Temperature-dependent expression of sarcolemmal K⁺ currents in rainbow trout atrial and ventricular myocytes

MATTI VORNANEN, ARI RYÖKKYNEN, AND ANTI NURMI
University of Joensuu, Department of Biology, 80101 Joensuu, Finland
Received 18 June 2001; accepted in final form 3 January 2002

Vornanen, Matti, Ari Ryykkynen, and Antti Nurmi. Temperature-dependent expression of sarcolemmal K⁺ currents in rainbow trout atrial and ventricular myocytes. Am J Physiol Regulatory Integrative Comp Physiol 282: R1191–R1199, 2002; 10.1152/ajpregu.00349.2001.—Temperature has a strong influence on the excitability and the contractility of the ectothermic heart that can be alleviated in some species by temperature acclimation. The molecular mechanisms involved in the temperature-induced improvement of cardiac contractility and excitability are, however, still poorly known. The present study examines the role of sarcolemmal K⁺ currents from rainbow trout (Oncorhynchus mykiss) cardiac myocytes after thermal acclimation. The two major K⁺ conductances of the rainbow trout cardiac myocytes were identified as the Ba²⁺-sensitive background inward rectifier current (I_{Kr}) and the E-4031-sensitive delayed rectifier current (I_{K1}). In atrial cells, the density of I_{K1} is very low and the density of I_{Kr} is remarkably high. The opposite is true for ventricular cells. Acclimation to cold (4°C) modified the two K⁺ currents in opposite ways. Acclimation to cold increases the density of I_{Kr} and depresses the density of I_{K1}. These changes in repolarizing K⁺ currents alter the shape of the action potential, which is much shorter in cold-acclimated than warm-acclimated (17°C) trout. These results provide the first concrete evidence that K⁺ channels of trout cardiac myocytes are adaptable units that provide means to regulate cardiac excitability and contractility as a function of temperature.

IN NORTH TEMPERATE LATITUDES, ectothermic animals can gain partial independence of seasonal temperature changes by adaptive processes that alter the function of cells in a temperature-dependent manner (20). The expression of different isoforms of metabolic enzymes and contractile proteins in muscle cells with altered catalytic properties and temperature tolerance are examples of such adaptation process (15, 32). Fish hearts are no exception in this regard. For example, in rainbow trout, acclimation to low temperatures increases the rate of cardiac contraction and decreases the refractoriness of the heart, which enable higher heart rates than were otherwise possible in the cold (1). However, the subcellular and molecular mechanisms through which this partial temperature independence in excitability and excitation-contraction coupling is achieved are largely unknown.

The shape and duration of cardiac action potential (AP) differ markedly among different vertebrate species and different types of cardiac myocytes, largely owing to a variety of K⁺ conductances in the sarcolemma (SL) (27). The great diversity of K⁺ channels allows fine tuning of the cardiac AP configuration and thus provides the means for regulating myocyte excitability and maintaining electrical stability in the heart. Due to the multiplicity of K⁺ channel types and their functional differences, K⁺ currents also have a central role in cardiac adjustment to altered demand (17, 18, 22, 27, 30). K⁺ channels seem to be plastic entities of the cardiac myocyte, whose expression is changed when environment of the cell is altered, e.g., in cell culture. Accordingly, we reasoned that one potential mechanism of temperature acclimation in fish heart might be modulation of K⁺ conductance and the resulting effects on the shape of the cardiac AP. Therefore, the objective of the current research was to identify the major K⁺ conductances of the rainbow trout cardiac myocytes and to explore whether K⁺ currents are affected by thermal acclimation. It is shown that the two main K⁺ currents of the rainbow trout cardiac myocytes, I_{K1} and I_{Kr}, are changed in opposite manner by cold adaptation: the background inward rectifier current, I_{K1}, is reduced and the rapid component of the delayed rectifier, I_{Kr}, is markedly increased. Temperature-induced changes in K⁺ conductance reduce AP duration and thereby decrease cardiac refractoriness at low ambient temperatures.

