

## **TD1**

Quel type d'ion passe dans ce canal ?

Expérience en V-clamp ou I-clamp ?

Protocole de stimulation ?

Courant temps-dépendant (inactivation, activation) ? voltage-dépendant ?

Modèle ?

Courant total ou unitaire ?

Réalisé en patch-clamp ou en double-microélectrodes ?

Résultats et conclusion ?

## **TD2**

Quel type d'ion passe dans ce canal ?

Expérience en V-clamp ou I-clamp ?

Protocole de stimulation ?

Courant temps-dépendant (inactivation, activation, rectification) ? voltage-dépendant ?

Courant total ou unitaire ?

Réalisé en patch-clamp ou en double-microélectrodes ?

Effets de la concentration en  $K^+$  sur le potentiel d'équilibre au  $K^+$  et sur la conductance ?

Résultats et conclusion ?

## **TD3**

Quel type d'ion passe dans ce canal ?

Expérience en V-clamp ou I-clamp ?

Courant temps-dépendant (inactivation, activation, rectification) ? voltage-dépendant ?

Courant total ou unitaire ?

Potentiel d'inversion du courant ?

Seuil d'activation ?

Conductance maximale à quel potentiel ?

Effet de la sous-unité ?

Conclusion ?

## **TD4**

Quel type d'ion passe dans ce canal ?

Expérience en V-clamp ou I-clamp ?

Courant temps-dépendant (inactivation, activation) ? voltage-dépendant ?

Courant total ou unitaire ?

Effet du Bay-K, de la toxine, et du PMA sur les courants ?

Conclusion ?

### **TD5-TD6**

Quel type d'ion passe dans ce canal ?

Expérience en V-clamp ou I-clamp ?

Courant temps-dépendant (inactivation, activation, rectification) ? voltage-dépendant ?

Courant total ou unitaire ?

Potentiel d'inversion du courant ?

Seuil d'activation ?

Effets du baryum et du césium (voltage-dépendant, temps-dépendant) ?

Conclusion ?

### **TD7**

Technique utilisée ?

Quel type d'ion passe dans ce canal ?

Expérience en V-clamp ou I-clamp ?

Courant total ou unitaire ?

Effets du calcium, de l'ATP et du magnésium ?

Conclusion ?

### **TD8**

Quel type d'ion passe dans ce canal ?

Expérience en V-clamp ou I-clamp ?

Courant total ou unitaire ?

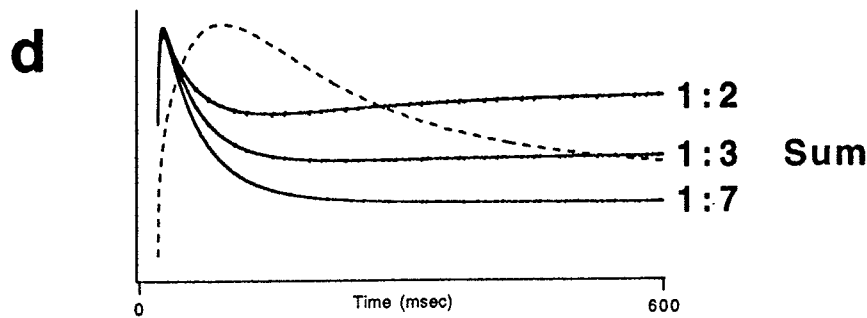
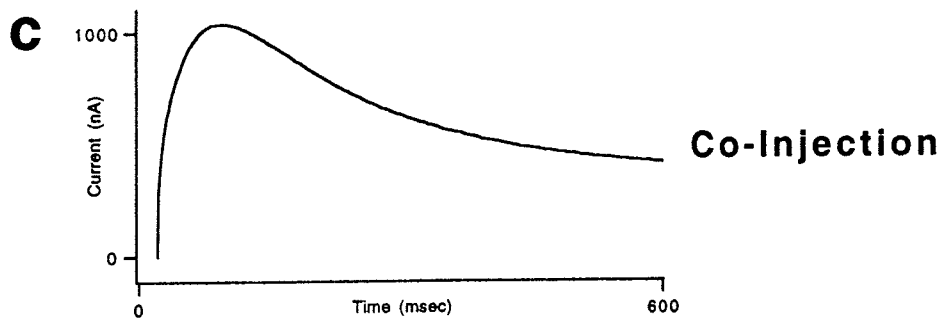
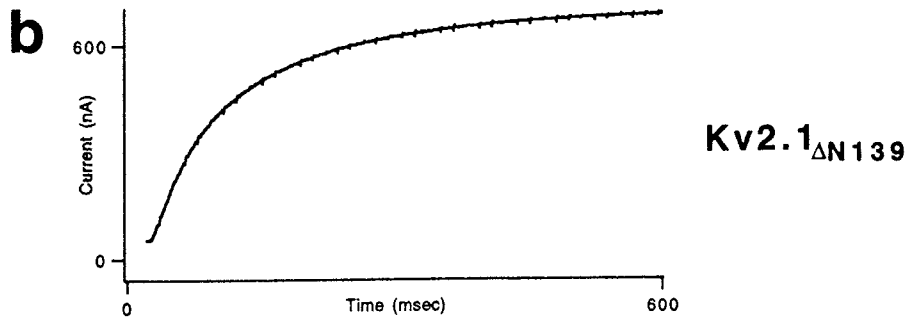
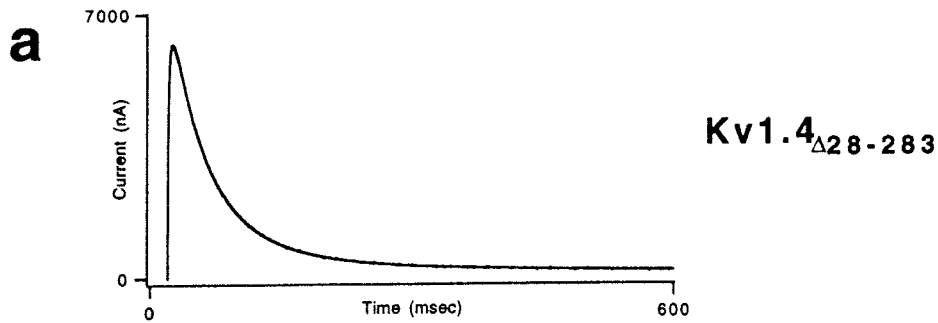
Potentiel d'inversion du courant ?

