The Neuropeptide FMRF-amide Decreases Both the Ca^{2+} Conductance and a Cyclic 3',5'-Adenosine Monophosphate-dependent K^{+} Conductance in Identified Molluscan Neurons¹

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Abstract

The molluscan neuropeptide FMRF-amide (10 to 50 μM) decreases the duration of the Ca^{2+} -dependent action potential recorded in the cell body of identified neurons of the snail *Helix aspersa* (cells D3 and E2). In these neurons, FMRF-amide evokes a decrease of the Ca^{2+} current resulting from a decrease in Ca^{2+} conductance. In another single neuron, cell E11, FMRF-amide, besides evoking a decrease of the Ca^{2+} conductance, induces a decrease of the S-current (Klein, M., J. S. Camardo, and E. R. Kandel (1982) *Proc. Natl. Acad. Sci. U. S. A.* 79: 5713–5717), a K^{+} current controlled by cyclic AMP. However, in this E11 cell, FMRF-amide also evokes a decrease of the amplitude of the Ca^{2+} spike plateau. As discussed in the preceding paper (Paupardin-Tritsch, D., L. Colombaioni, P. Deterre, and H. M. Gerschenfeld (1985) *J. Neurosci.* 5: 2522–2532), it is suggested that these FMRF-amide-induced modulations of ionic conductances involved in the Ca^{2+} -dependent spike recorded in these neuronal somata may intervene in processes of presynaptic inhibition and facilitation.

In the preceding paper (Paupardin-Tritsch et al., 1985), we have shown that dopamine (DA) modulates the amplitude of the Ca^{2+} -dependent plateau of the action potential recorded in the cell body of identified snail neurons. Such DA-induced modulation results from two different effects: DA induces in some neurons a decrease of the spike plateau by decreasing the membrane Ca^{2+} conductance, whereas it evokes in other neurons an increase of the amplitude of the spike plateau by depressing an outward current (the S-current),

i.e., by evoking a cAMP-mediated decrease of K^{+} conductance. In another small group of identified snail neurons, DA induces both the decrease in the Ca^{2+} current and the suppression of the S-current. However, in these cells DA only evokes a decrease of the Ca^{2+} -dependent plateau of the spike.

The molluscan neuropeptide Phe-Met-Arg-Phe- NH_2 (FMRF-amide), first isolated from the ganglia of the clam, *Macrocallista nimbosa* (Price and Greenberg, 1977a, b), was found to exert positive inotropic and chronotropic actions on the heart of the mollusc *Mercenaria* and to evoke contracture of the smooth muscles of other molluscs (Price and Greenberg, 1980). More recently, Price (1982) has reported that a longer peptide, structurally similar to FMRF-amide, is present in the ganglia of *Helix aspersa*, and Brown et al. (1984) have observed in *Aplysia californica* that many identified and unidentified neurons of the abdominal ganglion were immunoreactive to FMRF-amide antisera. In *H. aspersa*, Cottrell and his colleagues (Cottrell, 1982; Cottrell et al., 1984) have also observed that FMRF-amide and some related peptides elicit multiple responses in identified neurons: an increase in cationic conductance, an increase in K^{+} conductance, and a decrease of a K^{+} conductance, possibly the Ca^{2+} -dependent K^{+} conductance.

In the present paper we have examined the modulatory effects of FMRF-amide on the Ca^{2+} -dependent action potential of snail neurons. This neuropeptide, like DA, was found to elicit a decrease of the Ca^{2+} -dependent plateau of the somatic action potential of some identified snail neurons by inducing a decrease in their membrane Ca^{2+} conductance. It was also observed that, in a single identified snail neuron, cell E11, FMRF-amide not only decreases the Ca^{2+} current but also depresses the S-current. Moreover, when the E11 neuron was held at $-20/+10$ mV, FMRF-amide induced an inward current which was mimicked by the intracellular injection of cyclic 3',5'-adenosine monophosphate (cAMP). It was also found that FMRF-amide stimulated the adenylate cyclase activity of single E11 neurons. In spite of evoking such a cAMP-mediated decrease in K^{+} conductance in cell E11, FMRF-amide was observed to decrease the spike plateau. No FMRF-amide-induced increase of the spike plateau was observed in any other of the examined snail neurons.

Some of these results were previously reported in preliminary form (Paupardin-Tritsch et al., 1984).