MATERIALS AND METHODS

Isolation of atrial and ventricular myocytes. Rainbow trout (mean body mass ± SE of 147 ± 17 g; n = 44) were obtained from a local fish farm (Kontiolahti, Finland) where they had been raised at 9°C. In the laboratory, the fish were randomly divided into two groups and placed in 500-liter stainless steel tanks filled with aerated and circulating (~0.5 l/min) tap water. Water temperature was gradually (1°C/day) changed from 9°C to 17°C [warm-acclimated fish (WA)] or 4°C [cold-acclimated fish (CA)], and the fish were acclimated for >4 wk. The trout were fed commercial food pellets daily (Ewos; Turku, Finland). Photoperiod was a 15:9-h light-dark cycle.

Address for reprint requests and other correspondence: M. Vornanen, Dept. of Biology, Univ. of Joensuu, PO Box 111, 80101 Joensuu, Finland (E-mail: matti.vornanen@joensuu.fi).

http://www.ajpregu.org 0363-6119/02 $5.00 Copyright © 2002 the American Physiological Society R1191
The fish were stunned by a sharp blow to the head and the spine was cut. Atrial and ventricular myocytes were isolated by retrograde perfusion of the heart with a nominally Ca\(^{2+}\)-free isolation solution (in mM: 100 NaCl, 10 KCl, 1.2 KH\(_2\)PO\(_4\), 4 MgSO\(_4\), 50 taurine, 20 glucose, and 10 HEPES at pH 6.9) for 8 min, followed by enzymatic digestion with collagenase (Sigma IA, 1.5 mg/ml) and trypsin (Sigma VIII, 0.5 mg/ml) with added fatty acid-free serum albumin (1 mg/ml; BSA, Sigma) for 20 min at room temperature (~20°C). Atrium and ventricle were separated and cut in small pieces. Myocytes were freed by triturating the muscle pieces through the opening of a Pasteur pipette. Cells were stored in isolation solution at 6°C and were used within 8 h from the isolation.

**Patch-clamp recording.** Standard patch-clamp methods in whole cell configuration (12) were used to record ionic currents and APs in enzymatically isolated cells (28, 29). A small aliquot of dissociated cells were placed in a 150-μl chamber (RC-26, Warner Instrument) mounted on the stage of an inverted microscope (Nikon Eclipse 200). Cells were allowed to adhere to the bottom of the chamber and then were superfused continuously with the external solution prewarmed to either 10 ± 1°C or 20 ± 1°C (25). Complete replacement of external solution was achieved in <1 min with the perfusion rate of ~2 ml/min. Glass pipettes were pulled from borosilicate capillaries (Modulohm) and had a resistance of 1.5–3 MΩ when filled with pipette solutions. Voltage and current clamp recordings were made using an Axopatch 1D amplifier (Axon Instruments) equipped with a CV-4 1/100 headstage. Junction potentials were zeroed before formation of the seal. The pipette capacitance (4–8 pF) was compensated for after formation of the giga seal. The patch was ruptured by delivering a short voltage pulse (zap) to the cell, and capacitive transients were eliminated by iteratively adjusting the series resistance and cell capacitance circuits. The cell capacitance was read directly from the dial of the amplifier. Membrane potentials and currents were low-pass filtered at 10 and 2 kHz, respectively, and were sampled at 5 and 1 kHz with analog-to-digital converter (TL-1 DMA, Axon Instruments). The external solution contained (in mM) 150 NaCl, 5.4 KCl, 1.5 MgSO\(_4\), 0.4 NaH\(_2\)PO\(_4\), 2.0 CaCl\(_2\), 10 glucose, and 10 HEPES at pH 7.6. Na\(^+\) and Ca\(^{2+}\) currents were blocked with 1 μM TTX and 10 μM nifedipine, respectively. The Na\(^+\) channels of the rainbow trout cardiac myocytes are sensitive to TTX with half-maximal and maximal inhibition at the concentration of ~1 and 30 nM, respectively (results not shown). Pipette solution for K\(^+\) current recordings contained (in mM) 140 KCl, 4 MgATP, 1 MgCl\(_2\), 5 EGTA, and 10 HEPES at pH 7.2. For AP recordings, the pipette solution contained (in mM) 140 KCl, 5 Na\(_2\)ATP, 1 EGTA, and 10 HEPES at pH 7.4. TTX and nifedipine were omitted from the external solution when recording APs. Results are given as means ± SE. Currents were normalized to the capacitive membrane area and are expressed as current density (pA/pF) or slope conductance (nS/pF). Differences between acclimation groups were assessed with Student’s t-test with a P value <0.05 as the limit of statistical significance.