Seuil d'activation ?

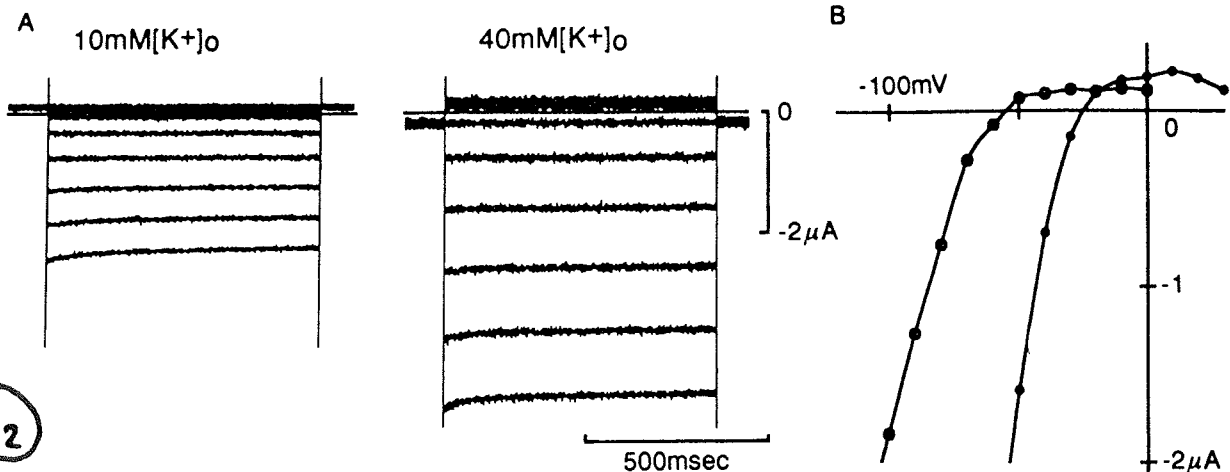
Effets du magnésium sur la rectification ?

Effets de la sous-unité sur la rectification ?

Conclusion ?

