voltages ($V_{0.5}$) derived from the Boltzmann equation fits to activation curves are shown in Table 3. In addition to a higher $I_{Ca}$ density in males, voltage-dependent activation was shifted approximately 2–3 mV negative compared with females. Steady-state voltage-dependent inactivation is shown in Fig. 5, with corresponding $V_{0.5}$ values provided in Table 3. Exercise training had no effect on voltage-dependent inactivation. However, gender influenced channel inactivation, as indicated by an approximate 5-mV negative shift in $V_{0.5}$ for both resistance and conduit arteries in males compared with females. Neither exercise training, gender, nor arterial size had any effect on the slope of either voltage-dependent activation or inactivation (data not shown).

**DISCUSSION**

The present study provides the first direct evidence that gender plays a significant role in determining both basal $I_{Ca}$ in CSM and the adaptive response of this current to endurance exercise training. Voltage-clamp data demonstrate that CSM from male swine exhibits both an increased $I_{Ca}$ density and a negative shift in voltage-dependent activation and inactivation compared with female swine. A second novel and important finding of the present study is the influence of gender on the adaptive response of $I_{Ca}$ to exercise training. Exercise training increased current density in CSM from both conduit and resistance arteries in a gender-specific manner [i.e., an increase in females as previously demonstrated (8, 14) with no effect in males]. This gender influence on $I_{Ca}$ density may play an important role in determining previously reported gender-related differences in vascular reactivity and, perhaps, the incidence and severity of cardiovascular disease.

Table 3. Effect of training, gender, and vessel size on voltage for half-maximal current activation and inactivation

<table>
<thead>
<tr>
<th></th>
<th>Female</th>
<th>Male</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Sed</td>
<td>Ex</td>
</tr>
<tr>
<td><strong>Conduit</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Voltage-dependent activation</td>
<td>$9.0 \pm 0.5$</td>
<td>$8.2 \pm 0.8$</td>
</tr>
<tr>
<td>(31)</td>
<td>(31)</td>
<td>(35)</td>
</tr>
<tr>
<td>Resistance</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Voltage-dependent activation</td>
<td>$10.8 \pm 0.5$</td>
<td>$11.0 \pm 0.3$</td>
</tr>
<tr>
<td>(22)</td>
<td>(19)</td>
<td>(26)</td>
</tr>
<tr>
<td><strong>Conduit</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Voltage-dependent inactivation</td>
<td>$-9.3 \pm 0.8$</td>
<td>$-7.9 \pm 0.4$</td>
</tr>
<tr>
<td>(16)</td>
<td>(19)</td>
<td>(35)</td>
</tr>
<tr>
<td>Resistance</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Voltage-dependent inactivation</td>
<td>$-8.6 \pm 0.5$</td>
<td>$-8.4 \pm 0.7$</td>
</tr>
<tr>
<td>(19)</td>
<td>(18)</td>
<td>(25)</td>
</tr>
</tbody>
</table>

Values are means $\pm SE$; n values are in parentheses. $^\dagger P < 0.05$ vs. female.
Gender and ICa. On the basis of existing epidemiological and experimental evidence, the initial hypothesis was that CSM from males would exhibit increases in ICa density and/or a negative shift in voltage-dependent activation compared with females. This original hypothesis was based on epidemiological studies that demonstrated a greater incidence of hypertension and CHD in men and postmenopausal women compared with premenopausal women (1, 20) and the proposed role of Ca2+ influx via VGCCs in the etiology of both diseases (18). Furthermore, in vitro studies have demonstrated increased contractile responses and 45Ca2+ influx in aortic smooth muscle from males compared with females (12), providing indirect evidence for an increased VGCC activity in aortic smooth muscle of males compared with females. The present findings in porcine coronary artery provide the first direct evidence to support this hypothesis. Smooth muscle ICa from both conduit and resistance coronary arteries was significantly greater in Sed male swine compared with Sed female swine. In addition, voltage-dependent activation and inactivation of ICa in CSM from male swine demonstrated a negative shift compared with female swine. This can be interpreted as a negative shift in the “window current” of ICa (i.e., the range of membrane potential over which Ca2+ channels are active) in males compared with females. Knot and Nelson (21) have demonstrated a dihydropyridine-sensitive, steep relationship between membrane potential, intracellular Ca2+, and diameter in cerebral microvessels, indicating that small changes in membrane potential can have significant effects on arteriolar diameter. Thus, although small, negative shifts in voltage-dependent activation in males may have a substantial impact on Ca2+ influx and vasomotor tone. The Boltzmann distribution fits derived in this study predict that a 3-mV negative shift in V0.5 would increase relative VGCC conductance (i.e., P_o) at a given membrane potential by ~50%. All other factors being equal (e.g., Ca2+ buffering, extrusion), both the increased ICa density and negative shift in voltage-dependent activation in arteries of males would increase Ca2+ influx via VGCCs in response to vasoactive agonists, which directly or indirectly activate VGCCs, resulting in an increased contractile response. In addition, long-term increases in Ca2+ influx in CSM of males may contribute to increases in the incidence and severity of cardiovascular disease such as atherosclerosis and/or hypertension (18, 28).

