Anion Permeability and Conduction of Adenine Nucleotides Through a Chloride Channel in Cardiac Sarcoplasmic Reticulum*

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Cardiac sarcoplasmic reticulum (SR) membrane contains several chloride (Cl⁻) channels. We have characterized a 116-pS Cl⁻ channel (500 mM cis, 50 mM trans Cl⁻) in cardiac SR that is activated by protein kinase A-dependent phosphorylation. To understand its function further, we examined the permeation of various anions and adenine nucleotides using the planar lipid bilayer-vesicle fusion technique. This Cl⁻ channel showed a high selectivity to anions and its permeability sequence was Br⁻ > Cl⁻ > I⁻ > NO₃⁻ > F⁻. When all anions were replaced with ATP in the cis solution, channel activity persisted. The conductance was 78 pS with 200 mM ATP and 68 pS with 100 mM ATP. The reversal potentials were +63 mV and +41 mV in 200 mM ATP and in 100 mM ATP, respectively. With 100 mM ADP or AMP in the cis solution, channel activities were also observed. The conductances were 87 pS with 100 mM ADP and 115 pS with 100 mM AMP. The apparent adenine selectivity of this channel was ATP > ADP > AMP, assuming that they exist as divalent anions. These results suggest that the SR Cl⁻ channel in cardiac cells may serve as a transporter for the movement of adenine nucleotides between cytosol and SR lumen.

Recently, it has been suggested that certain types of anion channels in the plasma membrane conduct adenine nucleotides such as ATP (9–11), but this suggestion has been contested (12–14). It has also been suggested that the mitochondrial voltage-dependent anion channel (VDAC) (15) may conduct adenine nucleotides. It is not known, however, whether the 116-pS Cl⁻ channel in cardiac SR conducts ATP or not. In the present study, we examined the anion permeability and whether ATP might permeate through this Cl⁻ channel or not. We found that this channel could conduct adenine nucleotides, ATP, ADP, and AMP. A preliminary report of this work was presented in abstract form (16).

EXPERIMENTAL PROCEDURES

Preparation—Porcine cardiac heavy SR was isolated by discontinuous sucrose gradient centrifugation as reported previously (5, 6).

Solutions and Drugs—The cis bath solution contained (mM) CsCl, 500; EGTA, 1; HEPES, 10; MgATP, 2; and CaCl₂, 5. The free Ca²⁺ concentration was 1 μM calculated with the method proposed by Fabiato (18). The trans solution contained (mM) CsCl, 50; EGTA, 1; HEPES, 10; and 1 μM free Ca²⁺. The pH of these solutions was adjusted to 7.3 by adding CsOH. In the experiments to determine anion permeability, CsCl was replaced by 500 mM CsBr, CsF, CsNO₃, or CsI. The 200 mM ATP solutions were made by replacement of 500 mM CsCl with 198 mM Tris ATP, Na₂ATP, K₂ATP, or Cs₂ATP and 2 mM MgATP. In some experiments, 500 mM CsCl was replaced with 200 mM MgATP. The 100 mM ATP solutions were made by replacing 100 mM CsCl with 98 mM Tris ATP or Na₂ATP and 2 mM MgATP. 10 μM ryanodine (Wako Chemical Co., Osaka, Japan) was added to the cis solution to block the RyR channels. A protein kinase inhibitor (PKI) specific to PKA (Sigma P3294) was purchased from Sigma Chemical Co. Enzymes were diazotized against the cis solution at 0 °C for 1 h before use. DIDS (Sigma) was dissolved in dimethyl sulfoxide (Me₂SO). Concentrations of Me₂SO in final solutions were less than 0.1%.