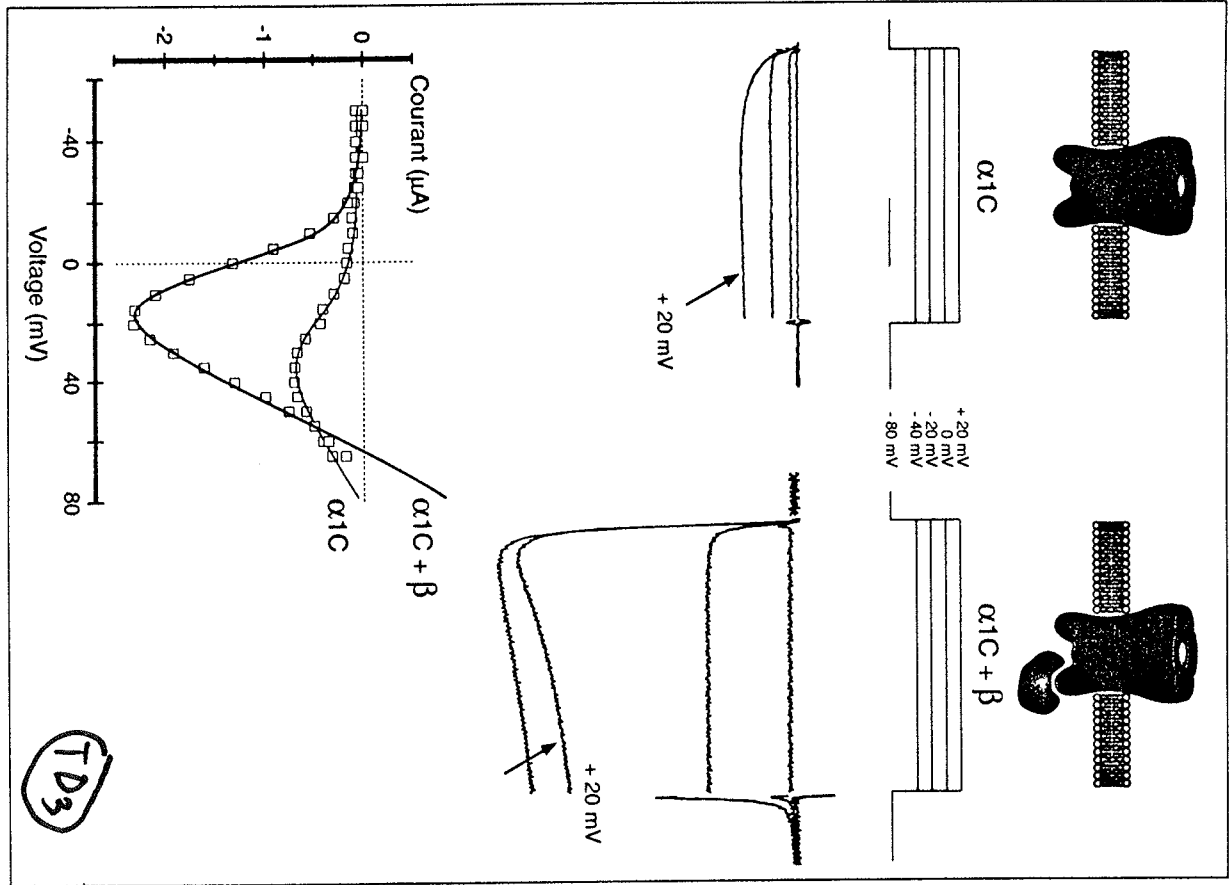


Formation of functional heteromultimeric channels between hKv1.4<sub>Δ28-283</sub> and Kv2.1<sub>ΔN139</sub>. (a) hKv1.4<sub>Δ28-283</sub> current during a voltage step to +40 mV from a holding potential of -100 mV. (b) Kv2.1<sub>ΔN139</sub> current during a voltage step to +40 mV from a holding potential of -50 mV. (c) Heteromultimeric hKv1.4<sub>Δ28-283</sub>-Kv2.1<sub>ΔN139</sub> heteromultimeric current during a voltage step to +40 mV from a holding potential of -50 mV, where none of the homomultimeric hKv1.4<sub>Δ28-283</sub> current would be available for activation (see Fig. 1). (d) Arithmetic sum of the homomultimeric hKv1.4<sub>Δ28-283</sub> current at a holding potential of -100 mV and homomultimeric Kv2.1<sub>ΔN139</sub> current measured at -50 mV (solid line) digitally added in the ratios noted to the right of each trace. The heteromultimeric current from c is shown as a superimposed dashed line for comparison. If hKv1.4<sub>Δ28-283</sub> current were to be available at -50 mV, the inactivation kinetics of the current measured in the coinjection experiments cannot be explained by the arithmetic sum of the independent expression of hKv1.4<sub>Δ28-283</sub> and Kv2.1<sub>ΔN139</sub> channel currents. In all experiments, oocytes were held at the specified holding potential for 1 min prior to the depolarizing step to +40 mV.

