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A Mutation that Increases a Novel Calcium-Activated Potassium Conductance of *Paramecium Tetraurelia*

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Summary. Under two-electrode voltage clamp, a mutant of *P. tetraurelia, restless* (*rst*/*rst*), showed a large increase in induced current and an outward tail current when compared to the wild-type cell for hyperpolarizing voltage steps. An increase in the induced and tail currents is also observed for depolarizing voltage steps. The larger current during voltage steps and tail in the mutant were eliminated by the use of CsCl-filled electrodes and tetraethylammonium ion (TEA^+) in the bath solution, characterizing the lesion as affecting a K^+ conductance. Ionophoretic injection of ethylene glycol bis-(beta-aminoethyl ether) n,n,n′,n-tetraacetic acid (EGTA) to buffer internal Ca^{2+} concentration reduced the increased K^+ current and tail of the *restless* cell, indicating Ca^{2+} activation of the K^+ current. Time course and amplitude of remaining currents after blockage of K^+ conductances with Cs^+ and TEA^+ were similar in wild-type and *restless* cells suggesting no *restless* defect in entry of calcium. The Ca^{2+}-activated sodium current was similar in the mutant to that in wild type arguing against a defect in calcium regulation activating the K^+ channel in the *restless* cell. We conclude that the *restless* mutation alters a Ca^{2+}-activated potassium conductance other than the one previously described. The multiplicity of Ca^{2+}-activated potassium conductances in *Paramecium* is discussed.

Key Words mutation · *Paramecium* · calcium-activated potassium conductance · neurogenetics

Introduction

Calcium-activated potassium channels have been shown to exist in a wide range of organisms and cell types. They are involved with secretion, oscillatory membrane potentials, negative feedback on excitatory responses, and maintaining membrane potential (*see* Latorre & Miller, 1983; Schwarz & Passow, 1983; Petersen & Maruyama, 1984, for reviews). Calcium-activated potassium currents have been reported as altered by mutation in three instances. In *Drosophila*, the *slopoke* mutation decreases I_C (Elkins, Ganetzky, & Wu, *in preparation*). In *Paramecium*, the *pantaphobiac A* mutant greatly decreases a Ca^{2+}-activated potassium current participating in regeneration of the resting membrane potential during the action potential (Saimi et al., 1983). The *Tea A* (*tetraethylammonium insensitive A*) mutation, also in *Paramecium*, increases the current through this latter conductance (Hennessey & Kung, *in preparation*).

A new mutant of *P. tetraurelia* has been isolated with behavioral and other phenotypes indicating a defect in the control of potassium permeability (Richard & Saimi, 1984; Richard et al., 1985). Unlike that of wildtype (Naitoh & Eckert, 1968), the membrane voltage of *restless* cells depends on external potassium concentration as a potassium electrode. The membrane potential of wild-type cells hyperpolarizes to an apparent limit of approximately −45 mV as external potassium is lowered, while *restless* hyperpolarizes −66 mV per decade decrease in external potassium concentration. The *restless* cells are also found to die within hours in dilute potassium solutions that wild type can survive for days. Genetic crosses of *restless* to other mutants affecting potassium regulation demonstrated complementation, independent segregation, and expression of both phenotypes (Richard et al., 1985; Richard, *unpublished*). Behavioral responses to most stimuli are attenuated in this mutant, which is interpreted as being related to an increased potassium permeability rather than a direct effect on excitability (Richard et al., 1985).

In light of the many functions ascribed to Ca^{2+}-activated potassium channels and the indirect effects on excitability of the *restless* mutation, we tested the *restless* mutant for such a conductance. We found that the *restless* mutation affected a novel Ca^{2+}-activated potassium channel of *Paramecium tetraurelia*.

Materials and Methods

Strains and Culture

Wild type refers to *trichocyst nondischarge nd6*/*nd6* (Sonneborn, 1975; a gift of K. Aufderheide), a mutant of stock 51s (kappa