Materials and Methods

The experiments were performed on the isolated and desheathed abdominal ganglionic mass of the snail, *Helix aspersa*, bathed in a circulating saline of the following composition (in millimolar concentration): NaCl, 120; KCl, 2.5; CaCl_2 , 6; MgCl_2 , 3.5; HEPES, 10 (pH 7.4). The neurons (D2, D3, E2, E11, and E13) were identified according to the ganglion maps of Kerkut et al. (1975).

The methods of intracellular recording, of modification of the ionic composition of the saline, and of extracellular and intracellular application of

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pharmacological agents were similar to those described in the previous paper (Paupardin-Tritsch et al., 1985). FMRF-amide (Cambridge Research Biochemicals, United Kingdom) was dissolved (0.1 M) in distilled water for iontophoretic application and in saline for both pressure and bath application. In the last two cases, the pH of the solutions was adjusted to 7.4.

The protocol of the biochemical assay of adenylate cyclase activity in isolated single snail neurons has been also described in the previous paper (Paupardin-Tritsch et al., 1985).

Results

FMRF-amide decreases the calcium-dependent plateau phase of snail neurons E2, D3, and E13. Figure 1A, control, illustrates action potential elicited by injecting short pulses of current in the soma of an E2 neuron bathed in a saline containing 30 mM tetraethylammonium (TEA) chloride. The control spike showed a large Ca^{2+} -dependent plateau having an amplitude that was decreased by the bath application of 20 μM FMRF-amide (Fig. 1A, FMRF).

To analyze the changes in the transmembrane currents associated with this effect of FMRF-amide, the E2 neuron was voltage clamped and the composition of the TEA-containing saline bathing the neuron was modified by replacing the Ca^{2+} content of the saline by Ba^{2+} . The cell membrane potential was initially held at -60 mV and stepped to $+10$ mV for 40 msec, and at the end of the pulses the membrane was repolarized to -75 mV, the potential corresponding to the K^+ equilibrium potential (E_K). An inward current followed by an inward tail current (Fig. 1B, arrow) was then recorded (Fig. 1B, control). The application of 20 μM FMRF-amide evoked a 30% decrease of the amplitude of the inward current and a marked decrease of the inward tail current (Fig. 1B, FMRF). In contrast, when the Ca^{2+} content of a TEA-containing saline was replaced by Mg^{2+} and the membrane potential was held at -50 mV and stepped for 1 sec to $+10$ mV, the TEA-resistant outward current components were not affected by the same concentration of FMRF-amide. The trace of Figure 1C actually corresponds to two superimposed current recordings obtained in the absence and in the presence of the peptide. It can be concluded, therefore, that FMRF-amide only induced a decrease of the inward current in cell E2 (and similarly in neuron D3)

The I-V curves of Figure 2 were obtained in a D3 neuron bathed in a TEA/TTX (tetrodotoxin)-containing extracellular medium in which Ca^{2+} was replaced by Ba^{2+} . These I-V curves were corrected by subtracting the leakage current. The graph of Figure 2 clearly shows

that the application of 20 μM FMRF-amide (triangles) decreased the control Ca^{2+} current values (circles) throughout the range of membrane potential examined and that the values of the Ca^{2+} current in the presence or absence of the peptide in the bath became maximal

Figure 2. Effect of FMRF-amide on the I-V curves obtained in a D3 neuron bathed in a TTX/TEA-containing saline in which Ca^{2+} was replaced by Ba^{2+} . The maximum inward currents obtained in the absence (●) and in the presence (▲) of 20 μM FMRF-amide were plotted against the values at which the membrane potential was stepped. Both curves were corrected for linear leakage conductance.

Figure 1. Decrease of the Ca^{2+} -dependent plateau of the action potential and of the inward current evoked in neuron E2 by FMRF-amide. A, Bath application of 20 μM FMRF-amide to an E2 cell bathed in a saline containing 30 mM TEA evokes a marked decrease of the spike plateau and a small decrease of the overshoot. B, Inward current recorded after voltage-clamping the same E2 neuron. The cell is bathed in a TEA-containing saline in which Ca^{2+} was replaced with Ba^{2+} . The cell initially held at -60 mV is depolarized for 40 msec to $+10$ mV and then repolarized to -75 , the potential level corresponding to E_K . The application of 20 μM FMRF-amide (FMRF) decreases the amplitude of both the inward current and the tail inward current. C, Outward current recorded in the same E2 cell bathed in a TEA-containing saline in which Ca^{2+} was replaced with Mg^{2+} . The cell was held at -60 mV and the voltage was stepped to $+20$ mV for 1200 msec. The trace corresponds to two superimposed currents recorded either in the absence or in the presence of FMRF-amide which has no effect on the outward current.

at the same potential level. Moreover, the maximal decrease of the Ca^{2+} current evoked by FMRF-amide is observed at the membrane potential value at which the control Ca^{2+} current reached its maximum.