Electrophysiological Methods and Data Analysis—The planar bilayer was composed of brain phosphatidylethanolamine and brain phosphatidylserine (Avanti Polar Lipids, Alabaster, AL) at a ratio of 1:1, dissolved in decane (20 mg/ml). Purified cardiac heavy SR vesicles were added to the cis chamber and fused into the lipid bilayer formed in the hole (0.25 mm in diameter) in a Lexan polycarbonate partition. In the present experiments, the cis chamber was defined as the side to which SR vesicles were added, and the opposite side was referred to as the trans chamber. The cis side was equivalent to the cytoplasmic side of the incorporated channel, and the trans side was equivalent to the lumen of the SR as determined previously (5, 6). Currents flowing through the ion channels were measured by using the voltage-clamp technique. Applied voltages were defined with respect to the trans chamber held at ground. Channel activities were recorded at room temperature (22 ± 1 °C), amplified by a patch-clamp amplifier (Axopatch 1C, Axon Instruments, Inc, Foster City, CA), and stored on a videocassette tape recorder through a PCM converter system (RP-880, NF Instruments, Yokohama, Japan) digitized at 10 kHz. Data were reproduced and low pass filtered at 2,000 or 1,000 Hz by a filter with Bessel characteristics (octave attenuation, 48 dB) and analyzed off-line on a computer (P5–200, Gateway 2000). For single-channel analysis, the threshold used to judge to open state was set at a half-amplitude of the single-channel currents (19).

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The abbreviations used are: SR, sarcoplasmic reticulum; VDAC, voltage-dependent anion channel; DIDS, 4,4'-diisothiocyanatostilbene-2,2'-disulfonic acid; PKA, protein kinase A; CFTR, cystic fibrosis transmembrane regulator; pS, picosiemens; PKI, protein kinase inhibitor.

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RESULTS
Permeation of Various Anions—We have reported that a 116 pS Cl\textsuperscript{2}\textsuperscript{-} channel in cardiac SR is activated via PKA-dependent phosphorylation or in the presence of MgATP (5, 6). Fig. 1, panel A, shows a continuous recording of Cl\textsuperscript{2}\textsuperscript{-} channel openings with 500 mM CsCl in the cis and 50 mM CsCl in the trans chamber solutions. Because SR membrane contained not only Cl\textsuperscript{2}\textsuperscript{-} channels, but also ryanodine receptor Ca\textsuperscript{2}\textsuperscript{+} release channels (RyRs) and K\textsuperscript{+} channels (2, 7), these channels were blocked by replacement of K\textsuperscript{+} with Cs\textsuperscript{+} and the application of 10 \textmu M ryanodine to the cis solution. The current amplitude of the channel remaining after this treatment was −15 pA at −40 mV. In the absence of MgATP, channel activity ran down spontaneously within several minutes after incorporation into the lipid bilayer. In the presence of 2 mM MgATP, however, channel openings were sustained until the experiments were interrupted by a break of bilayer (Fig. 1, panel A, b). The open-time and closed-time histograms could be fitted by the sum of two exponentials (Fig. 5, panel A). The time constants for the open-time histogram were 1.5 ± 0.2 (mean ± S.D.) ms (n = 6) and 43 ± 4 ms (n = 6). The time constants for the closed-time histograms were 0.7 ± 0.1 ms (n = 6) and 5.0 ± 0.5 ms (n = 6). 

**FIG. 1.** Single-channel activity of Cl\textsuperscript{-} channel. Panel A, tracings showing the single-channel activity of the SR Cl\textsuperscript{-} channels incorporated into planar lipid bilayers. The continuous recording of Cl\textsuperscript{-} channel activity is shown in a. The cis solution contains 500 mM Cl\textsuperscript{-} and the trans solution contains 50 mM Cl\textsuperscript{-}. Voltage was held at −40 mV. Channel activity ran down soon after being incorporated into the planar lipid bilayer in the absence of MgATP in the cis solution. Shown in b, in the presence of 2 mM MgATP in the cis solution, the Cl\textsuperscript{-} channel openings were sustained until the break of bilayer. o, open channel; c, closed channel. Downward deflection indicates current flow from the trans to cis side. Panel B, current traces with various anions through Cl\textsuperscript{-} channel. Cl\textsuperscript{-} ions in both cis and trans solutions were replaced with various anions: Br\textsuperscript{-}, I\textsuperscript{-}, NO\textsubscript{3}\textsuperscript{-}, and F\textsuperscript{-}, respectively. The concentration of each anion is 500 mM in the cis solution and 50 mM in the trans solution. In all experiments, the cis solutions contained 2 mM MgATP. Holding potentials are indicated on the left side of each tracing. Panel C, the plot of unitary current amplitude (mean ± S.D.) with five different anions as a function of voltages. All currents reached 0 current level at around +60 mV. The slope conductance of 500 mM Cl\textsuperscript{-} (n = 18) and Br\textsuperscript{-} (n = 5) is about 116 pS and those of 500 mM NO\textsubscript{3}\textsuperscript{-} (n = 6), I\textsuperscript{-} (n = 6), and F\textsuperscript{-} (n = 6) are about 40 pS.