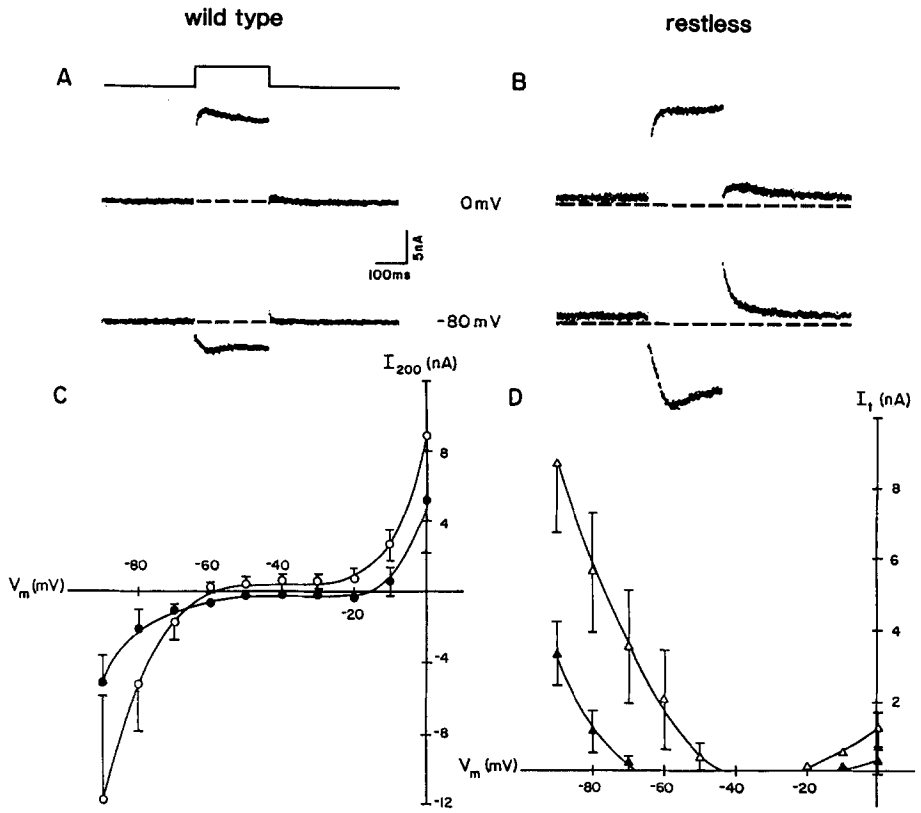

Fig. 1. Membrane properties of wild type and *restless* under voltage clamp. Membrane currents induced by 250-msec steps from the holding level of −40 to 0 or −80 mV are shown for wild type (*A*) and *restless* (*B*). The dashed lines indicate the zero-current level. Total current is shown without leakage current subtraction. Bathed in Solution A; recorded with 2 M KCl electrodes. (*C*) The *I-V* relationships of the total membrane current at 200 msec under these conditions (bars show standard deviation) are shown for wild type (filled circles) and *restless* (open circles). The leakage current has been subtracted ($n = 6$ except for *restless* at −90 mV where $n = 5$; slope resistance for wild type = 35 ± 10 MΩ; *restless* = 30 ± 10 MΩ). (*D*) Maximum amplitude of tail currents upon return to the holding level after 250-msec voltage steps (V_m) for wild type (filled triangles) and *restless* (open triangles). Amplitude is measured from the holding current level; these are the same cells as in (*C*)

free) of *P. tetraurelia* with no observed defect in behavior or electrophysiology. The mutant strain was *restless*, stock d4-647 (Richard et al., 1985), with the nondischarge background, *rst/rst nd6/nd6*. Well-fed cells (Saimi et al., 1983) were chosen for electrophysiological studies.

Electrophysiology

Cell recordings using two-electrode voltage clamp were done as described previously (Hinrichsen & Saimi, 1984). Leakage corrections were estimated by measuring under voltage clamp the slope resistance of the membrane using small voltage steps from the holding voltage of −40 mV. The extrapolated current for large steps was subtracted from the measured values. The electrodes were filled with 2 M KCl or 2 M CsCl, depending on the experiment. The electrode resistance was between 15 and 25 MΩ. Cells were held at −40 mV, near the resting level in Solution A. The experiments were done at room temperature, 21 ± 2°C.

The solutions had compositions in addition to 1 mM HEPES (N-2-hydrozyethylpiperazine-N′-2-ethanesulfonic acid), 0.01 mM EDTA (ethylenediaminetetraacetic acid), and 1 mM $CaCl_2$ as follows (in mM): Solution A: 4 K^+; Solution B: 10 TEA^+, 4 K^+; Solution C: 10 TEA^+, 8 Na^+. All salts were reagent grade and either Cl^- or OH^- form; HCl was used to adjust pH to 7.1 to 7.2.

EGTA Ionophoresis

Current injection and voltage electrodes for clamping the voltage were filled with 2 M KCl. A third electrode containing EGTA was also used. The EGTA ionophoresis electrode contained 100 mM EGTA and 1 mM HEPES. KOH (approximately 200 mM) was added to adjust the pH to 7.1. The resistances of the ionophoresis electrodes were 100 to 200 MΩ. EGTA injection was done for a total of 3 min at −5 nA under voltage clamp while maintaining the holding voltage between −40 to −45 mV.