**Specific blockers.** The delayed rectifier K\(^+\) channels were selectively blocked with sotalol (Tocris Cookson) or E-4031 (Wako). Sotalol was dissolved in external saline and E-4031 in ethanol. TTX was obtained either from Alomone Labs or Tocris Cookson. Other chemicals were purchased from Sigma.

**RESULTS**

**Steady-state current-voltage relations.** After blocking other membrane currents, K\(^+\) conductances of the SL were measured by clamping the membrane from a holding potential of ~80 mV to various voltages between −120 and +20 mV at 10° or 20°C. The steady-state current-voltage relationship measured at the end of 500 ms square-wave pulses varied greatly between atrial and ventricular cells as well as between WA and CA fish (Fig. 1).

Striking differences appeared between atrial and ventricular myocytes in the densities of inward and outward currents. Atrial cells had little inward current at membrane potentials negative to the equilibrium potential (~74 mV) of K\(^+\) ions, whereas the outward current at the positive side of the equilibrium potential was relatively large. In the ventricular cells, the situation was opposite: the inward current was much larger than the outward current (Fig. 1).

Thermal acclimation strongly modified sarcolemmal K\(^+\) conductances, and the effects were qualitatively similar in atrial and ventricular myocytes. The inward current was strongly reduced by cold acclimation (Fig. 1). The inhibitory effect of cold acclimation on the inward current was particularly clear in ventricular cells where this current component is well developed. In ventricular cells, the slope conductance between −120 and −80 mV (at 20°C) was 0.737 ± 0.094 nS/pF for WA trout but only 0.269 ± 0.035 nS/pF for CA trout (P < 0.001), indicating ~64% loss of the current after acclimation to cold. Due to the very small amplitude of the inward current, the effect of thermal acclimation on K\(_{\text{in}}\) could not be resolved in atrial cells. The slope conductance of the atrial inward current of WA fish (0.022 ± 0.005 nS/pF) was not significantly different from the value of CA fish (0.034 ± 0.005 nS/pF) (20°C) (P = 0.09). Any temperature-induced changes in the inward current might, however, be obscured by leakage current in atrial cells.

In contrast to the inward current, the density of the outward current increased after acclimation to cold (Fig. 1). The densities of the outward current at +20 mV (20°C) were 2.85 ± 0.35 and 0.96 ± 0.30 pA/pF (P < 0.001) in ventricular cells of CA and WA fish, respectively. The corresponding values of the atrial cells were 8.93 ± 0.61 and 1.88 ± 0.32 pA/pF (P < 0.001) for CA and WA fish, respectively. Thus the steady-state current-voltage relations indicate that acclimation to cold suppresses inward K\(^+\) current(s) but enhances late outward K\(^+\) current(s) in the cardiac myocytes of the rainbow trout heart (Table 1).

