On the Involvement of Endogenous μ- and δ-Opiate Receptor Agonists in the Antiarrhythmic Effect of Adaptation

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Rats adapted to stress showed a decreased severity and incidence of cardiac arrhythmias induced by epinephrine, and these effects of adaptation were abolished by naloxone. It is suggested that stress adaptation mitigates arrhythmia by activating the endogenous opioid system and stimulating the μ-opiate receptors.

Key Words: *opiate receptors; adaptation; arrhythmia; stress*

Recent studies have demonstrated that adaptation of animals to a stressor through periodic short-term exposures to it can mitigate the arrhythmogenic effect of subsequent coronary occlusion [2]. The mechanism of this phenomenon is not clear and has attracted the attention of many investigators because its elucidation will not only add to our basic knowledge of adaptation processes but also break new ground in pharmacology and cardiology.

As we showed previously, animals adapted to stress by being repeatedly immobilized for short periods contain elevated levels of opioid peptides, some of which exhibit antiarrhythmic activity after coronary occlusion [3]. This suggested to us that the antiarrhythmic effect of adaptation might be mediated by the endogenous opioid peptides accumulating in the body. There also remained the question of whether the antiarrhythmic effect is confined to the arrhythmias following coronary occlusion or extends to those caused by other arrhythmogenic agents or factors. The present study attempts to answer these two questions.

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MATERIALS AND METHODS

Male Wistar rats weighing 180-200 g were used. Test rats were adapted to stress by being immobilized in the supine position for 15 min on day 1, 30 min on day 2, 45 min on day 3, and 60 min on alternate days subsequently, for a total of 12 days. We found earlier that this method of adaptation results in elevated levels of opioid peptides in the organs and tissues [3]. Intact rats served as unadapted controls.

Arrhythmias were produced by injecting rats intravenously with epinephrine (100 μg/kg) or $CaCl_2$ (100 mg/kg) under etherrausch. During the 5-minute postinjection period, the electrocardiogram was recorded and the number of ventricular arrhythmias counted. Fifteen minutes prior to the injection of these arrhythmogenic agents, the animals received intravenously isotonic sodium chloride solution (adapted controls) or an opiate receptor (OR) antagonist - either naltrindole (synthesized by Professor P. S. Portoghese, Department of Medical Chemistry, College of Pharmacy, Minneapolis, USA), in a dose high enough to cause complete inactivation of the central and peripheral δ-OR (10 mg/kg) [4], or the nonselective μ-OR inhibitor naloxone (Sigma) in a dose sufficient for blocking only the μ-OR (0.2 mg/kg) or for "cutting off" the bulk of OR [7,9].

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TABLE 1. Effects of Opiate Receptor Antagonists on Epinephrine- and $CaCl_2$-Induced Arrhythmias in Stress-Adapted Rats

Group	No. of rats with epinephrine-induced arrhythmias				No. of rats with $CaCl_2$-induced arrhythmias			
	n	without VE	with VE	with VF	*n*	without VE	with VE	with VF
Unadapted controls	19	7	12	-	29	9	12	6
Adapted controls	21	21**	-**	-	15	10*	3	1
Adapted+naloxone, 0.2 mg/kg	16	2**	7**	3*	16	2**	8	5
Adapted+naloxone, 2.0 mg/kg	19	6**	9**	1	20	4**	8	5
Adapted+naltrindole, 10 mg/kg	21	21**	-**	-	20	14**	5	1

Note. VE = ventricular extrasystole; VF = ventricular fibrillation. Significance of differences: *$p<0.02$, **$p<0.001$ from intact controls; *$p<0.05$, **$p<0.001$ from adapted controls by the χ^2 test.

The results were subjected to statistical analysis by the χ^2 test.

RESULTS

As shown in Table 1, the preliminary adaptation of rats to stress by repeated immobilization for short periods prevented the occurrence of arrhythmia under the action of epinephrine and $CaCl_2$. These two substances are known to provoke the emergence of ectopic pacemakers in the myocardium, but the molecular mechanisms of their action differ. Thus, $CaCl_2$ induces arrhythmias by causing Ca^{2+} ions to exit from the sarcoplasmic reticulum and by generating an "input current" [5,6], whereas epinephrine does so by activating the β-adrenoreceptors with a consequent elevation of the intracellular cAMP concentration [8].

It seems, then, that the antiarrhythmic effect of adaptation to stress is due both to a diminished epinephrine responsiveness of cardiac muscle cells and to an enhanced elimination of excess Ca^{2+} from the cytoplasm of these cells. This hypothesis is in accord with observations that adaptation lowers the affinity of the β-adrenergic receptors and boosts the activity of the sarcoplasmic Ca^{2+} pump in the myocardium [1,2].

To ascertain whether endogenous opioids can mediate the antiarrhythmic effect of adaptation, we carried out a series of tests in which stress-adapted rats were administered an OR blocker before the induction of arrhythmias. Naloxone at 0.2 mg/kg, a dose high enough to block only μ-OR, completely abolished the antiarrhythmic effect, and a similar effect was observed with a naloxone dose of 2 mg/kg, which also inhibits OR of other types (Table 1). Naltrindole, a selective δ-OR blocker, failed to abolish the antiarrhythmic effect of adaptation. It should be noted that the arrhythmogenic activity of epinephrine or $CaCl_2$ in intact (i.e., unadapted) rats was not altered by naloxone in the indicated doses (data not shown).

The results of this study indicate that endogenous agonists of μ-OR play a key role in mediating the antiarrhythmic effect of adaptation to stress, and that δ-OR and their agonists are most likely not involved in regulating the electrical stability of the heart in adapted animals.

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