Results

Membrane currents of wild-type and *restless* cells were recorded under voltage clamp using KCl-filled electrodes and external 4 mM K^+ (solution A). The

current traces for wild type (Fig. 1*A*) and *restless* (Fig. 1*B*) demonstrated differences in membrane current during the voltage step, tail current amplitude, and tail current kinetics. The increased current in the *restless* cell was compared to wild type in Fig. 1*C* as measured 200 msec after the start of the voltage step for different voltage steps. Mean currents were significantly different for all voltage levels (two-tailed *t*-test, $P < 0.10$) except at the crossover point of −70 mV. At the holding potential of −40 mV, the mutant had a 0.8 nA more positive holding current. The parallel *I-V* curves around the holding voltage suggested this difference was due to the steady-state properties of the membrane at the holding voltage rather than to activation of a current. When this difference in holding level was adjusted, there was a significantly greater activation of current in *restless* for voltage steps less than or equal to −70 mV and greater than or equal to −10 mV (two-tailed *t*-test, $P < 0.10$). The causes of induced current by voltage steps of opposite polarities are discussed below.

The *restless* difference from wild type was more apparent upon examination of the tail currents upon return to the holding level at the end of these voltage steps. The tail currents (Fig. 1*D*) were measured as the maximum amplitude of the outward current measured upon return to the holding level of −40 mV. Amplitude of this outward tail current was always much larger in the *restless* mutant. In those cases where a wild-type current was observed ($\leqq$ −70 mV, $\geqq$ −10 mV), the difference between wild type and *restless* was highly significant (two-tailed *t*-test, $P < 0.01$). For the voltage steps to −60 and −50 mV where a wild-type tail current was not observed, the *restless* tail current was statistically different from 0 nA (*t*-test, $P < 0.10$). The composition of this tail current in the *restless* mutant was apparently dominated by the conductance affected by the *restless* lesion. The tail current kinetics were also different. Wild-type cells showed only a single exponential for all voltage steps. At the end of steps to −90 mV the $t_{1/2}$ equaled 9 ± 3 sec ($n = 6$). The *restless* cells exhibited more complex kinetics as the amplitude of the tail current increased. At the end of steps to −90 mV, *restless* cells had at least three exponentials with $t_{1/2} = 23 \pm 6$ sec ($n = 5$).

Since, in the 4 mM K^+ solution, the tail current at the end of the voltage steps was always outward, the affected current was likely to be carried by K^+ or Cl^-. As chloride has been shown not to be significantly permeant across the wild-type cell membrane (Naitoh et al., 1972), we tested whether the current enhanced by the mutation was carried by K^+. External 10 mM TEA^+ and internal Cs^+ through diffusion from electrodes have been shown to eliminate more than 90% of membrane K^+ currents

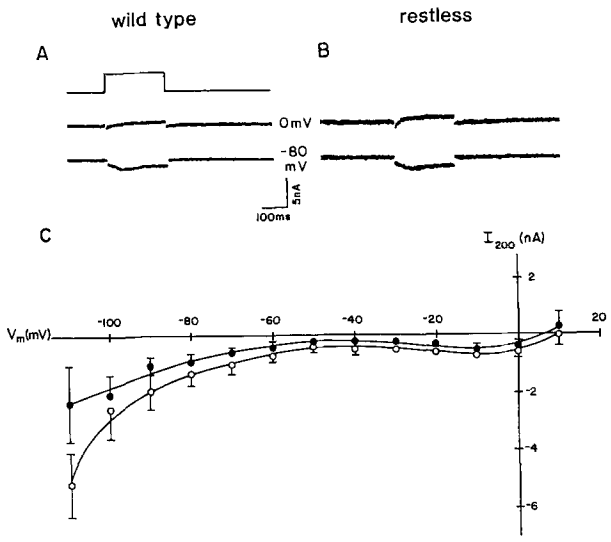

Fig. 2. Suppression of K^+ currents with internal Cs^+ and external TEA^+ in wild type and *restless*. Membrane currents induced by 250-msec steps from the holding voltage of −40 to 0 or −80 mV are shown for wild type (*A*) and *restless* (*B*). The leakage current is not corrected. Bathed in Solution B; recorded with 2 M CsCl electrodes. (*C*) The *I-V* relationships of the total membrane currents at 200 msec under these conditions (bars show standard deviation) are shown for wild type (filled circles) and *restless* (open circles). The leakage current has been subtracted. ($N = 6$ except for −100 and −110 mV where $n = 3$; slope resistance for wild type = 70 ± 10 MΩ; *restless* = 45 ± 10 MΩ)

(Hinrichsen & Saimi, 1984). Blocking potassium conductances with this treatment made the mutant membrane nearly identical to wild type (Fig. 2). The holding current of wild type did not change from −0.2 nA, while that of *restless* decreased from 0.6 to −0.4 nA (Figs. 1*C*, 2*C*). If this difference in holding current is again subtracted on the assumption that it reflects steady-state properties of the membrane, there is a significant difference in current amplitudes only at −110 and −90 mV (two-tailed *t*-test, $P = 0.06$ for each; at −100 mV, $P = 0.77$). The outward tail currents of both wild type (Fig. 2*A*) and *restless* (Fig. 2*B*) were effectively eliminated by this treatment. Since Cs^+ and TEA^+ are potent potassium channel inhibitors, the mutation affected a K^+ current. This result indicates that the parallel effects of the *restless* mutation on amplitudes of current during the voltage step and the tail current were due to effects on potassium currents only.