**Blockade of inward and outward currents.** In vertebrate cardiac myocytes, there are several inwardly rectifying K\(^+\) currents, including background inward rectifier current (I\(_{\text{K1}}\)), ATP-sensitive K\(^+\) current (I\(_{\text{K,ATP}}\)), and acetylcholine-activated K\(^+\) current (I\(_{\text{K,ACh}}\)). The inward current, measured in the absence of external ACh and in the presence of intracellular ATP, was completely blocked by 200 μM BaCl\(_2\) (Fig. 2) but unaffected by 10 μM glibenclamide (data not shown), a blocker of
These findings suggest that the inward current was flowing through the inwardly rectifying background $K^+$ channels.

There are two relatively late activating delayed rectifier currents in vertebrate cardiac myocytes, a rapid methanesulfonanilide-sensitive component ($I_{Kr}$) and a more slowly activating methanesulfonanilide-resistant component ($I_{Ks}$) (11, 21). The delayed rectifier was completely blocked by 2 $\mu$M E-4031 (Fig. 3) and by 100 $\mu$M sotalol (data not shown), selective blockers of $I_{Kr}$. Thus a major part of the outward current in trout cardiac myocytes flows through the rapidly activating delayed rectifier channels. Taken together these blockade experiments strongly suggest that cold-acclimation reduces the background inward rectifier current, $I_{K1}$, and enhances the rapid component of delayed rectifier conductance, $I_{Kr}$, in the rainbow trout cardiac myocytes.

Table 1. Slope conductance of inward current and the density of outward current in atrial and ventricular myocytes of WA and CA rainbow trout

<table>
<thead>
<tr>
<th></th>
<th>$I_{inw}$ (nS/pF) 20°C</th>
<th>$I_{inw}$ (nS/pF) 10°C</th>
<th>$I_{outw}$ (pA/pF) 20°C</th>
<th>$I_{outw}$ (pA/pF) 10°C</th>
</tr>
</thead>
<tbody>
<tr>
<td>CA atrium</td>
<td>0.022 ± 0.005</td>
<td>0.009 ± 0.003</td>
<td>8.93 ± 0.61*</td>
<td>2.49 ± 0.31*</td>
</tr>
<tr>
<td>WA atrium</td>
<td>0.034 ± 0.005</td>
<td>0.014 ± 0.003</td>
<td>1.88 ± 0.32</td>
<td>0.98 ± 0.11</td>
</tr>
<tr>
<td>CA ventricle</td>
<td>0.267 ± 0.035*</td>
<td>0.198 ± 0.029*</td>
<td>2.85 ± 0.35*</td>
<td>0.84 ± 0.16</td>
</tr>
<tr>
<td>WA ventricle</td>
<td>0.737 ± 0.094</td>
<td>0.402 ± 0.026</td>
<td>0.96 ± 0.30</td>
<td>0.89 ± 0.29</td>
</tr>
</tbody>
</table>

Results are means ± SEM from 7 to 15 cells. *Statistically significant difference ($P < 0.05$) between warm-acclimated (WA) and cold-acclimated (CA) fish. $I_{inw}$, steady-state inward current; $I_{outw}$, steady-state outward current.
Characterization of the delayed rectifier current. The current density of the inward rectifier is very small in trout atrial cells and, therefore, the properties of the delayed rectifier currents could be examined in these cells without much interference from the inward K⁺ currents. Current-voltage relation of the delayed rectifier in atrial cells were examined by clamping the membrane for 2,000 ms to potentials between -80 and +80 mV from a holding potential of -40 mV. In myocytes of the CA fish, the amplitude of the activating current (10°C) during the pulse increased with voltage to a maximum (5.03 ± 0.50 pA/pF) at 0 mV and then declined at more positive potentials (Fig. 4). In atrial cells of the WA fish, the current density was much lower (1.53 ± 0.15 pA/pF) (P < 0.001) and the maximum amplitude was attained at +20 mV with some inward rectification at more positive voltages (Fig. 4). The density of the tail current, activated by repolarization to -40 mV, was also much less in WA (1.91 ± 0.19 pA/pF) than CA fish (5.09 ± 0.55 pA/pF) (P < 0.001). These findings indicate that acclimation to cold increases the peak density of I_Kr by 266% in atrial cells of the rainbow trout.