**FIG. 2.** Anion selectivity. Current traces were recorded with 500 mM various anions (Br\textsuperscript{-}, I\textsuperscript{-}, NO\textsubscript{3}\textsuperscript{-}, or F\textsuperscript{-}) in the cis solution and 50 mM Cl\textsuperscript{-} in the trans solution. Holding potentials are indicated on the left side of each trace.
ms (n = 5). Thus, the kinetic properties and channel conductance were identical to those of the 116 pS Cl⁻ channel reported previously (5, 6). These channel activities could be restored in the presence of 2 mM or higher MgATP or by adding of PKA with 0.05 mM MgATP. Therefore, we performed the following experiments with the cis solution containing 2 mM MgATP.

To test the anion selectivity, current-voltage relationships were obtained with various anions in both the cis and trans solutions. When Cl⁻ was replaced with equimolar Br⁻, the unit amplitude of the current was not changed significantly (Fig. 1, panel B). The slope conductance with Br⁻ was about 116 pS, which was identical to that with Cl⁻. When Cl⁻ was replaced with equimolar I⁻, NO₃⁻, or F⁻, the unit amplitudes were decreased (Fig. 1, panel C). The slope conductances were 40 pS under these conditions. The reversal potential (E_{rev}) was approximately equal to the calculated equilibrium potential, which was +58 mV (Fig. 1, panel C). From these results, we concluded that this SR-Cl⁻ channel was highly permeable to these anions.

|| Anion | E_{rev} (mV) | n | P_{anion}/P_{Cl} |
|---|---|---|---|
| Cl⁻ | 60 ± 0.5 | 18 | 1.0 |
| Br⁻ | 75 ± 10 | 5 | 2.1 |
| I⁻ | 45 ± 10 | 5 | 0.59 |
| NO₃⁻ | 28 ± 6.8 | 5 | 0.30 |
| F⁻ | 23 ± 2.9 | 4 | 0.25 |

E_{rev}, reversal potential; P, permeability; n, numbers of experiments.

ATP Conduction through the Cl⁻ Channel—We examined
FIG. 4. ATP currents at different concentrations of ATP. Panel A shows single-channel activity at different holding potentials indicated on the left of each tracing in 200 and 100 mM ATP in the cis solution. Panel B, the plot of unitary current amplitude (mean ± S.E.) with different ATP concentrations in the cis solution as a function of holding potential. Open circles (○) indicate current with 100 mM ATP (n = 9 experiments) and closed circles (●) indicate current with 200 mM ATP (n = 15 experiments).

FIG. 5. Kinetics of Cl– channel activity with Cl– and ATP. Panel A, open-time and closed-time histograms with 500 mM Cl– in the cis solution are shown at –60 mV holding potential. The open-time histogram was fitted by double exponential curves with time constants of the fast (T1) and the slow component (T2) of 1.5 and 43 ms, respectively. The closed-time histogram was fitted by double-exponentials with T1 and T2 of 0.6 and 5.0 ms, respectively. Panel B, when all Cl– ions were replaced with 100 mM ATP, open-time and closed-time histograms at –60 mV and –80 mV are shown. At –60 mV both open-time and closed-time histograms are fitted by single exponential with time constants of 28 and 0.4 ms, respectively. At –80 mV, both histograms were fitted by a single exponential with time constant of 21 and 0.4 ms, respectively. All cis bath solutions contained 2 mM Mg-ATP.
whether adenine nucleotides permeated this Cl\textsuperscript{−} channel. After incorporation of Cl\textsuperscript{−} channel into the lipid bilayers, 500 mM Cl\textsuperscript{−} in the cis solution was replaced with 200 mM ATP (198 mM Tris ATP and 2 mM MgATP). Fig. 3 shows a continuous recording of channel openings before and after replacement of the cis solution, where the membrane potential was held at −80 mV. After replacement of 500 mM Cl\textsuperscript{−} with 200 mM ATP, the current amplitudes decreased (Fig. 3, panel A). We have often observed a subconductance level of the SR-Cl\textsuperscript{−} channel in the Cl\textsuperscript{−} solution and similar subconductance levels were recognized in the ATP solution as shown in Fig. 3, panel B.