EGTA Ionophoresis

A Ca^{2+} current is suggested to activate by hyperpolarization, and the Ca^{2+}-activated Na^+ current upon hyperpolarization has been recorded (Y. Saimi, *submitted*). We tested the possibility of calcium ac-

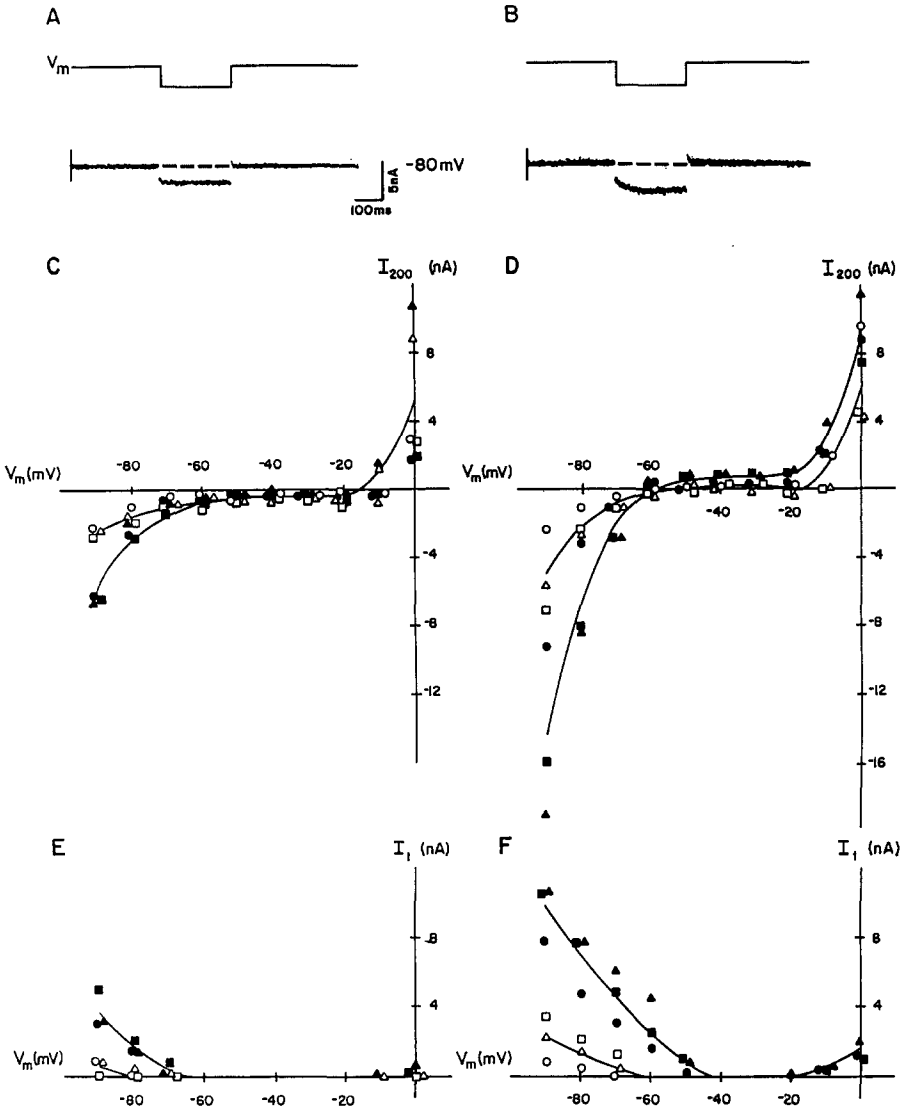

Fig. 3. Ionophoretic injection of EGTA reduces membrane current of wild type and the *restless* mutant. Current induced by a 250-msec step to −80 mV from the holding level of −40 mV after 3 min of EGTA ionophoresis are shown for wild type (*A*) and *restless* (*B*). The dashed lines indicate zero current level; the traces are not corrected for leakage current. Figure 1 (*A–B*) illustrates the same cells before EGTA injection. The individual before (filled symbols) and after (open symbols) *I-V* relationships of total membrane currents at 200 msec are shown for three wild-type (*C*) and three *restless* (*D*) cells. Leakage current has been subtracted (slope resistance for wild type: before = 30 ± 5 MΩ, after = 35 ± 5 MΩ; *restless*: before = 30 ± 5 MΩ, after = 35 ± 20 MΩ). Maximum amplitude of tail currents upon return to the holding voltage of 250-msec voltage steps are shown for wild type (*E*) and *restless* (*F*). Each symbol corresponds to the cells in (*C*) and (*D*). Amplitude was measured from the holding current level. A smooth curve has been drawn through the mean currents for wild type and *restless* (*C–F*)

tivation of the K^+ current enhanced in the mutant with ionophoretic injection of EGTA to reduce the intracellular free calcium concentration. Injection of EGTA was expected to decrease the induction of Ca^{2+}-activated currents and alter tail kinetics of such currents by increasing the rate of removal of free calcium that enters the cell. A decrease in holding current similar to that observed upon potassium channel blockage with Cs^+ and TEA^+ (Fig. 2*C*) would also be expected in the mutant cell.