Unfortunately, in the voltage-clamp conditions of our experiments we could not resolve the question of a possible modification of the inward current reversal potential by FMRF-amide. However, the calculations made by Dunlap and Fischbach (1980) for the Ca^{2+} current decrease by noradrenaline in chick sensory ganglion cells in culture indicate that, to account for a 30 to 40% decrease of the Ca^{2+} current, the shift of the reversal potential should be much more important than may be suspected from Figure 2. These results strongly suggest that FMRF-amide decreases the Ca^{2+} conductance of the D3 neuron.

The intracellular injection of either cAMP or cyclic 3',5'-guanosine monophosphate (cGMP) did not mimic the effects of FMRF-amide on the Ca^{2+} conductance of neurons E2 and D3. Prolonged intracellular injections of EGTA in these cells evoked an increase of the amplitude of the control Ca^{2+} current amplitude. Nevertheless, the FMRF-amide-induced decrease of the Ca^{2+} current in these EGTA-treated neurons was proportional to that observed on the control recordings.

Composite effects of FMRF-amide on the membrane currents of snail E11 neuron. When FMRF-amide was assayed on the action potential of cell E11, it was also observed that the peptide depressed the amplitude of the Ca^{2+} -dependent plateau (Fig. 3A). However, the voltage-clamp analysis of the effects of FMRF-amide on the membrane currents of cell E11 yielded a result somewhat different from that obtained in cells E2 and D3. Figure 3B, *control*, illustrates the Ca^{2+} current of an E11 neuron bathed in a TTX/TEA-containing extracellular medium in which Ca^{2+} was replaced by Ba^{2+} . FMRF-amide decreased the Ca^{2+} conductance of cell E11 (Fig. 3B, *FMRF*). Nevertheless, when the E11 neuron was bathed in a normal saline containing 30 mM TEA and its potential, initially held at -50 mV, was stepped to $+10$ by 1-sec voltage pulses, it could be observed that FMRF-amide also evoked a decrease of the TEA-resistant outward current (Fig. 3C). Further experiments applying 4-aminopyridine (4-AP) and removing Ca^{2+} from the extracellular medium, similar to those described in the previous paper (Paupardin-Tritsch et al., 1985), showed that FMRF-amide, as with DA, did not affect I_A , I_{DR} , or I_C (see D. J. Adams et al., 1980; Adams, 1982), but that it suppressed the S-current of cell E11. The S-current (see Klein et al., 1982) is associated with the opening of a group of background K^+

channels which constitutes the main population of open channels when the membrane potential of certain neurons, such as the sensory neurons of *Aplysia* and some identified neurons of *Helix*, is held at $0/+10$ mV (see Siegelbaum et al., 1982). As shown previously, the closing of these channels by either the extracellular application of either 5-HT or DA, or the intracellular injection of cAMP, generated an inward current (Deterre et al., 1982; see Paupardin-Tritsch et al., 1985).

The experiment of Figure 4, A and B, illustrates that the iontophoretic application of FMRF-amide on an E11 cell voltage clamped at $+10$ mV also evoked an inward current with a time course similar to that evoked by iontophoretically applied DA on a neighboring

Figure 4. Iontophoretic application of either FMRF-amide (A) or DA (B) on to an E11 neuron held at $+10$ mV evokes inward currents of similar features. B, Bath application of $20 \mu M$ DA evokes a saturating inward current and further iontophoretic application of FMRF-amide becomes ineffective. (see the text).

Figure 3. Composite effect of FMRF-amide on the membrane currents of neuron E11. A, Bath application of $20 \mu M$ FMRF-amide (*FMRF*) decreases the amplitude of the spike plateau of an E11 cell bathed in a saline containing 30 mM TEA. B, Inward current of the same F5 cell recorded in a saline containing both 30 mM TEA and $5 \mu M$ TTX and in which Ca^{2+} was replaced with Ba^{2+} . The neuron held at -50 mV is depolarized to 0 mV using a 50-msec duration pulse (*control*). Bath application of $20 \mu M$ FMRF-amide decreases the inward current (*FMRF*). C, The outward current of the same F5 neuron is recorded in a TEA-containing saline. The neuron held at -60 mV is depolarized for 1200 msec to $+20$ mV (*control*). The application of the same concentration of FMRF-amide also decreases the outward current (*FMRF*).

