ATRIONATRIURETIC PEPTIDE (ANP) ENHANCES THE SELECTIVITY OF Na^+ CHANNEL TO Ca^{2+} IN RAT (RATTUS NORVEGICUS) AND GUINEA PIG (CAVIA COBAYA) VENTRICULAR MYOCYTES

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ANP is a vasoactive peptide which is released from the atrium in response to volume expansion. ANP reduces Na^+ absorption by the kidney and thereby reduces blood volume. It has been reported that ANP reduces i_{Ca} in frog ventricular myocytes, through an adenylate cyclase-dependent pathway (Gisbert & Fischmeister, Circ. Res. 1988; 62: 660). In this report, we examined the effect of ANP on the Na^+ current, i_{Na}, and Ca^{2+} current, i_{Ca}, in isolated, whole cell clamped adult rat and guinea pig ventricular myocytes.

Myocytes were prepared by enzymatic dissociation (Mitra and Morad, Am. J. Physiol. 249: H1056-H1060, 1985). The myocytes were placed in a chamber coated with fibronectin to stabilize them mechanically, making it possible to exchange the bathing solutions using an electronically controlled, multibarrelled concentration-clamp system. K^+ was omitted from both external and internal solutions and was replaced by either Cs^+ or n-methyl glucamine. Intracellular Ca^+ and H^+ concentrations were highly buffered, with 14 mM EGTA and 10-20 mM HEPES, at pH 7.2. ANP (rat ANF, 1-28 amino acids) was obtained from Peninsula Labs (San Diego, California).

Application of 100 nM ANP suppressed the Na^+ current that had been activated by a depolarizing pulse from -80 to -40 mV (Figure 1A). Panel B shows the suppression of the current-voltage relation of i_{Na} by ANP. The Ca^{2+} current was similarly reduced (panel C). The current-voltage relations are based on Panel D.

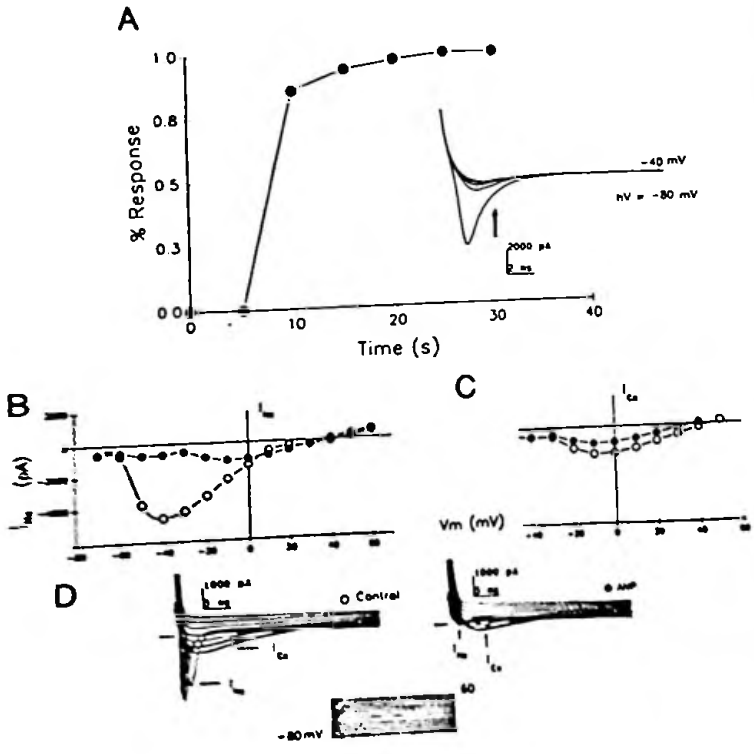

Figure 1. Suppression of i_{Na} (Panels A, B, and D) and i_{Ca} (Panels C and D) by 100 nM ANP. Panel A shows the rapid suppression of i_{Na} (inset traces) by ANP. Current-voltage relations (Panels B and C, -80 mV holding potential) measured before (open circles, Panel D left) and after (filled circles, Panel D right) addition of ANP show that both i_{Na} (Panel B, fast inward current in panel D) and i_{Ca} (Panel C, slow inward current in panel D) were suppressed by ANP.

When Ca^{2+} was omitted from the external solution, ANP failed to suppress I_{Na}, suggesting that the ANP effect was Ca^{2+} dependent. When external Na^+ was completely replaced by Cs^+ or n-methyl glucamine, ANP induced a rapidly activating and inactivating current with kinetics and voltage dependency similar to that of the Na^+ channel. In the absence of external Ca^{2+}, ANP failed to activate this current suggesting that Ca^{2+} was the charge carrier. Since 10 μM TTX blocked this current, but the current was unaffected by 5 mM Ni^{2+} or 10 μM nifedipine, Na^+ channel were thought to carry this Ca^{2+} current.

Our results therefore suggest that ANP induces a molecular transformation in the Na^+ channel making it more selective to calcium, without affecting its kinetics and pharmacological sensitivity. The combined suppressive effect of ANP on Na^+ and Ca^{2+} channels may render the secretory atrial tissues inexcitable and non-contracting. Thus, this mechanism may be involved in the feedback regulation of ANP secretion.

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