PROTECTIVE ACTION OF α_2-ADRENORECEPTOR AGONISTS IN HYPOXIC HYPOXIA

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Selective stimulation of α-adrenoreceptors (of the α_2-subtype) by catecholamines and other adrenoreceptor agonists leads to a marked decrease in the oxygen consumption of mice [2, 5], and of animals of other species and man [2, 3]. It has been suggested that this so-called "anticalorigenic" effect of α-agonists is due to their inhibitory effect on oxidative processes in the tissues. In this case stimulators of α-adrenoreceptors may reduce the oxygen consumption of the body and, consequently, may increase resistance to hypoxia.

The aim of this investigation was to test this hypothesis experimentally and to study the receptor mechanism of the antihypoxic action of α-adrenoreceptor agonists.

METHODS

Experiments were carried out on 728 satiated (CBA × C57BL)F_1 female mice weighing 18-21 g. Hypoxic hypoxia was induced in three ways: 1) by "lifting" the group of animals in a pressure chamber to an "altitude" of 8000 m at the rate of 15-20 m/sec (acute hypobaric hypoxia); 2) placing the animals individually in an airtight glass container with a volume of 100 ml (hypercapnic hypoxia, asphyxia); 3) lowering the animals into a vessel containing water under a lid so that the whole of the animal was under the water, although still completely able to move (asphyxia by drowning). Death was recorded in the first two cases as respiratory arrest, and in the case of drowning, as the sudden cessation of motor activity. The air and water temperature during the experiments was 19-21°C. All substances were injected in a volume of 10 ml/kg body weight in optimal doses. The adrenoreceptor agonists naphazoline, phylephrine, norphenylephrine, and clonidine, were injected subcutaneously 10-15 min before exposure to hypoxia, i.e., at the time of maximal development of maximal development of the anitcalorigenic effect; the adrenoreceptor antagonists phenolamine, prazosin, piperoxan, and yohimbine were injected 15-20 min before the agonist. The results were subjected to statistical analysis by the t and chi-square tests, using Yates' correction. Differences were considered significant at the $P \leq 0.05$ level.

RESULTS

Injection of the nonselective α_1- and α_2-adrenoreceptor agonist naphazoline into mice caused a marked increase in the animals' resistance to hypoxia (antihypoxic action). This effect of naphazoline was observed on all three models of hypoxic hypoxia studied, and it was clearly manifested in all the parameters determined (Table 1): Both the survival rate of the animals after stipulated time intervals and the mean life span (MLS; under hypoxic conditions were increased.

To determine the subtype of the α-adrenergic structures mediating the effect of naphazoline, more selective α-adrenoreceptor agonists were used. It was found that phenylephrine, agonists of α_1-adrenoreceptors, did not increase the resistance of mice to hypoxia (Table 1): When both drugs were used, the survival rate and MLS were not significantly higher than in the control, and norphenylephrine actually reduced MLS of the animals which

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TABLE 1. Effect of α-Adrenoreceptor Agonists on Resistance of Mice to Various Forms of Hypoxic Hypoxia

Drugs	Dose, μmoles/kg	Hypobaric hypoxia			Hypercapnic hypoxia			Asphyxia by drowning		
		n	survival rate after 20 min, %	MLS, sec	n	survival rate after 20 min, %	MLS, sec	n	survival rate after 20 min, %	MLS, sec
Control	—	96	7±3	11,8±0,3	51	0+2	12,2±0,2	20	10+7	23,0±0,5
Naphazoline	24	24	45±11***	16,4±0,7***	4	100±25*4	31,1±11*4	19	74±10*4	26,8±0,5***
Phenylephrine	40	20	13±9	12,8±0,9	6	17±17	14,4±1,8	—	—	—
Norphenylephrine	27	—		—	9	0±11	11,2±1,2	10	0+10	20,8±0,6**
Clonidine	0,4	45	76±6*4	18,9±0,3*4	7	86±14*4	21,9±1,1*4	13	85±10*4	28,2±1,0*4
Clonidine	2	15	100±8*4	19,2±0,4*4	27	78±8*4	23,4±1,5*4	20	70±10*4	27,0±0,6*4

Legend. Level of significance compared with control: *P ≤ 0.1, **P ≤ 0.05, ***P ≤ 0.01, *4P ≤ 0.001. In Tables 1 and 2, n denotes number of experiments.

