THE ANALGESIC EFFECT OF CLOFELIN

A. A. Zaitsev, Yu. D. Ignatov, and A. V. Dmitriev

UDC 615.225.2 + 615.212].03

KEY WORDS: nociceptive reactions; clofelin; naloxone.

Recent investigations have shown that clofelin*, whose hypotensive action is well known, also induces an analgesic effect which is even stronger than that of morphine [10, 12]. It has been suggested that an important role in this effect is played by opioid- and peptidergic mechanisms of action of clofelin [7, 10, 13]. According to one hypothesis, clofelin and its structural analogs may even be the starting point of a new class of analgesics [7].

However, the analgesic activity of clofelin has been studied on models (tail flick, hot plate tests, etc.) which can determine only changes in the pain threshold. Yet we know that an essential role in drug-induced analgesia is played by changes in the emotional-behavioral and autonomic components of pain [3]. Furthermore, the relationship between the hypotensive and analgesic effects of clofelin has not been investigated at all, although it has been shown that the response of animals to nociceptive stimuli can vary substantially with changes in the systemic arterial pressure (BP) [9, 15].

In the investigation described below the effect of clofelin on a combination of emotional behavior and cardiovascular manifestations of nociceptive responses, on the BP level, and on changes in these effects of clofelin when administered together with naloxone, was compared.

EXPERIMENTAL METHOD

Seventeen experiments were carried out on unrestrained cats and rats. Nociceptive stimulation of cats was carried out by stimulating the pulp of the upper canine teeth through chronically implanted electrodes, and in rats by stimulation of the base of the tail through ring electrodes. The emotional-behavioral nociceptive responses were evaluated on scales drawn up previously [1, 5] and BP and (in cats) momentary values of intersystolic intervals (ISI) were recorded [4].

Clofelin (clonidine, from Boehringer Ingelheim) was injected subcutaneously in doses of 0.001 to 4 mg/kg; naloxone (narcan, from Endo Laboratories) was injected intravenously in a dose of 4 mg/kg, and papaverine was injected intravenously in doses of 2 to 8 mg/kg.

EXPERIMENTAL RESULTS

The character of emotional-behavioral and hemodynamic responses to stimulation of the dental pulp in cats was described by the writers previously [4, 5]. Clofelin in doses of 0.001 to 0.01 mg/kg induced slight hypotension but did not affect the structure of the emotional-behavioral reaction or hemodynamic shifts caused by nociceptive stimulation (Table 1, Fig. 1A). Significant depression of manifestations of the emotional and motor response and also of responses of BP and ISI to stimulation of the dental pulp occurred only under the influence of clofelin in a dose or 0.02-0.03 mg/kg, which produced a marked decrease in the initial BP level and bradycardia.

Clofelin in this dose, incidentally, inhibited spontaneous motor activity and the responses of cats to provoking nonnociceptive stimuli (a loud bell, a jet of water aimed at the animal's head). Some cats assumed the lateral position.

*Clonidine.

Department of Pharmacology, Academician I. P. Pavlov First Leningrad Medical Institute. (Presented by Academician of the Academy of Medical Sciences of the USSR A. D. Ado.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 92, No. 12, pp. 690-692, December, 1981. Original article submitted July 15, 1981.

 0007-4888/81/9212-1662$07.50

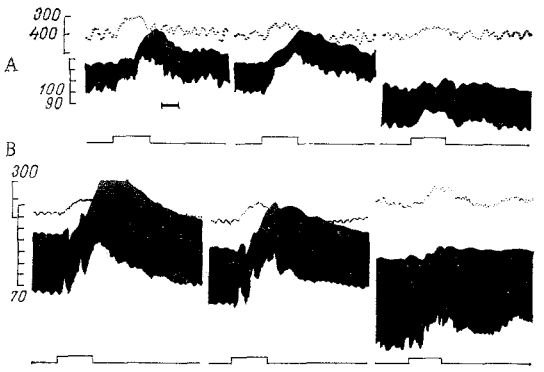

Fig. 1

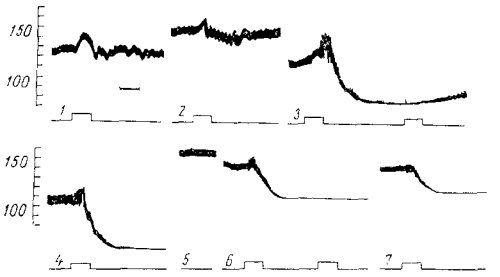

Fig. 2

Fig. 1. Effect of clofelin (A) and papaverine (B) on hemodynamic responses to stimulation of dental pulp in cats. From top to bottom: ISI, BP, time calibration (5 sec), marker of stimulation of dental pulp. From left to right: hemodynamic reactions before injection of drugs, after injection of clofelin in doses of 0.005 and 0.03 mg/kg (A) and of papaverine in doses of 2 and 8 mg/kg (B).