To confirm the ATP conduction, current-voltage relationships were obtained at two different ATP concentrations in the cis solution and 50 mM Cl\textsuperscript{−} in the trans solutions (Fig. 4). With 200 mM ATP, the slope conductance of the inward current was 83 pS (\(n = 15\)), and \(E_{rev}\) was +62.6 mV. With 100 mM ATP, the slope conductance decreased to 68 pS (\(n = 9\)) and \(E_{rev}\) shifted to +40.5 mV (Fig. 4, panel B). Under these experimental conditions, the only anion present in the cis solution was ATP. Therefore, inward currents could only be carried by ATP at negative potentials. It is difficult to estimate the theoretical \(E_{rev}\) for the ATP current, because we do not know the actual free and complexed ATP concentrations under our experimental conditions. If ATP was assumed to exist as a divalent anion, \(F_{ATP}/F_{Cl}\) was 0.54 at 200 mM ATP and was 0.5 at 100 mM ATP calculated by the following equation (20).

\[
E_{rev} = \frac{RT}{2F} \times \ln(4P_{ATP}[ATP]_{cis}/P_{Cl}[Cl]_{trans}).
\]  
(Eq. 2)

The value of \(E_{rev}\) at each condition was almost identical to the theoretical value. From these results, we concluded that the currents were mostly carried by ATP\textsuperscript{2−}.

**The Kinetics of ATP Currents**—As reported previously (5, 6), the channel open-time and closed-time histograms could be fitted with two exponentials when currents were carried by Cl\textsuperscript{−} (Fig. 5, panel A). By analyzing the currents carried by ATP, the open-time histograms could be fitted by a single exponential with time constants of 26 ± 11 ms at −60 mV and 23 ± 10 ms at −80 mV (\(n = 4\)). The fast component of the open-time histogram observed when chloride was charge carrier seemed to disappear when ATP was the charge carrier. The closed-time histogram analyzed with a single-active channel could be fitted by a single exponential with time constants of 0.4 ± 0.4 ms at −60 mV and −80 mV (\(n = 4\)). The fast component was not different from those in the Cl\textsuperscript{−} currents. Thus, the slow component of the closed-time histogram with chloride as charge carrier disappeared with ATP as charge carrier (Fig. 5, panel B). In this analysis we chose the data in which only one channel existed in the bilayer and no subconductance levels were observed.

**ADP and AMP Conduction through the Cl\textsuperscript{−} Channels**—We examined whether SR-Cl\textsuperscript{−} channel might conduct other adenine nucleotides besides ATP. ADP and AMP were tested using the same approach as shown in Fig. 3, panel A. When the cis solution was replaced with 100 mM Na\textsubscript{2}ADP or Na\textsubscript{2}AMP for 500 mM CsCl, the channel activities were maintained (Fig. 6). Thus,

![Fig. 6. ADP and AMP conduction through Cl\textsuperscript{−} channel. Panel A, a shows single-channel activities in 100 mM ADP in the cis solution and 50 mM Cl\textsuperscript{−} in the trans solution at −80 mV. Amplitude histogram with Gaussian fit is shown in b. In c, open- and closed-time histograms are shown. Both histograms were fitted by double exponential curves with time constants of 4.5 and 49 ms in open time and 0.2 and 163 ms in closed time. Panel B, a shows single-channel activities in 100 mM AMP in the cis and 50 mM Cl\textsuperscript{−} in the trans solutions at −100 mV. In b, the amplitude histogram is shown. In both ADP and AMP current measurements, the cis solution contained 2 mM MgATP.](http://www.jbc.org/)

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**Note:** The image contains a figure (Fig. 6) illustrating the conduction of ADP and AMP through the Cl\textsuperscript{−} channel. The figure includes panels showing single-channel activities, amplitude histograms, open- and closed-time histograms, and the theoretical calculation of \(E_{rev}\).
the SR-Cl\textsuperscript{−} channel also conducts these adenine nucleotides. We compared the slope conductances and relative permeabilities in bi-ionic conditions with 100 mM ATP, ADP, or AMP in the cis and 50 mM Cl\textsuperscript{−} in the trans solution. The slope conductance was 68 pS in 100 mM ATP, 87 pS in ADP, or 115 pS in AMP. The reversal potentials were +41 mV, +20.6 mV, and +3.0 mV, respectively (Fig. 7). The relative permeabilities ($P_A/P_{Cl}$) are summarized in Table II.