The membrane properties of cells were first measured under the conditions used in Fig. 1. After insertion of an electrode containing 100 mM EGTA, membrane properties were tested to insure there was no damage caused by the additional electrode. EGTA was injected for 3 min and membrane currents again measured. The current traces for a wild-type (Figs. 1*A*, 3*A*) and a *restless* cell (Figs. 1*B*, 3*B*) are shown before and after EGTA injection. Current amplitudes at 200 msec for voltage steps from the holding level are shown for wild type (Fig. 3*C*) and *restless* (Fig. 3*D*). The maximum outward tail amplitudes upon return to the holding voltage at 250 msec also decreased for wild type (Fig. 3*E*) and *restless* (Fig. 3*F*). Paired before-and-after comparison of wild type showed significant reductions in the membrane current during voltage steps and tails for −80 and −90 mV (*t*-test, $P < 0.10$). The *restless* cells had significant decreases within cells for membrane current induced by voltage steps in the range of −90 to −70 mV (*t*-test, $P < 0.10$), and highly significant decreases in tail currents for this range and for −10 and 0 mV (*t*-test, $P < 0.05$). Wild-type tails after injection were too small and rapid to be measured with the resolution of these experiments. The decay of the *restless* tails where measurable

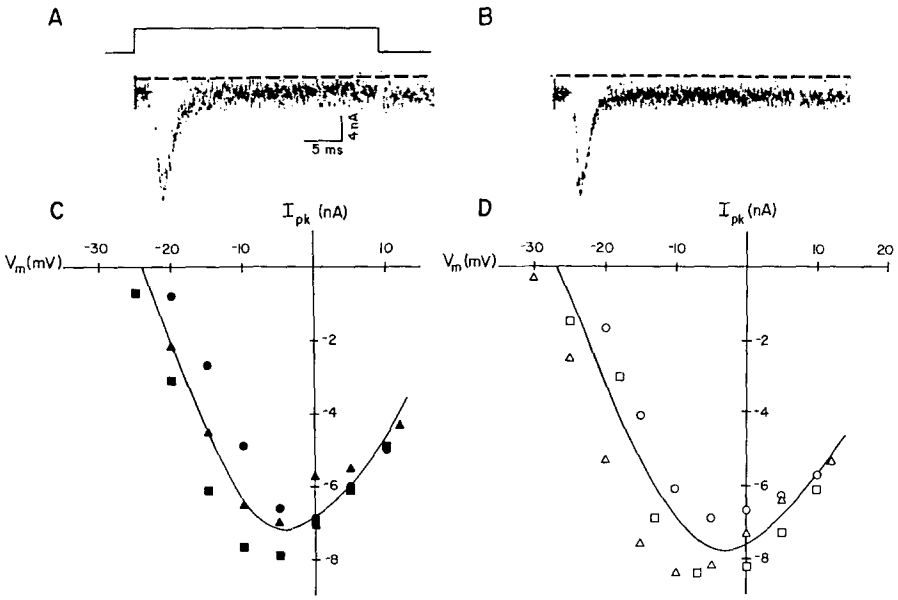

Fig. 4. The depolarization-dependent calcium current measured in wild-type and *restless* cells. Current traces under voltage clamp of maximum calcium currents induced by 25-msec depolarizing voltage steps (to −5 mV) from the holding level of −40 mV for wild type (*A*) and *restless* (*B*). Conditions are the same as in Fig. 2. The dashed line indicates zero current level; leakage current has not been corrected. The peak inward currents induced by a series of voltage steps to different levels are shown for three wild-type (*C*) and three *restless* (*D*) cells. Each symbol represents the measurements from one cell. Leakage currents have been subtracted (slope resistance for wild type = 70 ± 10 MΩ, *restless* = 60 ± 15 MΩ). Curves are drawn through mean current values

was simplified to an apparent single exponential with a $t_{1/2}$ of approximately 7 msec. The change in mean holding current of the *restless* cell was highly significant (two-tailed *t*-test, $P < 0.05$). These results indicate that increased conductance in the *restless* cell was calcium activated.

The *restless* cells were not identical to wild-type cells after EGTA treatment. EGTA injection was not done to higher concentrations due to death of *restless* cells with larger ionophoretic currents or longer injection times.