Gender and exercise training. Previously, exercise training has been shown to increase L-type ICa in conduit, small artery, and arteriolar CSM of female miniature swine (8, 14). A primary purpose of the present study was to determine whether similar training adaptations occur in males. In contrast to females, exercise training did not affect ICa in conduit or resistance arteries from males, indicating that the adaptation of CSM ICa to endurance training is gender specific. As noted previously (8), the increased ICa density in females appears paradoxical within the context of VGCC-dependent Ca2+ influx as a mediator of increased vasomotor activity and cardiovascular disease (as discussed above for gender differences in sedentary individuals). This “ICa paradox” is perhaps resolved by the finding of Heaps et al. (14) that increased L-type Ca2+ influx in CSM of Ex female swine is compensated such that cytosolic Ca2+ responses are unchanged. Thus the increased ICa density that occurs with training may be a single component in a coordinated adaptation in CSM Ca2+ regulation involving sarcoplasmic reticulum Ca2+ unloading, coupled K+ -channel activation, and depressed contractile responses to vasoactive agonists (6, 9, 10, 23). Similar potential adaptations in cellular Ca2+ regulation following exercise training.

![Fig. 5. Effect of gender, exercise and, arterial size on voltage-dependent inactivation of ICa. Voltage-dependent inactivation of L-type current in CSM from conduit (A and B) and resistance (C and D) arteries from female (A and C) and male (B and D) Sed (◆) and Ex (●) swine. Exercise training had no effect on activation curves in either gender or artery size. In both arterial sizes, half-maximal inactivation potentials were shifted to more negative membrane potentials in males vs. females (Table 3). Relative peak current (I/I_max) data were fit to a conventional Boltzmann distribution equation. Peak current was determined during step depolarization to 20 mV following 4-s pre-pulses at potentials (V_m) indicated. Data are means ± SE.](http://jap.physiology.org/Content/HttpBase/figs/fig5.png)
may occur in males to reduce sensitivity of coronary arteries to vasoactive agents separate from changes in $I_{Ca}$ (17). In pathological conditions, such as hypertension, increases in $I_{Ca}$ may develop separately from the associated compensating mechanisms that occur with exercise training, resulting in increased cytosolic Ca$^{2+}$ and contractile responses (12, 30) and, in the long term, contribute to increased vascular disease.

Potential mechanisms. As whole cell $I_{Ca}$ is the product of the number and $P_o$ of active channels, differences in $I_{Ca}$ density due to gender, training, and arterial size must result from differences in synthesis and membrane targeting of functional channels and/or modulation of channel $P_o$. It is reasonable to speculate, especially for gender differences, that hormones may be responsible for $I_{Ca}$ differences. Testosterone, estrogen, glucocorticoids, catecholamines, and aldosterone have been shown to influence L-type Ca$^{2+}$-channel synthesis and activity in cultured vascular smooth muscle and other cell types (4, 13, 33). Recent sequencing of the Ca.1.2 gene promoter region has provided evidence for a hormone response element strongly activated by testosterone (26). Conversely, estrogen inhibits L-type Ca$^{2+}$ channels (31), whereas L-type Ca$^{2+}$ channel expression in cardiac muscle is increased in estrogen-receptor knockout mice (16). If estrogen acts in vivo to decrease $I_{Ca}$, this would provide a logical candidate mechanism for the lower $I_{Ca}$ in females compared with males. However, it is unlikely that training-induced reductions in circulating estrogens is a mechanism for increased $I_{Ca}$ in exercise-trained female swine, as this training model does not alter estrus cycle length or estrogen or progesterone levels (24). Another consideration regarding this model is that male swine have higher circulating levels of 17β-estradiol than females (2, 24). Whether the effect of higher estrogen levels in males is overridden by a dominant effect of testosterone, as has been proposed for sex hormone effects on endothelin receptors in porcine CSM (2), is unknown. Clarification of the role of either androgens or estrogens in determining gender-related difference in $I_{Ca}$ will require additional studies (e.g., hormone replacement in gonadectomized animals of both sexes).

Apart from increased channel synthesis, changes in channel $P_o$ may account, in part or whole, for the $I_{Ca}$ differences observed. As phosphorylation plays a dominant role in regulating $P_o$ of VGCCs, kinase/phosphatase regulation may be central to the observed gender and exercise-training differences in $I_{Ca}$. Accordingly, an intriguing candidate is protein kinase C (PKC). Recently, Kanashiro and Khalil (19) reported increased PKC-α, -δ, and -γ levels in aorta of male, compared with female, rats and PKC activation increased L-type $I_{Ca}$ in porcine CSM (15). Furthermore, exercise training has been reported to increase PKC-α levels in porcine coronary arterioles (22). Whether PKC-dependent increases in VGCC activity contribute to gender and training-induced differences in coronary $I_{Ca}$ remains to be determined.

In conclusion, the present study provides the first direct evidence that gender significantly influences CSM L-type Ca$^{2+}$-channel activity and the subsequent adaptive response to endurance exercise training. Future studies directed at understanding the underlying mechanisms and functional consequences of these differences may aid in determining the basis for gender-related differences in cardiovascular disease and the cardioprotective effect of endurance training.

The authors thank Cathy Galle for invaluable technical assistance in this project and Dr. Chris Heaps and Joyce Warwick for critical reading. This work was supported by National Heart, Lung, and Blood Institute Grant HL-52490.

REFERENCES


21. Knot HJ and Nelson MT. Regulation of arterial diameter and wall 


25. Leblanc N, Wan X, and Leung PM. Physiological role of 