Fig. 2. Effect of clofelin and naloxone on responses of BP to stimulation of base of rat tail. From top to bottom: BP, time marker (5 sec), marker of stimulation of base of tail. 1) Before injection of drugs; 2-4) after injection of clofelin in doses of 1, 2, and 4 mg/kg respectively; 5-7) 2, 5, and 20 min respectively after injection of naloxone (4 mg/kg) preceded by clofelin in a dose of 4 mg/kg.

A combination of depression of emotional behavior and hemodynamic manifestations of nociceptive responses and of marked hypotension also was observed after injection of papaverine — a typical myolytic agent. As the data in Table 1 and Fig. 1B show, it was only after injection of papaverine in doses causing a marked decrease in BP that the whole combination of nociceptive responses was inhibited. Clofelin in a dose of 0.02-0.03 mg/kg and papaverine in a dose of 8 mg/kg, giving equal hypotensive effects, at the same time caused equal inhibition of the nociceptive responses, the response of the cats to nonnociceptive provocative stimuli, and inhibition of the animals' spontaneous behavior.

Correlation between the inhibitory effect of clofelin on nociceptive responses and its hypotensive effect also was found in the experiments on rats (Fig. 2). Clofelin, in a dose of 1 mg/kg, did not lower BP (on the contrary, it caused hypertension) and it reduced the pressor shifts of BP in response to stimulation of the base of the tail only a very little. In the same dose clofelin did not change the emotional-behavioral components of nociceptive responses. Inhibition of behavioral components, like that of the pressor responses of BP, took place under the influence of clofelin in doses of 2 to 4 mg/kg. The analgesic effect of clofelin and its analogs was found to be in these same doses in screening tests [8, 12, 14].

However, as will be clear from Fig. 2, clofelin in a dose of 2 mg/kg and, in particular, of 4 mg/kg, had a powerful hypotensive action and also sharply inhibited the spontaneous behavior of the rats and their responses to provocative stimuli. Nociceptive stimulation after injection of clofelin in doses of 2 to 4 mg/kg was accompanied by a deep and prolonged fall in BP, against the background of which absolute areactivity of the animals to nociceptive stimuli was observed.

Naloxone, in a dose of 4 mg/kg, abolished the hypotensive effect of clofelin (Fig. 2). The spontaneous behavior of the rats and all components of the emotional-behavioral response to stimulation of the base of the tail were fully restored. However, against the background of naloxone the nociceptive responses of BP were not restored and, moreover, the depressor shifts of BP evoked by clofelin were fully preserved.

These investigations showed that clofelin significantly inhibits nociceptive responses in doses giving rise to considerable hypotension and worsening of the animals' functional state. Naloxone, by abolishing clofelin hypotension, at the same time restores spontaneous

TABLE 1. Effect of Clofelin and Papaverine on Background Hemodynamic Indices and Structure of Emotional-Behavioral Response to Stimulation of Dental Pulp

Drug	Dose, mg/kg	BP, mm Hg	ISI, msec	Levels of nociceptive response and their features								
				1		2			3			
				reflex opening of mouth	licking	piloerection	changes in respiration	uncontrolled movements	defensive movements	scratching	running	vocal response
Control		102±4	411±12	1,0	1,4	2,4	2,2	2,4	6,3	6,4	8,8	7,8
Clofelin												
	0,001—0,005	103±3	423±13	1,0	1,4	2,2	2,2	2,4	6,4	6,6	8,6	8,0
	0,0075—0,01	90±3*	441±16	1,0	1,6	2,4	2,2	2,6	6,8	6,8	9,2	8,2
	0,02—0,03	76±5*	503±14*	1,0	1,6	2,8	2,8	3,4*	7,8*	7,6*	11,4*	9,8*
Control		105±3	434±15	1,0	1,2	2,6	2,4	2,4	5,8	6,2	8,4	8,2
Papaverine												
	2	98±6	422±18	1,0	1,2	2,4	2,6	2,4	6,2	6,4	8,8	8,6
	5	85±4*	403±16	1,0	1,4	2,6	2,8	2,8	7,8*	6,8	9,8	9,2
	8	71±4*	374±19	1,0	1,4	3,2*	3,4*	2,8	9,8*	9,8	12,4	—

Legend. Numbers in column "levels of nociceptive response and their features" denote mean intensity of stimulation of dental pulp (in thresholds) needed to cause the appearance of the given feature; minus sign denotes that the feature was not produced; an asterisk indicates a significant change in the index compared with the control by Student's test (for hemodynamic indices) and by van der Waerden's criterion (for behavioral features) at the $P < 0.005$ level.

behavior and the emotional-behavioral but not the hemodynamic response of the animals to nociceptive stimulation.