Figure 5. I-V curves relating the normalized net currents evoked by the iontophoretic applications of FMRF-amide (■) and of DA (●) in an E11 neuron bathed in saline containing 30 mM TEA. The steady-state I-V curves were first obtained by stepping the voltage from the holding level (+10 mV) to -90 and +50 by using 1-sec-duration pulses and measuring the current at the end of the pulses. The net currents evoked by either FMRF-amide or DA were obtained by subtracting the steady-state I-V curves obtained in the presence of the two agents from the control steady-state I-V curves.

membrane spot of the same cell. When FMRF-amide was bath applied for 10 min it induced in cell E11 a sustained inward current which did not show any desensitization. This current reached a maximal amplitude and saturated at 20 μ M concentrations of the peptide. The application of this maximal concentration of FMRF-amide made ineffective any further increase of the peptide concentration or repetitive iontophoretic application of FMRF-amide. Moreover, in the presence of a maximal saturating current evoked by DA, FMRF-amide also became ineffective (Fig. 4C). This observation suggested that both FMRF-amide and DA close the same population of membrane channels. This was confirmed by the I-V relations of Figure 5. In this graph, the normalized values of the net currents elicited by the extracellular application of either FMRF-amide or DA to neuron E11 were plotted against the voltage values at which the membrane potential was stepped to measure the currents (for the method of establishing the I-V curves, see the legend of Fig. 4). The curve of Figure 4, traced by eye, fits well the values of the net currents induced by both FMRF-amide and DA. Both currents reversed at -50 mV. As in the case of the DA- and cAMP-induced net currents presented in a similar graph of the previous paper (Paupardin-Tritsch et al., 1985), the value of the reversal potential of the FMRF-amide-induced current was not too close to the value of E_K in snail neurons (-75/-80 mV). However, the inversion potential of the FMRF-amide-induced net current was sensitive to changes in the extracellular K^+ concentration, even if it did not strictly follow the changes predicted by the Nernst equation. This difference between E_K and the inversion potential of the FMRF-amide-induced current could be due to the masked activation of other FMRF-amide-induced currents (Cottrell et al., 1984) in cell E11, or because the steady holding of the membrane potential at +10 during the voltage stepping procedure could have produced an extracellular K^+ accumulation, or because of a lack of selectivity of the channels. In any case, these results support the idea that FMRF-amide, as did DA, induced

a decrease in K^+ conductance. This conclusion is further supported by the results of other experiments (not shown) in which K^+ channel blockers were assayed. The intracellular injection of either TEA or Cs ions in cell E11 blocked the inward current elicited by FMRF-amide as it was previously shown to block those induced by 5-HT and DA (Deterre et al., 1981, 1982).

As described in the previous paper (Paupardin-Tritsch et al., 1985) for the effects of DA, the replacement of the Ca^{2+} content of the saline by either Co^{2+} or Ba^{2+} blocked both the S-current and the inward current evoked by FMRF-amide in cell E11. Nevertheless, the K^+ conductance decreased by both FMRF-amide and DA did not appear to involve a Ca^{2+} -dependent K^+ conductance, since prolonged intracellular injections of EGTA in cell E11 did not affect the FMRF-amide-induced inward current, even if this procedure blocked in the same neuron the Ca^{2+} -dependent K^+ conductance

Figure 6. The iontophoretic application of FMRF-amide (A) and the intracellular pressure injection of cAMP (B) evoke inward currents of similar characteristics in an E11 cell held at +10 mV. In C and D, the iontophoretic application of FMRF-amide (FMRF) on two other E11 neurons evokes biphasic responses composed of a small outward current followed by a much larger inward current. The iontophoretic application of FMRF-amide becomes unable to evoke an inward current in the presence of a saturating inward current elicited by the application of either 20 μ M forskolin (C) or 100 μ M IBMX (D). In these cases, the FMRF-amide-induced outward currents appear somewhat larger. Also in C and D, the records show "spontaneous" synaptic currents independent from the effects of FMRF-amide.

which was associated with the afterhyperpolarization that followed a train of action potentials.

Figure 6, *A* and *B* shows that the intracellular injection of cAMP in neuron E11 mimicked the effects of FMRF-amide and DA. The cAMP-induced inward current showed the same ionic and pharmacological properties as those evoked by FMRF-amide. When a maximal saturating inward current was obtained by applying 20 μ M FMRF-amide, the intracellular injection of cAMP became ineffective, thus suggesting that the ionic channels closed by both agents were the same.