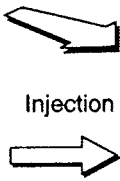
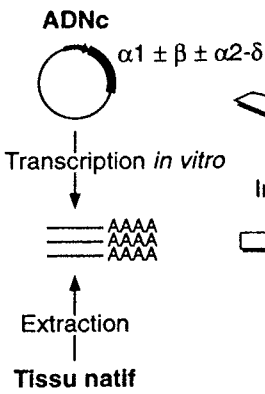


Inward rectifier K<sup>+</sup> currents expressed by injection of the IRK1-cRNA into the oocyte. Fifty nanoliters of the cRNA at a concentration of 5 ng/μl were injected into the stage V oocyte and the expressed currents recorded one day after injection. (A) Families of currents elicited by 650 msec voltage steps from a holding potential of -30 mV at 10 mM (left) and 40 mM (right) [K<sup>+</sup>]<sub>o</sub>. Voltage steps were 0, -30, -50, -60, -70, -80, -90 and -100 mV from top to bottom (left), and 30, 10, -20, -30, -40, -50, -60, -70 and -80 mV (right). (B) The isochronal I-V relationship measured just after the capacitive transient at each voltage step in A (open circles, 10 mM; filled circles, 40 mM [K<sup>+</sup>]<sub>o</sub>). Note that there are "crossover" in the I-V curves and a slight negative slope at potentials positive to E<sub>rev</sub>.

**Modulation des propriétés électrophysiologiques de la sous-unité  $\alpha 1C$  par la sous-unité  $\beta$ .** Courants calciques enregistrés après injection dans des ovocytes de xénope des ADNc codant pour la sous-unité  $\alpha 1$  (classe C) ou  $\alpha 1 + \beta$ . La co-injection de la sous-unité  $\beta$  augmente l'amplitude du courant, accélère les cinétiques d'activation et déplace le pic de la courbe courant-potential d'environ 20 mV dans le sens des hyperpolarisations. Les courants présentés sont enregistrés pour des dépolarisations de -40, -20, 0 et +20 mV (marqués par une flèche) à partir d'un potentiel de membrane de -80 mV.

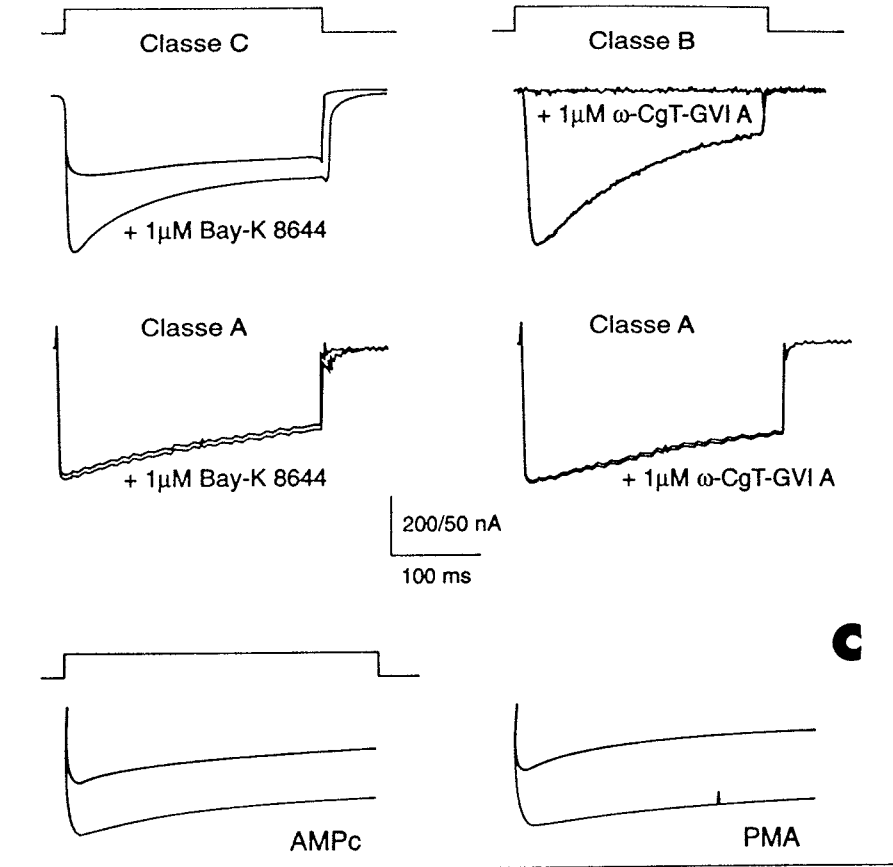
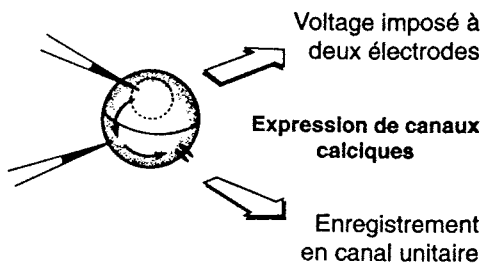