Calcium Entry and Regulation

An increase in conductance through a Ca^{2+}-activated K^+ channel could result from a direct alteration of the conductance itself or an increase of internal free calcium. Although the results from blockage of potassium conductances (Fig. 2) suggested no alteration of calcium currents, we investigated two biological indicators of the regulation of internal calcium available in *Paramecium*.

The depolarization-dependent calcium current is inactivated by elevated internal calcium. This has been shown by double-pulse experiments and EGTA injection (Brehm et al., 1980). The peak calcium current amplitude and its time course were identical for wild type (Fig. 4*A*) and *restless* (Fig. 4*B*) when potassium currents were blocked by Cs^+ and TEA^+ as in Fig. 2. The peak current amplitude as plotted against voltage steps was unaltered (two-tailed *t*-test, $P > 0.10$) by the *restless* mutation (Fig. 4*D*) as compared to wild type (Fig. 4*C*) except for steps to 10 mV. The time to maximum inward current (*data not shown*) was not significantly different at all voltages tested (two-tailed *t*-test, $P > 0.10$, at 10 mV: $P = 0.56$) between wild type and *restless*, further suggesting no alteration in internal calcium regulation.

The Ca^{2+}-activated sodium current in *Paramecium* can be induced by calcium entry due to depolarization (Saimi & Kung, 1980) as well as hyperpolarization (Saimi, *submitted*). The sodium channel current was measured under conditions similar to those used to suppress potassium conductances (Fig. 2) with the addition of 8 mM Na^+ externally (solution C). An increase in calcium entry or of free internal calcium would be expected to increase the conductance through this channel induced by voltage steps, and the magnitude of an inward sodium tail current upon return to the holding level. Current traces from wild type (Fig. 5*A*) and *restless* (Fig. 5*B*) showed neither of these effects. Measurements of current amplitude 200 msec after initiation of voltage steps from the holding voltage were ob-