Another group of experiments using agents which caused an increase in the intracellular cAMP concentration completed the evidence indicating that cAMP mediated the decrease in K^+ con-

ductance evoked by FMRF-amide. Thus, the application of 20 μ M forskolin, a diterpene activator of adenylate cyclase in broken and intact cells (Seamon and Daly, 1981), also evoked in the E11 cell a maximal inward current. In this case, further application of FMRF-amide (Fig. 6*C*) was unable to evoke an inward current. In contrast, the application of 100 μ M isobutylmethylxanthine (IBMX), a phosphodiesterase inhibitor, was observed to evoke a maximal saturating inward current in cell E11—in this condition, further application of FMRF-amide became ineffective (Fig. 6*D*). Finally, in single isolated E11 neurons, 20 μ M FMRF-amide induced a 3-fold stimulation of the adenylate cyclase activity (Table I).

A comparison of the effects of FMRF-amide and DA on the action potential of different identified snail neurons. In the present paper and in the previous one (Paupardin-Tritsch et al., 1985), FMRF-amide and DA were shown to decrease the Ca^{2+} -dependent plateau phase of the action potential of snail neurons. We therefore explored whether the two agents acted in a similar or a different manner on a group of identified snail neurons. Figure 7 illustrates the results of these experiments. Three different situations were observed. The two top recordings of Figure 7 (*a*₁ and *a*₂) correspond to neuron D3, in which both FMRF-amide and DA decreased equally the spike plateau. In contrast, in other neurons the effect of each agent was independent of the effect of the other. Thus, in cell D2 (Fig. 7, *b*₁ and *b*₂), DA decreased the spike plateau, whereas FMRF-amide did not affect it, and in neuron E13 FMRF-amide decreased the spike plateau, whereas DA evoked an increase of the amplitude of this plateau (see Paupardin-Tritsch et al., 1985).

Discussion

The FMRF-amide-induced decrease of calcium conductance. The present results indicate that the neuropeptide FMRF-amide decreases the Ca^{2+} -dependent plateau phase of the snail neurons D3, E2, and E13 by decreasing their Ca^{2+} conductance. This effect is identical to that exerted by DA on the Ca^{2+} spike of other identified snail neurons (Paupardin-Tritsch et al., 1985) and by other neuropeptides on the Ca^{2+} spike of some vertebrate neurons. Thus, Met-enkephalin and somatostatin have been reported to evoke a decrease in the duration of the Ca^{2+} spike of chick embryo sensory neurons in culture by decreasing their Ca^{2+} conductance (Dunlap and Fischbach, 1980). Met-enkephalin was also observed to shorten the Ca^{2+} spikes of amphibian Rohon-Beard neurons (Bixby and Spitzer, 1983) and Leu-enkephalin, as well as other opioid peptides, to decrease the duration of the somatic spike of mice sensory ganglion neurons in culture (Werz and Macdonald, 1982).

The fact that the Ca^{2+} conductance decrease evoked by FMRF-amide or DA can be observed in different snail neurons and independently from each other indicates that the receptors involved in this effect of the peptide and of DA are different. Moreover, the independence of these actions also suggests that these agents do not exert their effects directly on the Ca^{2+} channels, but that they are probably mediated by a common second intracellular messenger. We did not succeed in identifying this second messenger. Our experiments showed, however, that the Ca^{2+} conductance decrease elicited by both FMRF-amide and DA (see Paupardin-Tritsch et al., 1985) is not mediated by an increase in the intracellular concentration of cAMP, cGMP, or Ca^{2+} .

The cell body of neurons carrying a Ca^{2+} spike has constituted an interesting model to understand the events involved in the transmitter-induced modulation of the Ca^{2+} channels located at the synaptic endings. In the previous paper (Paupardin-Tritsch et al., 1985), we have discussed some proposed examples of presynaptic inhibition involving a decrease of the Ca^{2+} conductance induced by GABA²⁺, by catecholamines, or by an unidentified transmitter (see also Dunlap and Fischbach, 1980; Shapiro et al., 1980). The mediation of such an effect by peptides was clearly raised by the work of Mudge et al. (1979), which showed that enkephalin decreases both the duration of the Ca^{2+} spike of chick embryo sensory ganglion neurons in culture and the release of substance P from these

TABLE I

Adenylate cyclase activity in identified snail neurons

cAMP values are means $\pm$ SEM for 10 different neurons of the same class. Observed increases are given as percentages of the basal activity.