(TD3)



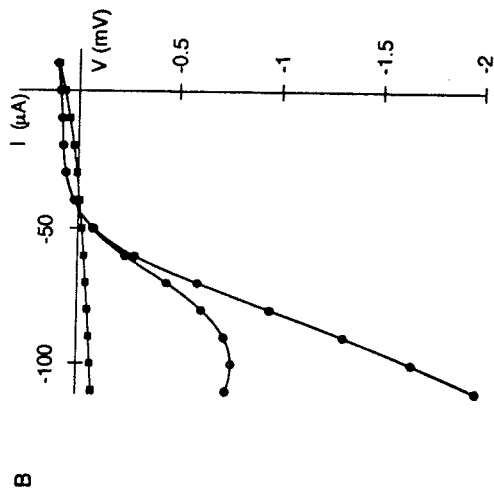
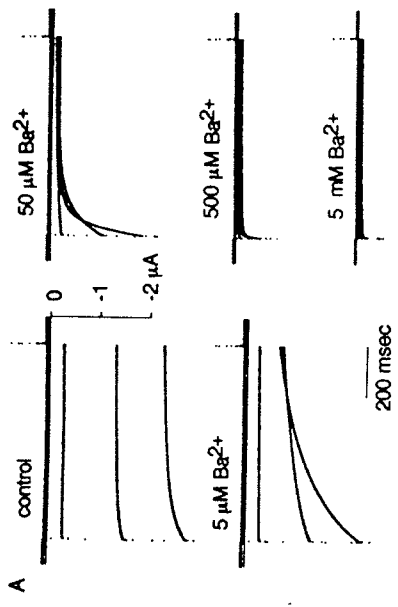
(TD4)

**A**



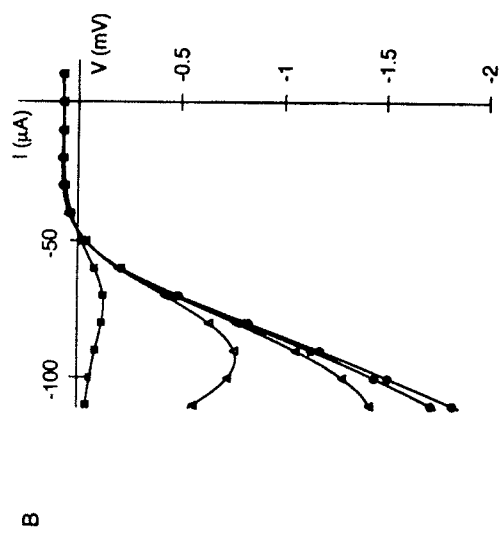
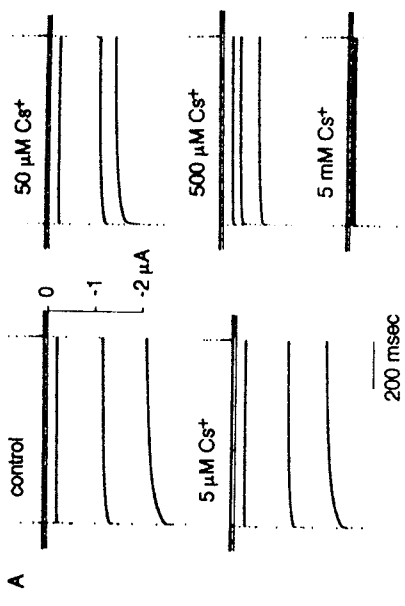
**Pharmacologie comparée des trois types de canaux calciques (classes C, B, et A).** (A). L'injection d'ARN codant pour les différentes sous-unités du canal calcique ( $\alpha 1$ ,  $\beta$  et  $\alpha 2\delta$ ) dans des ovocytes de xénope induit, après deux à trois jours, la formation de canaux présentant des propriétés électrophysiologiques et pharmacologiques similaires à celles mises à jour sur des cellules natives. (B). Le canal de classe C (type L) est activé par une dihydropyridine (Bay-k 8644), le canal de classe B (type N) est bloqué par l' $\omega$ -CgT-GVIA et le canal de classe A (type Q ou P), est insensible à ces deux agents. (C). Après expression dans l'ovocyte de xénope (par injection d'ARN de cœur), les courants calciques de type L restent sensibles aux régulations par la protéine kinase A (stimulation par l'AMPc) ou C (stimulation par l'ester de phorbol, PMA). Les courants sont activés par des dépolarisations de +10 mV à partir d'un potentiel de membrane de -80 mV.