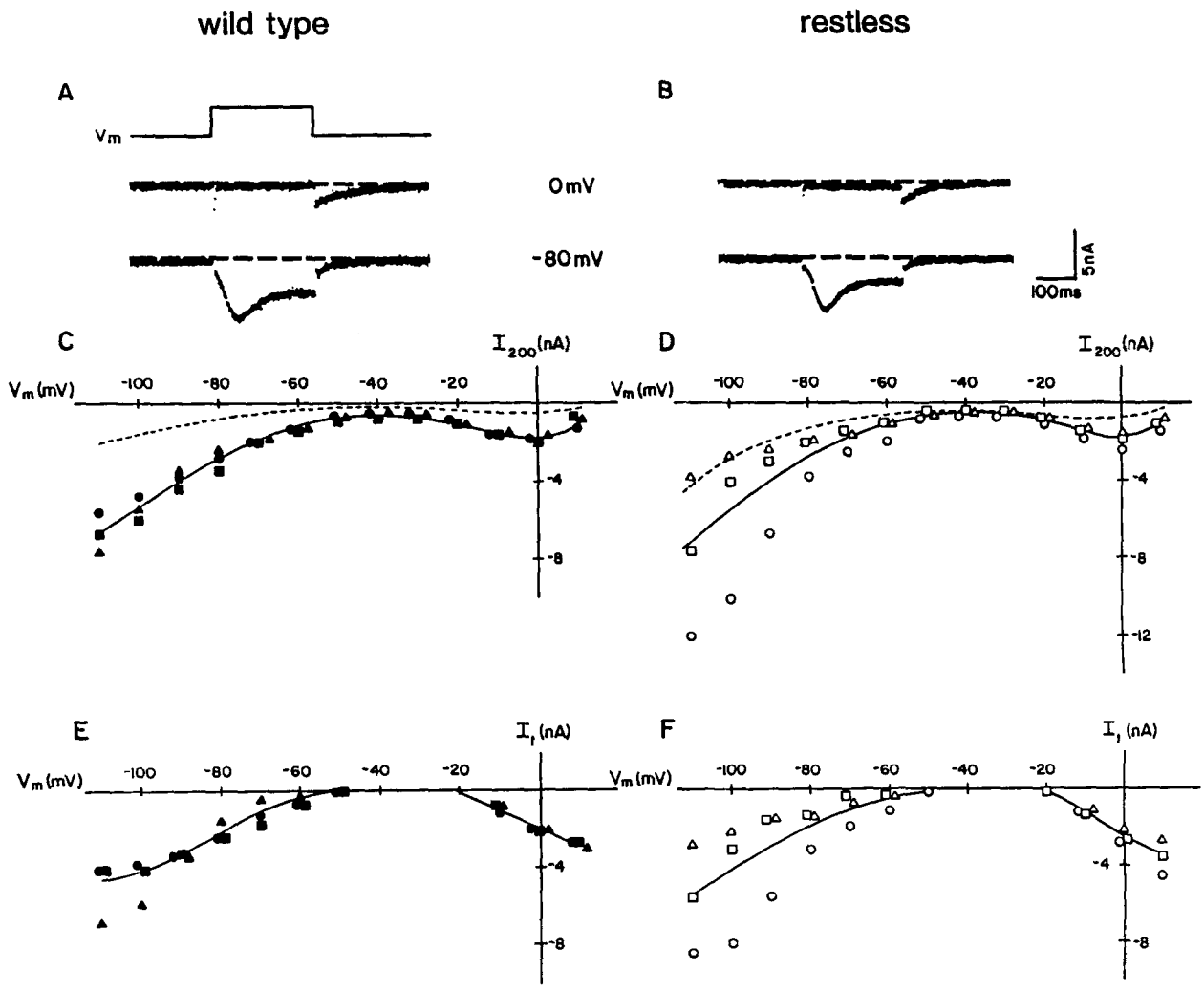

Fig. 5. Membrane currents in a sodium solution show that the Na^+ current is not significantly affected by the *restless* mutation. CsCl-electrodes and external TEA^+ were used to eliminate potassium currents (Solution C, 2 M CsCl electrodes). Membrane currents induced by 250-msec steps from the holding voltage of −40 mV to the voltages indicated beside each trace are shown from wild type (*A*) and *restless* (*B*). The dashed line indicates the zero-current level. Leakage current is not corrected. The *I-V* relationships of the total membrane currents at 200 msec induced by voltage steps from the holding voltage of −40 mV are shown for wild-type (*C*) and *restless* cells (*D*). Leakage currents have been subtracted (slope resistance for wild type = 35 ± 5 MΩ, *restless* = 45 ± 20 MΩ). Data from three cells each are shown with a curve drawn through mean values. A dotted line has been drawn in each case from the data in Fig. 2*C* for comparison. Maximum amplitude of tail currents induced by the return to the holding voltage from 250-msec voltage steps is shown for wild type (*E*) and *restless* (*F*). Amplitude was measured from the holding current level

tained for wild type (Fig. 5*C*) and *restless* (Fig. 5*D*). Although there was a difference in variability, a smooth curve through mean values is nearly identical for wild type and *restless*. The probability of the mean values being different is not significant (two-tailed *t*-test, $P > 0.10$; at −110 mV, $p = 0.75$). The dashed lines (Fig. 5*C*, *D*) indicate the basal current observed in Fig. 2 (in the absence of Na^+ and K^+ currents) for comparison. The maximal amplitude of the inward tail currents present upon return to the holding voltage at 250 msec also showed greater variability in the *restless* cells (Fig. 5*F*) than wild type (Fig. 5*E*), but similar mean values (two-tailed *t*-test, $P > 0.10$; at −110 mV, $P = 0.60$).

Discussion

The identification that the *restless* mutation affects a potassium conductance is supported by several lines of evidence. 1) The membrane potential of the mutant shows a near-Nernstian dependence on external potassium concentration below 8 mM in the presence of 1 mM $CaCl_2$ (Richard et al., 1985). 2)

The increased tail currents associated with the currents induced by voltage steps are outward (solution A, Fig. 1) and eliminated by the use of external TEA^+ and internal Cs^+. 3) Other membrane properties of *restless* are similar to wild type when these latter ions are present (Figs. 2, 4, 5). 4) Effluxes of ^{86}Rb (Richard, 1985), an analog for potassium (Browning, 1976; Hansma, 1981), were faster for *restless* than wild type in solution A with 0.8 mM RbCl added.

Calcium Activation

The potassium conductance is calcium activated as indicated by the increased current being induced by both hyperpolarizing and depolarizing voltage steps (Fig. 1) and supported by the ionophoretic injection of EGTA (Fig. 3). Calcium entry is well characterized for depolarizing voltage steps (for a review *see* Kung & Saimi, 1985), and recently demonstrated for hyperpolarizing voltage steps (Saimi, *in preparation*; Hennessey, *unpublished*). These two calcium currents produce a similar activation of the sodium current of *Paramecium* (Fig. 5, Saimi, *in preparation*). Bipolar activation of the *restless* channel directly by voltage would be highly unusual and is not consistent with the effect of EGTA injection. Note that the current removed by EGTA in *restless* cells at −110 mV is greater than the total current in wild-type cells before the treatment. The reduction of membrane currents during voltage steps, tail currents, and simplification of tail kinetics are all consistent with calcium activation. Inhibition of Ca^{2+}-activated K^+ conductances with EGTA has been reported in *Paramecium* (Satow & Kung, 1980) and other organisms (for review *see* Meech, 1978).

