GENERAL PATHOLOGY AND PATHOLOGICAL PHYSIOLOGY

Modulating Effect of μ-Opiate Receptor Ligands on Adrenergic Stage in Pathogenesis of Stress-Induced Damage to the Heart

Yu. B. Lishmanov, N. V. Naryzhnaya, Yu. G. Revinskaya, and L. N. Maslov

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 126, No. 11, pp. 510-512, November, 1998
Original article submitted December 2, 1997

The agonists of μ-opiate receptors, DAGO and DALDA, prevent stress-induced enhancement of ^{99m}Tc-pyrophosphate accumulation in the myocardium, which attests to cardioprotective activity of these opioids. This phenomenon is presumably related to modulating effect of these agents on the adrenergic stage in pathogenesis of stress-induced damage to the heart.

Key Words: *opiate receptors; stress; catecholamines*

Fatal disturbances in cardiac ventricular rhythm and sudden death are often observed in patients without coronary anamnesis, who had a severe stress or long-term psychoemotional overload [11]. These phenomena can be considered as clinical equivalents of stress-induced damage to the heart (SIDH) [11]. One of the leading factors of SIDH is the cardiotoxic effect of an excess of endogenous catecholamines (CA) caused by hyperactivation of sympathoadrenal system [2,8,9]. However, there are no direct data to prove the key role of CA in pathogenesis of SIDH.

We showed that in contrast to ligands of other types of opiate receptors (OR), the ligands of μ-OR are capable of modulating the degree of SIDH. Thus, there is a firm basis to believe that opioids produce cardioprotective effect by curtailing the secretion of endogenous CA, because activation of some types of OR is accompanied by inhibition of CA release from the sympathetic nerve terminals. There is evidence on the antagonism between opioid peptides and CA for β-adrenoceptors [13]. However, the receptor specificity of these effects is barely known.

Our aim was to study the modulating role of μ-OR ligands to the adrenergic stage in SIDH pathogenesis.

MATERIALS AND METHODS

The study was carried out on male Wistar rats weighing 150-200 g. Stress was modeled by a 24-h immobilization in the supine position. The adrenergic structures were affected by intraperitoneal (10 mg/kg) bretylium (Sigma), a blocker of CA release from adrenergic terminals [6], or by daily administration of guanethidine (Sigma) for 3 days in a dose of 50 mg/kg, which suppressed the synthesis of CA in nerve terminals [5].

The following ligands were used to stimulate μ-OR: DAGO ([D-Ala^2,N-Me-Phe^4,Gly^5-ol]-enkephalin) [10] or DALDA ([D-Arg^2,Lys^4]dermorphin-(1-4)-amide), which were injected intraperitoneally in a single dose of 0.1 mg/kg: 30 min prior to immobilization and 12 h after imposing the stress.

DALDA was synthesized by Prof. P. W. Schiller (Clinical Research Institute of Montreal, Canada),

Department of Experimental Cardiology, Institute of Cardiology, Siberian Division of the Russian Academy of Medical Sciences, Tomsk

0007-4888/98/0011-1095$20.00

TABLE 1. Effect of μ-OR Ligands on Accumulation of ^{99m}Tc-PPh in Rat Myocardium under Immobilization Stress (*M±m*, $n \geq 12$)

Animals	Specific accumulation of ^{99m}Tc-PPh (% of total dose/g tissue, $\times 10^{-2}$)
Intact	1.85±0.26
Stress-control (immobilization for 24 h)	2.86±0.31*
Stress+DAGO, 0.1 mg/kg	2.1±0.25*
Stress+DALDA, 0.1 mg/kg	1.59±0.22*
Stress+bretylium, 10 mg/kg	2.13±0.09*
Stress+guanethidine, 50 mg/kg	1.98±0.12**

Note. Here and in Table 2: *$p<0.05$ compared with intact rats; *$p<0.05$ and **$p<0.001$ compared with the stress-control.

and DAGO was synthesized at the BioPro (Novosibirsk).

SIDH was assessed by accumulation of ^{99m}Tc-pyrophosphate (^{99m}Tc-PPh) in damaged heart [12]. The level of CA was determined histochemically on cryostat sections of the adrenal glands and myocardium, using a 2% glyoxylic acid solution in accordance to modification [4] of method [3].

The results were statistically analyzed using Student's *t* test.

RESULTS

Table 1 shows that 24-h immobilization was accompanied by a 1.5-fold increase in ^{99m}Tc-PPh accumulation in the myocardium in comparison with baseline level, indicating damage to the cardiomyocyte membrane [12]. Simultaneously, the intensity of CA fluorescence decreased in the chromaffine cells of the adrenal gland medulla, and the content of CA in myocardial adrenergic fibers dropped 2-fold in comparison with intact rats. These alterations indicate the depletion of adrenergic neurotransmitters in the examined tissues due to stress-induced activation of the sympathoadrenal system [1,17].

Intraperitoneal administration of DAGO, a selective agonist of μ-OR, led to a statistically significant decrease in ^{99m}Tc-PPh accumulation in the myocardium of experimental rats by 26% (Table 1). In addition, it increased the content of histochemically determined CA in adrenal glands and heart relative the stress-control level by 16 and 13%, respectively (Table 2). To some degree, the specificity of these effects of DAGO is supported by the effect of systemic administration of the other agonist of μ-OR, DALDA, that modified the examined indices in the same way. For example, accumulation of ^{99m}Tc-PPh by cardiomyocytes in this series was 45% smaller than that in the stress-control group (Table 1), while the intensity of CA fluorescence on myocardial histological sections was 27% higher. The content of CA in the adrenal gland medulla did not change significantly in comparison with the stress control group.