(705)



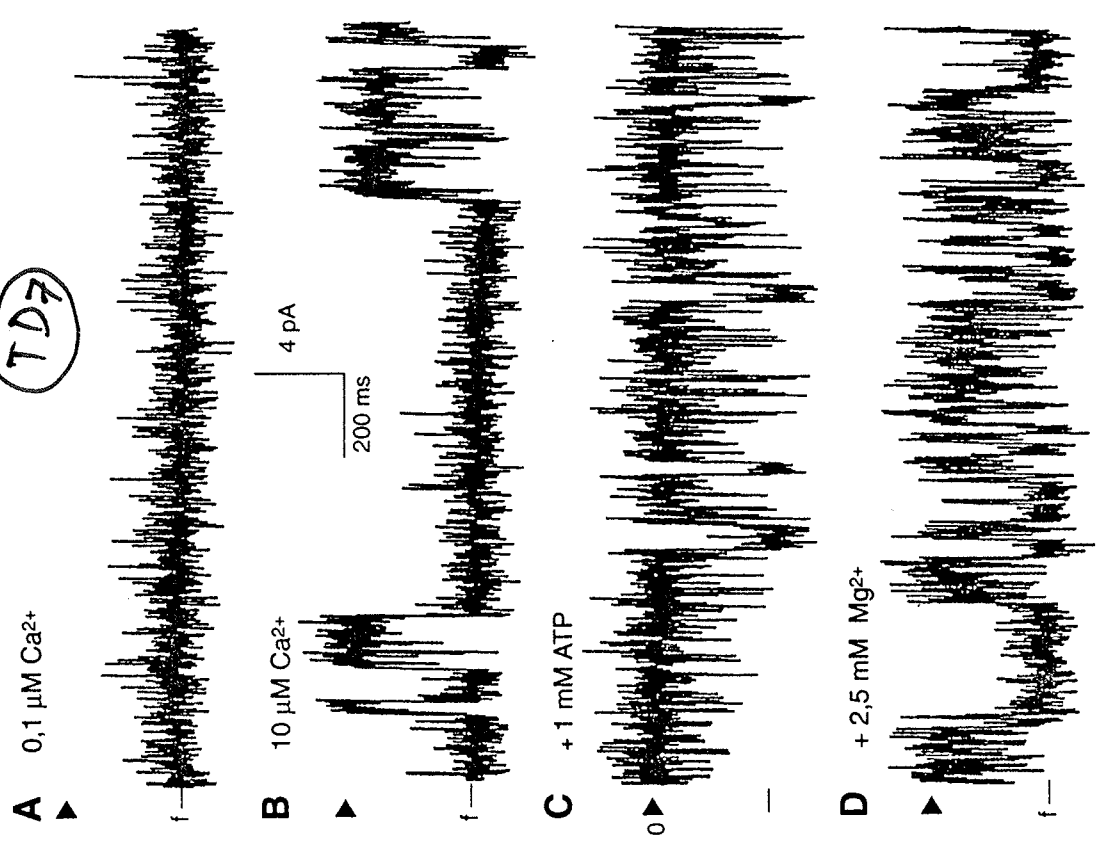
Time- and voltage-dependent block of the expressed current by external  $Ba^{2+}$ . (A) Current traces recorded at different concentrations of  $Ba^{2+}$ . The concentration of  $Ba^{2+}$  applied to the external solution is indicated on the top of each family of traces. Currents were elicited by stepping the membrane to  $-120$ ,  $-90$ ,  $-60$  and  $-30$  mV from a holding potential of  $-20$  mV. Horizontal lines indicate the zero current level.  $[K^+]_o = 20$  mM. (B) Isochronal  $I-V$  relationships obtained from the same experiment shown in A. Filled circles, control; open circles,  $5 \mu M Ba^{2+}$ ; squares,  $50 \mu M Ba^{2+}$ ; triangles,  $500 \mu M Ba^{2+}$ . Current amplitudes were measured at the end of 780 msec step pulses. To show the suppression of outward currents by  $5$  mM of  $Ba^{2+}$  clearly, only relationships obtained at  $5 \mu M$  and  $5$  mM  $Ba^{2+}$  are compared with the control curve.