TABLE 2. Antihypoxic Action of Clonidine

Drugs	Hypobaric hypoxia			Hypercapnic hypoxia		
	n	survival rate after 20 min, %	MSL, sec	n	survival rate after 20 min, %	MLS, sec
Phentolamine	21	5±5	7,1±0,8*4	30	0±6	8,8±0,3*4
Phentolamine + clonidine	15	0±7	7,5±0,4c	25	20±9c	14,5±1,1
Prazosin	25	16±8	13,1±0,6	34	3±3	11,5±0,6
Prazosin + clonidine	25	92±6***	19,7±0,3*4	35	54±9*4	20,2±0,8*4
Piperoxan	18	11±8	11,2±0,8	11	0+9	11,1±0,7
Piperoxan + clonidine	20	30±10b,**	15,4±0,6b,*4	12	33±15b,*	17,3±1,3c,*4
Yohimbine	—	—	—	7	0+14	12,6±0,4
Yohimbine + clonidine	—	—	—	7	0+14	15,6±0,8c,*4

Legend. The dose of clonidine in the experiments with hypobaric hypoxia was 0.4 μmole/kg, and in those with hypercapnic hypoxia it was 2 μmoles/kg; doses of adrenoreceptor antagonists (in μmoles/kg): phentolamine 15, prazosin 2.6, piperoxan 18, yohimbine 2.6. Significance of differences from control (for antagonists) and from level of corresponding blocker (for combination of drugs): *P ≤ 0.1, **P ≤ 0.05, ***P ≤ 0.01, *4P ≤ 0.001. Significance of differences between effects of combination of drugs and effect of clonidine: aP ≤ 0.1, bP ≤ 0.05, cP ≤ 0.01. For control and effect of clonidine, see Table 1.

drowned by a small degree. Conversely, the selective α_2-agonist clonidine protected the mice against hypoxia. This is in agreement with preliminary data on the increase in MLS in a pressure chamber due to clonidine [6, 7]. In the present experiments, clonidine had a marked antihypoxic action on all three models of hypoxia in doses much smaller than those of naphazoline.

The antihypoxic action of clonidine reached a maximum in hypobaric hypoxia in a dose of only 2 μmoles/kg, and in hypercapnic hypoxia in a dose of 20 μmoles/kg. This difference was probably due to the action of additional factors: a reduced pressure and hypercapnia.

With respect to the intensity of their protective action, all the α-adrenoreceptor agonists tested could be arranged in the following order: clonidine > naphtazoline ≥ phenylephrine = norphenylephrine. This order of the α-agonists is evidence that their antihypoxic effect is realized through the α_2-subtype of adrenoreceptors [8-10] and it agrees with the ability of the drugs to reduce the oxygen consumption of the body [2]. To test receptor selectivity of the antihypoxic action of the α-agonists, another technique of biochemical-pharmacological analysis was used, namely blocking of the effect by selective adrenoreceptor antagonists.

Injection of the nonselective α_1- and α_2-adrenoblocker phentolamine lowered the resistance of the animals to hypoxia. This was observed on both models of hypoxic hypoxia investigated (Table 2). Since acute hypoxia, like any other forms of stress, is accompanied by mass mobilization of endogenous catecholamines [11], it can be tentatively suggested

that the decrease in the animals' resistance to hypoxia under the influence of phentolamine was due to the effect of endogenous catecholamines on "free" β-adrenoreceptors. Reciprocal regulation of several metabolic processes in the body by catecholamines through α_2- and β-adrenoreceptors is well known, for example, for cAMP synthesis [1, 9], oxygen consumption [2, 5], lipolysis [8, 10], and several other processes [4, 8].

The selective α_1-antagonist prazosin and α_2-antagonists piperoxan and yohimbine had no regular effect on resistance to hypoxia, although they were used in optimal doses for manifestation of their adrenoblocking action.

The antihypoxic action of clonidine was considerably reduced (by 2-3 times; $P < 0.05$) by preliminary injection of phentolamine, piperoxan or yohimbine — substances capable of blocking adrenoreceptors of the α_2-subtype. The α_1-antagonist prazosin was ineffective or very slightly depressed the antihypoxic effect of clonidine. Consequently, adrenoblocker analysis confirmed the important role of α_2-adrenergic structures in the increase in the resistance of animals to hypoxia caused by adrenoreceptor agonists.

Characteristically the ability of α-agonists to exert an antihypoxic action and to reduce the oxygen consumption of the body correlate closely with one another: For hyperbaric hypoxia $r = +0.869$ ($P < 0.01$), and for hypercapnic hypoxia $r = +0.874$ ($P < 0.01$). In addition, ED_{50} of clonidine for theprotective and anticalorigenic effect are close. This is evidence that the anticalorigenic effect of the α_2-adrenoreceptor agonist leads to a reduction in the oxygen consumption of the tissues and, as a result, it determines their antihypoxic action.

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