Neuron	Base cAMP (10^2 fmol/30 min/cell)	FMRF (200 μ M)	Increase
E2	19 ± 2	25 ± 3	0%
E11	29 ± 3	61 ± 11^a	168%

^a $p < 0.001$.

Figure 7. Comparative effects of FMRF-amide and DA on Ca^{2+} spikes. All of the recordings correspond to preparations bathed with a TEA-containing saline. *a*₁ and *a*₂, Recording of action potentials of neuron D3. The application of DA and FMRF-amide both evoke a decrease of the plateau on the spikes. *b*₁ and *b*₂, In cell D2, the application of DA decreases the spike plateau, but FMRF-amide is ineffective; the trace of *b*₂ actually corresponds to the superimposed recordings of the action potential in the absence and in the presence of FMRF-amide. *c*₁ and *c*₂, In cell E13 the effect of DA is opposite to that of FMRF-amide. DA increases the amplitude of the spike plateau whereas FMRF-amide decreases it. In all experiments FMRF-amide and DA were applied, respectively, at 20 μ M and 10 μ M concentrations.

neurons. By analogy, it could be suggested that if FMRF-amide receptors involved in the decrease of the Ca^{2+} conductance were present at the synaptic endings of the snail neurons E2, D3, and E13, then FMRF-amide could be a transmitter mediating presynaptic inhibition at these endings.

The composite effects of FMRF-amide on neuron E11. In spite of evoking a decrease of the somatic spike plateau of cell E11, FMRF-amide was found to induce in this neuron, besides a decrease in Ca^{2+} conductance, a decrease in an outward current component. This outward current component suppressed by FMRF-amide was insensitive to TEA, 4-AP, and Ca-free media, but was blocked by both Ba^{2+} and Co^{2+} . In the previous paper (Paupardin-Tritsch et al., 1985), we have reviewed the evidence in favor of the identity of this outward current component, blocked by FMRF-amide (and by DA), with the S-current suppressed by serotonin (5-hydroxytryptamine, (5-HT) in *Aplysia* sensory neurons (Klein and Kandel, 1978, 1980; Klein et al., 1982). In E11 neurons voltage-clamped near the zero level, FMRF-amide could elicit an inward current which reflected the closing of the ionic channels generating the S-current (see Siegelbaum et al., 1982). A similar inward current was evoked by cAMP and by agents which increased the intracellular concentration of this nucleotide, and FMRF-amide stimulated the adenylate cyclase activity of cell E11. All of these effects of FMRF-amide resemble those described in the preceding paper (Paupardin-Tritsch et al., 1985) as being produced by DA in snail neurons E13 and F1. Therefore, we would suggest here that the outward current decreased by FMRF-amide in the E11 neuron, like the outward current decreased by DA in the E13 and F1 neurons, closely resembles the S-current suppressed by 5-HT in *Aplysia* sensory neurons (Klein et al., 1982); that is, FMRF-amide evokes in cell E11 a cAMP-mediated decrease of K^+ conductance.

Recently, Cottrell et al. (1984) have also observed that FMRF-amide elicited a K^+ conductance decrease in another identified snail neuron, the C1 cell. Since this response was blocked by Co^{2+} , these authors postulated that it was possibly associated with a decrease in Ca^{2+} -dependent K^+ conductance. However, the K^+ conductance decrease evoked by the peptide in cell E11 shows many properties similar to those observed in cell C1, including its sensitivity to Co^{2+} . We are therefore in favor of the idea that both responses are similar.

It was also previously shown (Deterre et al., 1982) that DA and 5-HT elicit cAMP-mediated decreases of K^+ conductance in identified snail neurons by activating different receptors. Since FMRF-amide does not affect the S-current in the majority of neurons in which 5-HT and/or DA suppress it, it is highly probable that the receptors activated by FMRF-amide are independent from the DA and 5-HT receptors. Therefore, the present results provide additional evidence that the S-current, generated by the closure of a population of K^+ channels controlled by the intracellular cAMP concentration, can be suppressed by two different amines and a peptide, which activate different membrane receptors linked to adenylate cyclase. Abrams et al. (1984) have also recently reported that, besides 5-HT, two other molluscan cardioexcitatory peptides, SCP_A and SCP_B , induced a similar cAMP-mediated decrease of the S-channel conductance in *Aplysia* sensory neurons.

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