(706)



Voltage-dependent block of the expressed current by external  $Cs^+$ . (A) Current traces recorded at different concentrations of  $Cs^+$ . The concentration of  $Cs^+$  applied to the external solution is indicated on the top of each family of traces. The currents were elicited by stepping the membrane to  $-120$ ,  $-90$ ,  $-60$ ,  $-30$  and  $0$  mV from a holding potential of  $-20$  mV. Horizontal lines indicate the zero current level.  $[K^+]_o = 20$  mM. (B) Isochronal  $I-V$  relationships obtained from the same experiment shown in A. Filled circles, control; open circles,  $5 \mu M Cs^+$ ; filled triangles,  $50 \mu M Cs^+$ ; open triangles,  $500 \mu M Cs^+$ ; squares,  $5$  mM  $Cs^+$ . Current amplitudes were measured at the end of 780 msec step pulses. Note that the outward currents were not blocked by external  $Cs^+$ .

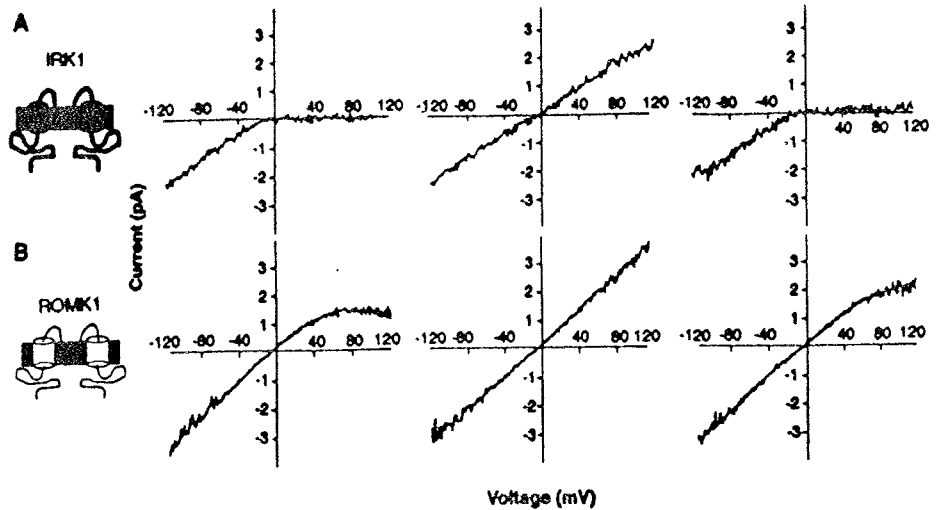
(707)