These data indicate that stimulation of peripheral μ-OR enhances the tolerance of the cardiomyocyte membrane to stress-induced damage and simultaneously it facilitates the retention of CA in adrenal glands and adrenergic nerve terminals in the heart.

Presumably, the ability of the ligands of μ-OR to inhibit the release of CA from adrenergic terminals is responsible for their CA-saving effect, which agrees with similar effects of opiates [13]. Assuming the dependence of cardioprotective effect of the ligands of μ-OR on their ability to curtail the adrenergic transmission to myocardium as a working hypothesis, one should expect that other agents have similar properties (for example, guanethidine and bretylium) which affect the deposition and release of CA.

We could not detect any stress-induced damage to the heart in the rats that were pretreated with guanethidine to deplete CA depot. Accumulation of

TABLE 2. Effect of μ-OR Ligands on the Content of Catecholamines (CA) According to Histochemical Indices in Adrenal Glands and Rat Heart during Immobilization Stress (*M±m*, $n>=12$)

Animals	Intensity of CA fluorescence in chromaffin cells of adrenal gland medulla, arb. units	Intensity of CA fluorescence in myocardium, vol. %
Intact	6.1±0.3	2.2±0.1
Immobilization stress for 24 h	3.1±0.1***	1.1±0.1*
Stress+DAGO, 0.1 mg/kg	4.1±0.2***	1.4±0.1**
Stress+DALDA, 0.1 mg/kg	3.2±0.1**	1.6±0.1**
Stress+bretylium, 10 mg/kg	4.2±0.4**	1.7±0.1**
Stress+guanethidine, 50 mg/kg	1.2±0.02****	0.0****

Note. **$p<0.01$, ***$p<0.001$ compared with intact rats.

^{99m}Tc-PPh in the myocardium of the rats in this group was similar to that in the intact group (Table 1). The histochemical methods did not reveal CA in the myocardium of the rats in this group (Table 2). The intensity of CA fluorescence in the chromaffin cells of adrenal gland medulla in the rats administered with guanethidine was decreased by more than 5-fold in comparison with intact rats and by almost 3-fold in comparison with stress-control rats.

The degree of cardiac damage in rats subjected to stress under the effect of bretylium significantly decreased (by 25%) relative to the stress-control level. However, in contrast to the effect of guanethidine, CA content in the myocardium and adrenal glands was increased relative to stress-control by 54 and 35%, respectively.

The experiments with guanethidine and bretylium supported the hypothesis on the key role of CA in stress-induced damage to cardiomyocytes. Indeed, in both cases we observed moderation of damage to the heart during stress against the background of drastically limited release of adrenergic agents from presynaptic terminals. The only difference was that guanethidine depleted the adrenergic depots in the myocardium and the adrenal gland medulla [5] and thus drastically curtailed the release of transmitters during stress, while bretylium directly blocked epinephrine release from the sympathetic nerve terminals [6].

Thus, our data attest to enhancement of cardiac tolerance to stress-induced damage by peripheral administration of the μ-OR ligands DAGO and DALDA. The potency of these opioids to modulate the activity of the peripheral mechanisms of sympathoadrenal system and the release of CA from the adrenergic terminals is responsible for this cardioprotective effect.

REFERENCES

1. B. N. Manukhin, V. I. Pavlova, T. G. Putintseva, *et al., Fiziol. Zh. SSSR.*, **67**, No. 8, 1182-1188 (1981).
2. F. Z. Meerson, *Pathogenesis and Prophylaxis of Stress-Induced and Ischemic Damage to the Heart* [in Russian], Moscow (1984).
3. R. A. Stropus, K. A. Tamashauskas, and V. V. Yakubauskaite, in: *General Mechanisms of Morphogenesis and Regeneration* [in Russian], Kaunas (1976), pp. 68-69.
4. V. N. Shvalev and N. I. Zhuchkova, *Arkh. Anat.*, **76**, No. 6, 114-116 (1979).
5. W. A. Abrams, R. A. Moe, H. Bates, *et al., Am. J. Cardiol.*, **12**, 711-720 (1963).
6. M. B. Bacaner, *Ibid.*, **21**, 504-512 (1968).
7. C. Chang and C. Su, *J. Pharm. Pharmacol.*, **19**, No. 2, 73-77 (1967).
8. M. S. Cebelin and C. S. Hirsch, *Hum. Pathol*, **11**, 123-132 (1980).
9. J. Haggendal, L. Jonsson, and G. Johansson, *Acta Physiol. Scand.*, **131**, No. 11, 447-450 (1987).
10. H. W. Kostrelitz and S. J. Paterson, *Br. J. Pharmacol.*, **73**, Suppl., 299 (1980).
11. B. Lown, R. DeSilva, and P. Reich, *Am. J. Psychiatry.*, **137**, No. 11, 1325-1335 (1980).
12. D. G. Miller and S. Mallov, *Pharmacol. Biochem. Behav.*, **7**, No. 2, 139-145 (1977).
13. R.-P. Xiao, S. Pepe, H. A. Spurgeon, *et al., Am. J. Physiol.*, **272**, H797-H805 (1997).