The *restless* defect appears not to be a secondary consequence of an increase in a calcium conductance or internal calcium levels. Calcium entry has been shown to reduce and delay the depolarization dependent calcium current (Brehm & Eckert, 1978). A difference between wild type and *restless* in magnitude (Fig. 4) is only evident at the extreme voltage of +10 mV, while time course and activation is normal at lower voltage levels. We feel this difference is too minor to explain the *restless* phenotype and is possibly due to small errors in measurement of slope membrane resistance. Testing the Ca^{2+}-activated sodium current (Fig. 5) might examine calcium entry for voltage steps of both polarities from the resting level. No aspect of the sodium current was clearly increased by the *restless* defect.

The *restless* cells consistently exhibited greater variation of induced currents than wild-type cells. This may be due to their being more difficult to voltage clamp or maintain during experiments without physical damage, unacceptably low membrane resistance, or decreases in the depolarization-dependent calcium current.

Other mutations affecting calcium regulation in *Paramecium* do not have the behavioral or survival phenotypes of *restless*. A mutation increasing current through the depolarization-dependent calcium channel, *Dancer* (Hinrichsen & Saimi, 1984), and a new mutation of *P. tetraurelia, k^+-shy A*, suggested to have a slowed reduction of free internal calcium following stimulation (Evans, Hennessey, & Nelson, *in preparation*) do not die in dilute potassium solutions as does *restless* (Richard et al., 1985; Richard, *unpublished*).

Unlike the depolarization related currents, the nature of the various currents in the hyperpolarizing region have not been fully explored. This present work is part of an effort to understand the currents in this region. Hyperpolarization-activated potassium conductances have been described in *Paramecium* (Satow & Kung, 1977; Oertel et al., 1978). These currents have similar properties as that affected in *restless* cells. Experiments investigating the pharmacology and time course of the *restless* current are being conducted to strengthen the identification. A hyperpolarization-dependent calcium current is being characterized (Saimi, *submitted*; Hennessey, *unpublished*).

Behavior and Electrophysiology

Wild-type cells can be induced to exhibit transient to extended periods of backward swimming by a variety of ions and chemicals. The *restless* cells show no reversals or shorter ones (Richard et al., 1985), depending on the stimulus, which can be explained by the electrophysiological results presented here. The entry of calcium during an action potential serves to both depolarize the membrane and induce ciliary reversal. In the *restless* cell an increased potassium conductance short circuits the action potential, and as external potassium concentration is lowered, hyperpolarizes the membrane (Richard et al., 1985) adding another barrier to action potentials.

However, an increase in conductance of a potassium channel is not sufficient to explain the hyperpolarization phenomenom. That wild-type cells do not hyperpolarize as external potassium is lowered implies active regulation of potassium conductances or increases in inward currents. The inward currents in *restless* are normal as indicated by the data in Figs. 2, 4 and 5. The lesion is therefore more than just an increase in a potassium conductance; the *restless* current appears to have lost an element

of control. The open and unregulated potassium channel in *restless* explains the Nerstian behavior of its membrane voltage to changes in external potassium. As this conductance does not appear to be directly involved in excitability in the wild-type cell, and the defect of the channel has such a great effect on membrane potential, we suggest this current may be a major determinant of resting membrane potential.

Calcium-Activated Potassium Channels

A Ca^{2+}-activated potassium current has been well characterized in *Paramecium* (Satow & Kung, 1980; Saimi et al., 1983). The *restless* mutation demonstrates the existence of a second Ca^{2+}-activated potassium conductance in this organism. A mutant increasing the previously described Ca^{2+}-activated potassium current, *Tea A*, does so only for depolarizing voltage steps (Hennessey & Kung, *in preparation*) while *restless* also increases current in response to hyperpolarizing steps. The *restless* phenotype was expressed in the double mutant with *pantaphobiac A* (Richard et al., 1985), a mutation that eliminates most of the previously described Ca^{2+}-activated potassium conductance (Saimi et al., 1983). Both these genes are phenotypically and genetically distinct from the *restless* mutation (Richard et al., 1985). TEA^+ is a weak blocker of the Ca^{2+}-activated potassium current (Saimi, *unpublished*), but was able to induce avoiding reactions in *restless* cells similar to those of wild type (Richard et al., 1985) by apparently blocking the short-circuiting potassium current during the action potential. *Tea A* cells do not respond to TEA^+ ions in the concentrations used for behavioral testing (Satow & Kung, 1976).

Two Ca^{2+}-activated potassium channels have been reported in nerve (Pennefather et al., 1985) and muscle (Romey & Lazdunski, 1984). In both types of cells, one current was described as helping to regain the resting membrane potential after depolarization. This is the same function ascribed to the previously described Ca^{2+}-activated potassium conductance in *Paramecium* (Saimi et al., 1983). The second current was described as inducing hyperpolarized levels of the cell's membrane potential. This novel Ca^{2+}-activated potassium conductance of *Paramecium* may serve an analogous function in control of membrane potential.

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