In addition to its sensitivity to class III antiarrhythmic drugs (E-4031, sotalol), the I_Kr of the mammalian cardiac cells is characterized by its inwardly rectifying properties, i.e., the channel conducts inward current much better than outward current. Inward rectification of the I_Kr in trout atrial myocytes was examined by eliciting tail currents with two slightly different voltage protocols. In the first series of experiments, the membrane was depolarized from -40 to +20 mV for 500 ms to activate and inactivate I_Kr and then clamped to various test potentials between -120 and +10 mV to measure the tail current. The initial increase or decrease of the tail current resulted from the recovery of channels from inactivation, and was followed by slower deactivation (Fig. 5A). In these experiments, the in-

---

**Fig. 2. Effects of 200 μM Ba²⁺ on K⁺ currents of trout cardiac myocytes.** Currents were elicited from a holding potential of -80 mV by a slow voltage ramp protocol as indicated at top. Representative current recordings for a ventricular myocyte of warm-acclimated (WA) trout in the absence and presence of Ba²⁺ is also shown (A). Representative current-voltage relationships from similar experiments on atrial and ventricular myocytes of cold-acclimated (CA) and WA trout (B). Experimental temperature was 10°C.
ward currents were substantially larger than the outward currents (Fig. 5C), thus indicating an inward rectification at positive voltages. The inward current was linear within a limited voltage range between $-110$ and $-80$ mV with slope conductances of $0.230 \pm 0.047$ and $0.061 \pm 0.005$ nS/pF ($P < 0.001$) for CA and WA fish, respectively.

The inward rectification of $I_{Kr}$ is considered to be due to the rapid inactivation (i.e., at positive voltages the channels tend to move from resting state directly, without opening, to inactivated state), which can be removed by a short hyperpolarizing voltage step (26). In accordance with this, the inward rectification was largely abolished when the above voltage protocol was modified to include a 5-ms hyperpolarizing voltage step (26). Inward rectification was also observed when the conditioning voltage was $-120$ mV after depolarization to $+20$ mV (Fig. 5B). After removing the inactivation by the short hyperpolarization, the channels were conducting also in outward direction and the resulting instantaneous current-voltage relation was linear up to $+20$ mV (Fig. 5B and C). Slope conductance of this linear current was 2.5 times higher in CA than WA trout ($0.25 \pm 0.05$ vs. $0.10 \pm 0.04$ nS/pF).

**APs.** Atrial and ventricular APs were recorded in the absence and presence of $2 \mu M$ E-4031, a specific blocker of the fast component of the delayed rectifier, at $10^\circ C$ (Fig. 6). There were marked differences in AP waveforms between atrial and ventricular myocytes. In atrial myocytes, resting membrane potential (RP) was less negative and AP duration much shorter than in ventricular cells (Fig. 6). The RP of unstimulated atrial cells was $-44 \pm 5$ and $-47 \pm 4$ mV ($P > 0.05$) for CA and WA fish, respectively. When stimulated to produce APs, repolarization of AP was followed by an undershoot to a maximum diastolic potential (MDP) of about $-65$ mV and subsequent diastolic depolarization toward RP. In contrast to atrial cells, ventricular myocytes had stable RPs measuring $-77 \pm 1.7$ (n = 17) and $-79 \pm 3.3$ mV (n = 11) for CA and WA trout, respectively. There was also considerable differences in AP duration between acclimation groups. In both atrial and ventricular myocytes, the duration of AP was much shorter in CA than WA trout (Fig. 6). Furthermore, AP duration was prolonged by blocking of the $I_{Kr}$ with $2 \mu M$ E-4031. The response to E-4031 could not be quantified in atrial myocytes, because E-4031 depolarized.
ized the RP and made the myocytes unexcitable. In ventricular cells, E-4031 had no effect on the RP but markedly increased the duration of AP (Fig. 7).