**Modulation of the Currents by Protein Kinase A-dependent Phosphorylation**—If these currents carried by adenine nucleotides were activated via PKA-mediated phosphorylation as reported previously (5, 6), currents should be blocked by removal of MgATP from the cis solution or by the application of PKI. After the ADP currents were activated in the presence of 2 mM MgATP, MgATP in the cis solution was removed. The ADP current was quickly and completely blocked (Fig. 8A), and restored by the reapplication of 2 mM MgATP (Fig. 8B). The application of PKI completely blocked the channel openings (Fig. 8C). Therefore we conclude that the anion channel-conducting adenine nucleotides can be activated via PKA-dependent phosphorylation.

**DISCUSSION**

**Anion Selectivity of Cl\textsuperscript{−} channel in the cardiac SR**—The order of anion selectivity of this Cl\textsuperscript{−} channel is consistent with the predicted Eisenman’s sequence III (21). These results suggest that the electrodiffusion through this Cl\textsuperscript{−} channel may be controlled by a so-called “weak field strength” selectivity site (21). The selectivity sequence of other Cl\textsuperscript{−} channels in cardiac or skeletal muscle SR are different from this (3, 8, 22). The Ca\textsuperscript{2+} - and voltage-sensitive Cl\textsuperscript{−} channel in cardiac SR displayed the order of SCN\textsuperscript{−} > I\textsuperscript{−} > NO\textsubscript{3}\textsuperscript{−}, Br\textsuperscript{−} > Cl\textsuperscript{−} > F\textsuperscript{−} > HCO\textsubscript{3}\textsuperscript{−}, which is consistent with sequence 1 of Eisenman (8). In skeletal muscle SR, the Cl\textsuperscript{−} channels displayed the sequence of NO\textsubscript{3}\textsuperscript{−} > Br\textsuperscript{−} > Cl\textsuperscript{−} (1) or NO\textsubscript{3}\textsuperscript{−} > SCN\textsuperscript{−} > I\textsuperscript{−} > Br\textsuperscript{−} = Cl\textsuperscript{−} (22). Therefore, we speculate that SR-Cl\textsuperscript{−} channel in this study may be a different type of Cl\textsuperscript{−} channel than others previously described in SR. The cystic fibrosis transmembrane regulator (CFTR) expressed in epithelial cells or cardiac sarcolemma has an anion selectivity sequence similar to the 116 pS SR Cl\textsuperscript{−} channel (23, 24). In addition, both CFTR and the 116 pS SR Cl\textsuperscript{−} channel are activated by PKA-dependent phosphorylation (Fig. 1) (5, 6), and exhibit voltage-independent activation (Figs. 1 and 2; Table I) (25). However, the conductance of the Cl\textsuperscript{−} channel in cardiac SR is much larger than that of CFTR (Fig. 1). Therefore, it seems unlikely that the SR-Cl\textsuperscript{−} channel is CFTR.

**Permeation Pathway for Cl\textsuperscript{−} and Adenine Nucleotides**—The data in this report first demonstrate ATP currents in cardiac SR by recording single-channel activities (Figs. 3 and 4). Our results suggest that ATP is permeable through the same channel, which mediates Cl\textsuperscript{−} permeation. First, the continuous channel activity before and after replacement of Cl\textsuperscript{−} with ATP (Fig. 3, panel A) is consistent with this interpretation. The alternative, that new channels are incorporated into the bilayer as a consequence of ATP addition, seems highly unlikely. Furthermore, we could always detect the channel openings in

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**Table II**

| Adenine | $E_{rev}$ (mV) | Conductance (pS) |
|---------|----------------|-----------------|
| 100 mM AMP | 3.0 | 115 |
| 100 mM ADP | 20.6 | 87 |
| 100 mM ATP | 41 | 68 |