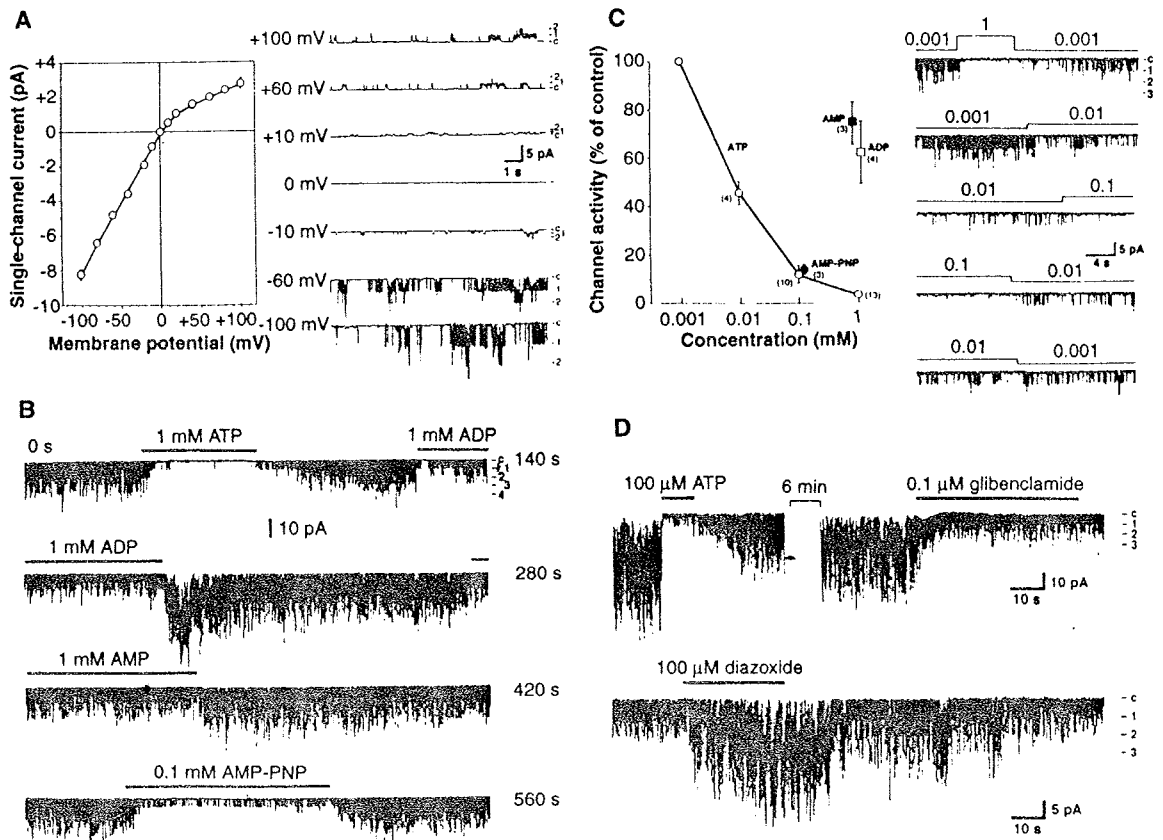
**Enregistrements de l'activité du canal calcique du réticulum sarcoplasmique modulé par différents ligands.** L'activité du canal est enregistrée en condition asymétrique après fusion d'une microvésicule dérivée du réticulum sarcoplasmique cardiaque dans une bicouche lipidique plane (voir figure 3). (A) En présence de  $0,1 \mu M$  de  $Ca^{2+}$  libre du côté cytoplasmique (cis), le canal est pratiquement toujours dans l'état fermé (f). (B) En présence de  $10 \mu M$  de  $Ca^{2+}$ , la probabilité d'ouverture augmente. (C) Après ajout d'ATP ( $1$  mM), le canal est maintenu dans son état ouvert (o). (D) L'addition de  $Mg^{2+}$  ( $2,5$  mM) du côté cytoplasmique (cis) inhibe l'activité de ce canal en modifiant sa cinétique.

TD<sub>8</sub>

Single-channel ramp currents of IRK1 and ROMK1 channels expressed in *Xenopus* oocytes. Single-channel ramp currents from either (A) IRK1 or (B) ROMK1 were recorded from the same patch in the cell-attached configuration (left panels), after patch excision in a Mg<sup>2+</sup>-free solution (middle panels), and after the addition of 0.288 mM Mg<sup>2+</sup> to the internal solution (right panels). Pipette solution: K<sup>+</sup>-Ringer; bath solution: iso-K<sup>+</sup>. The zero Mg<sup>2+</sup> ramp current in IRK1 came from a fully open channel. At depolarized potentials, the probability of opening was reduced as a consequence of the intrinsic gating that is known to occur in these channels (13-17). The  $g_{K^+}$  values are expressed as slope conductance measured between -160 and -20 mV in cell-attached patches by the use of voltage steps. In our solution conditions,  $g_{K^+}$  values for IRK1 and ROMK1 are consistent with those already reported (1, 2).



Electrophysiological recording from COS-1 cells coexpressing mBIR and haSUR. (A) Left, inwardly rectifying property of BIR channel ( $n = 6$ ). Right, representative traces from single-channel recordings in the inside-out mode at various holding potentials. (B) Effects of ATP and its analogs on channel activity. Representative traces of the excised inside-out patches are shown. Channel activity was inhibited by 1 mM dipotassium ATP and by 0.1 mM AMP-PNP, a nonhydrolyzable ATP analog. ADP (1 mM) and AMP (1 mM) in the presence of 1  $\mu$ M dipotassium ATP slightly suppressed channel activity. (C) Left, dose-dependent effect of ATP and its analogs on channel activity. Channel activity was calculated by integrating current flow during the channel openings and dividing the integral by the total sampling time (generally 20 to 40 s). The channel activity at each concentration of the agents is expressed as a percent of the control (at 1  $\mu$ M dipotassium ATP). Right, representative traces at the indicated ATP concentrations (mM). The numbers in parentheses give the number of patches analyzed for each concentration. [See (22, 23) for details of the transfection.] (D) Effects of glibenclamide (0.1



$\mu$ M) and diazoxide (100  $\mu$ M) on channel activity. Representative traces are shown. In (B) through (D), horizontal bars indicate application periods of the agents. Membrane potential was held at -60 mV, unless otherwise noted. Values are means  $\pm$  SEM. The numbers to the right of the traces indicate the numbers of open channels.

TD<sub>9</sub>

TD<sub>10</sub>