DISCUSSION

Potassium conductances of trout atrial and ventricular myocytes. The present results indicate that the two major components of sarcolemmal K⁺ conductance of the rainbow trout cardiac myocytes are the E-4031-sensitive delayed rectifier current, Iₖr, and the Ba²⁺-sensitive background inward rectifier current, Iₖ₁. Furthermore, thermal acclimation modifies these K⁺ conductances in opposite manners: Iₖr is increased and Iₖ₁ decreased in the cold.

Iₖ₁ belongs to the Kir2-family of the inwardly rectifying currents (for review, see Ref 27). Due to its large conductance around the Nernst potential of K⁺ ions and its negative slope conductance between −20 and −60 mV, Iₖ₁ maintains a stable RP and contributes to the final repolarization of the AP without having much effect on plateau duration (27). In mammalian cardiac myocytes, the channels responsible for Iₖ₁ are mainly located in t-tubular membrane (5). Because rainbow trout cardiac myocytes do not have any t tubules (23), the spatial distribution of Iₖ₁ channels in the fish SL must differ from that of mammals. The location of the channels in the peripheral SL might reduce or eliminate the depolarizing effect of fast spacing, which is considered to occur in mammalian hearts due to the accumulation of K⁺ in the narrow t tubules. The exact spatial distribution of the Iₖ₁ channels and its physiological consequences in fish cardiac myocytes remains to be shown.

In agreement with the earlier findings from mammalian (9, 14) and fish hearts (29), the density of Iₖ₁ was much higher in ventricular than atrial cells. The density of Iₖ₁ in ventricular cells of the rainbow trout heart is similar to what was recently measured in crucian carp (Carassius carassius) ventricular myocytes (29). In contrast, the density of Iₖ₁ in trout atrial cells is exceptionally low, even if compared with the atrial cells of the crucian carp heart (29). The small
magnitude of \( I_{K1} \) explains the relatively depolarized RP of the isolated trout atrial cells.

The delayed rectifier currents (\( I_{K} \)) gradually develop during the plateau phase of AP and conduct outward current at more positive voltages than the background inward rectifier channels. In mammalian hearts, \( I_{K} \) is comprised of two pharmacologically and biophysically distinct current components, rapid (\( I_{KR} \)) and slow (\( I_{KS} \)) delayed rectifiers (17). Two characteristics, complete block by low concentrations of E-4031 and inward rectification due to rapid inactivation, indicate that the delayed rectifier current of the trout atrial and ventricular myocytes is almost exclusively carried by the rapid component of the delayed rectifier, \( I_{KR} \). In contrast to the mammalian cardiac myocytes, where \( I_{KR} \) is usually a minor component among the diverse \( K^+ \) conductances (4, 7, 10, 31), the trout cardiac myocytes (especially the atrial cells) have a very prominent \( I_{KR} \).

In atrial and ventricular myocytes of the mammalian heart, the delayed rectifier regulates the plateau phase of AP but has little effect on the rate of final repolarization or on RP (6, 17, 22). E-4031-induced prolongation of AP and depolarization of RP suggest that in atrial cells of the rainbow trout heart, \( I_{KR} \) not only regulates the duration of AP but may also contribute to the maintenance of RP. Similarly, \( I_{KR} \) is involved in maintenance of RP in the esophageal smooth muscle cells (3, 19) and determines MDP of the cardiac pacemaker cells (19). In isolated atrial cells of the trout heart, the AP undershoot (hyperpolarization) and diastolic depo-
larization were regular findings and they were abolished by E-4031. Thus, in isolated atrial myocytes, $I_{Kr}$ seems to determine MDP and the decay of $I_{Kr}$ probably causes the diastolic depolarization. In ventricular myocytes of the trout heart, the large $I_{K1}$ sets RP and accordingly the inhibition of $I_{Kc}$ had no effect on RP.