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**Fig. 7. ADP and AMP currents, through Cl\textsuperscript{−} channel.** Panel A shows single-channel activities in 100 mM AMP in the cis solution (left) and 100 mM ADP (right) at different holding potentials indicated at the left of each tracing. The cis solution contained 2 mM MgATP. Panel B, the plot of unitary current amplitude (mean ± S.D.) with 100 mM Na\textsubscript{2}ATP ($n$ = 9), 100 mM Na\textsubscript{2}ADP ($n$ = 8), and 100 mM Na\textsubscript{2}AMP ($n$ = 5) as a function of holding potential.
the ATP solutions, whenever the activity of Cl⁻ channels is incorporated into the bilayer was recorded (15/15). On the other hand, ATP currents were never observed when Cl⁻ channels were not incorporated into the bilayer (10/10). Second, the anion channel requires phosphorylation to conduct either Cl⁻ or ATP (Fig. 8). Third, the pharmacological properties are similar for both currents. We have reported that the SR-Cl⁻ channel is insensitive to DIDS (5). Likewise, DPC or DIDS did not block ATP or ADP currents (data not shown). Therefore, we conclude that the 116 pS Cl⁻ channel in cardiac SR can conduct both Cl⁻ and adenine nucleotides.

Our data are in agreement with other reports indicating the existence of ATP conduction pathway in intracellular membranes (26–31). It is known that VDAC (mitochondrial porin) is responsible for most of the metabolite flux across the mitochondrial outer membrane and also provides a pathway for nucleotide transport (28–31). A working model of the VDAC pore proposes a barrel with a diameter of 2.4–3.0 nm, and it has been reported that VDAC is sufficient to mediate ATP flux through the mitochondrial membrane (32, 33). The molecular structures of ATP channels in a variety of intracellular membranes, including SR, have not been identified. Further studies are required to clarify the structural basis for the ATP conduction through the inner membrane channels.

In contrast to the inner membrane channels, there is no consensus regarding the existence of any ATP conduction pathway in sarcosomal membranes. Although recent studies have suggested ATP conduction through CFTR or the multidrug resistance channel (9–11), conflicting results have also been presented (12–14). Furthermore, it is suggested that the size of the ATP anion is much larger than the estimated size of the CFTR pore (34). A different view of these controversial findings has been presented by Pasyk and Foskett (26) and Sugita et al. (34), showing the existence of the CFTR-associated ATP channels in the plasma membrane by patch-clamp technique. Thus, an ATP conducting pathway appears to exist in the plasma membrane, but the identity of the responsible channels, whether they are the same or different from CFTR, are not clear.

**Selectivity of Adenine Nucleotides**—Based on the theoretical value of $E_{\text{rev}}$ at different ATP concentrations, our results were consistent with the assumption that ATP might move predominantly as a divalent anion (Fig. 4). Observed $P_{\text{ATP/Cl}}$ ratios did not fit with calculations based on ATP as a monovalent anion (calculated $P_{\text{ATP/Cl}}$ ratio was 0.73 at 200 mM ATP and 1.01 at 100 mM ATP), tetravalent ($P_{\text{ATP/Cl}} = 0.527$ at 100 mM ATP and 1.168 at 200 mM ATP) or trivalent ($P_{\text{ATP/Cl}}$ is 0.462 at 100 mM ATP and 0.71 at 200 mM ATP) anion. This suggests that ATP permeation as ATP⁴⁻, ATP³⁻, or ATP⁻ is unlikely. Sugita et al. (35) showed the ATP currents through CFTR-associated ATP channels with $P_{\text{ATP/Cl}} = 0.4$. This value is almost identical to our results (0.5). We have also demonstrated the permeation of ADP and AMP through this Cl⁻ channel (Figs. 6 and 7) and tried to determine the selectivity of adenine nucleotides (Tables I and II). It was quite difficult, however, to estimate precisely free concentrations of these anions in solution or those valences as charge carriers. If ADP and AMP were assumed to be present as a monovalent anion, $P_{\text{ADP/Cl}}$ was 0.71 and $P_{\text{AMP/Cl}}$ was 0.527. The order of apparent permeability became ADP > ATP > AMP. If they were assumed to be divalent anions, $P_{\text{ADP/Cl}}$ was 0.25, and $P_{\text{AMP/Cl}}$ was 0.14. Then the order of apparent permeability became ATP > ADP > AMP.

**Physiological Implication**—In the lumen of intracellular organelles, many processes may require adenine nucleotides for the functional source, and intraluminal ATP may be physiologically regulated. ATP is needed for energy-requiring processes (36) and is also the substrate for phosphorylation of intraluminal proteins in ER (37–39). Luminal ATP is required for protein translocation in the ER (40–42). Therefore, we speculate that the SR Cl⁻ channel may mediate the transport of ATP between lumen and cytosol, which may be responsible for important regulatory functions in cardiac excitation-contraction coupling.

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