Effect of temperature acclimation on $K^+$ conductances. Acclimation to cold depressed $I_{K1}$ of the ventricular myocytes and strongly increased $I_{Kr}$ in both atrial and ventricular myocytes of the rainbow trout heart. The cold-induced increase in $I_{Kr}$ can be regarded as compensatory adaptation, which limits AP duration (present study) and decreases refractoriness of the heart (1). This will allow compensatory increases in heart rate (2) and high cardiac output in cold-adapted trout.

The adaptive significance of the cold-induced depression of $I_{K1}$ is less evident, but might produce energy savings by reducing the demand for ATP-dependent ion pumping across the SL. Moreover, reduction of $I_{K1}$ might increase the excitability of the ventricular myocytes, because there would be less outward current opposing the initial depolarization in the beginning of AP. The reduction of $I_{K1}$ did not cause, however, any significant depolarization of RP in ventricular cells of the trout heart. In atrial cells, $I_{K1}$ was so small that RP must be, at least partially, maintained by the much larger $I_{Kr}$. Furthermore, it is unlikely that the atrial RP in vivo would be as depolarized as it was in enzymatically isolated myocytes. Above all, the tonic parasympathetic tone of the intact fish heart (8, 24) would be assumed to have an effect on atrial $K^+$ currents. The densities of the inwardly rectifying $K^+$ currents might in vivo be much larger than recorded in single atrial myocytes due to $G_{K1}$ protein-mediated stimulation of $I_{K,Ca}$. Channels (16).

Fig. 6. Effect of temperature acclimation on action potential (AP) duration of ventricular (A) and atrial (B) myocytes from rainbow trout heart. Thin and thick lines mark tracings from WA and CA fish, respectively. Bar graphs show mean (±SE) results for AP duration at 50% (APD50) and 90% (APD90) repolarization levels. *Statistically ($P < 0.05$) significant difference between WA and CA fish.

Fig. 7. Effect of 2 μM E-4031 on the duration of ventricular AP in CA (A) and WA (B) rainbow trout. The duration of AP increases with time after E-4031 application. Experimental temperature was 10°C.
more negative (about −65 mV; unpublished observations) than in isolated atrial myocytes.

**Perspectives**

The present findings indicate that sarcolemmal K\(^+\) currents are sensitive to chronic temperature stress in a eurythermic fish species and that temperature-dependent expression of K\(^+\) currents modifies AP duration and consequently cardiac excitability. In addition to regulation of excitability and refractoriness of the heart, K\(^+\) currents are closely involved in the regulation of cardiac contractility. AP duration and plateau height have strong effects on sarcolemmal Ca\(^{2+}\) influx through L-type Ca\(^{2+}\) channels and Na\(^+\)/Ca\(^{2+}\) exchange (25) and hence temperature-induced changes in repolarizing currents (\(I_{K1}\) and \(I_{K2}\)) might have a significant indirect effect on Ca\(^{2+}\) management of the fish cardiac myocyte. The shorter AP of the CA fish will probably allow less sarcolemmal Ca\(^{2+}\) influx during single excitation, but on the other hand the compensatory increase in heart rate, owing to the reduced refractoriness, will allow a larger frequency-dependent Ca\(^{2+}\)-loading of the SR (13). Therefore, it seems that the plasticity of excitation and excitation-contraction coupling of cardiac myocytes in fish is significantly based on temperature-induced changes in K\(^+\) conductances of the SL. The task of future research is to examine the presence of this acclimatory response in other ectotherms and to clarify its significance to cardiac contractility in species inhabiting different thermal environments.

This study was supported by a grant from the Academy of Finland (project No. 63090) to M. V. Kontiolahti fish farm is gratefully acknowledged for supplying the trout.

**REFERENCES**