The difference between the results of this investigation and data in the literature on the true analgesic effect of clofelin can be explained, in our opinion, by the fact that other workers used predominantly screening tests. It will be evident that methods of assessing analgesia based on changes in the pain threshold alone are insufficiently informative and may give rise to mistaken ideas on the efficacy of drugs during nociceptive stimulation [2], more especially because the present results confirmed observations [9, 15] indicating a significant change in the response of animals to aversive stimuli during shifts of the systemic BP.

The dissociation discovered in the action of naloxone on nociceptive behavioral manifestations, on the one hand, and on the response of BP on the other hand, makes the role of peptidergic mechanisms in the "analgesic" effect of clofelin questionable. The absence of antagonism between the analgesic effect of clofelin and naloxone is emphasized in reports of other investigations also [10, 14]. Crossed tolerance between morphine and clofelin likewise has not been found[14].

Meanwhile the abolition by naloxone of the hypotensive effect of clofelin probably indicates the presence of an opioidergic component in the hypotensive action of the adrenomimetic drug. It is suggested that opiate receptors participate in both the cardiovascular and the sedative effects of clofelin [6]. Since the use of labeled clofelin has demonstrated that the compound has no direct effect on opiate receptors [11], it can be postulated that the liberation of endogenous opiates under the influence of clofelin takes place indirectly through central adrenergic structures.

The results thus do not confirm the existing view that clofelin has a true (morphine-like) analgesic effect. The inhibition of nociceptive responses by clofelin in "hypotensive" doses is probably the result of lowering of the reactivity of normotensive animals to aversive stimuli, which is accompanied by (or associated with) marked hypotension.

LITERATURE CITED

1. B. V. Andreev, Yu. N. Vasil'ev, V. P. Kosinskii, et al., in: Neuropharmacological Aspects of Emotional Stress and Drug Dependence [in Russian], Leningrad (1978), pp. 38-48.
2. Yu. N. Vasil'ev and Yu. D. Ignatov, Farmakol. Toksikol., No. 6, 676 (1976).
3. A. V. Val'dman, The Neuropharmacology of Narcotic Analgesics [in Russian], Moscow (1972).
4. A. A. Zaitsev, A. V. Dmitriev, Yu. D. Ignatov, et al., Byull. Éksp. Biol. Med., No. 10, 391 (1980).
5. Yu. D. Ignatov and A. V. Dmitriev, Byull. Éksp. Biol. Med., No. 10, 1158 (1976).
6. H. Karppanen, in: Neurotransmitters and the Mechanism of Action of Neurotropic and Cardiovascular Drugs [in Russian], Moscow (1979), p. 9.
7. D. Atlas and S. L. Sabol, Biophys. Res. Commun., 94, 924 (1980).
8. S. G. Dennis, R. Melzack, S. Gutman, et al., Life Sci., 26, 1247 (1980).
9. B. R. Dworkin, R. J. Filewich, N. E. Miller, et al., Science, 205, 1299 (1979).
10. S. Fielding, J. Wilker, M. Hynes, et al., J. Pharmacol. Exp. Ther., 207, 899 (1978).
11. K. Golembiowska-Nikitin, A. Pilc, and J. Vetulani, J. Pharm. Pharmacol., 32, 70 (1980).
12. J. J. Lipman and P. S. J. Spencer, Neuropharmacology, 18, 731 (1979).
13. G. Paalzow, Arch. Pharm. Exp. Pathol., 304, 1 (1978).
14. T. G. Spaulding, S. Fielding, J. J. Venafro, et al., Eur. J. Pharmacol., 58, 19 (1979).
15. N. Zamir and M. Segal, Brain Res., 160, 170 (1979).

COMPARATIVE STUDY OF SUBSTANCE P AND ITS FRAGMENTS: ANALGESIC PROPERTIES, EFFECT ON BEHAVIOR, AND MONOAMINERGIC PROCESSES

V. E. Kluša, N. A. Abissova,
R. K. Muceniece, Sh. V. Svirskis,
M. Bienert, and A. Lipkowski

UDC 615.357.814.32.017:615.214

KEY WORDS: substance P and its fragments; analgesia; behavior; biogenic monoamines.

Despite much evidence of the mediator or neuromodulator role of substance P in the CNS [6, 9] and data showing the broad spectrum of its action [10, 11, 15], it is not yet known exactly what are the functions of substance P in the body or how it interacts with other central ligands. It has also been postulated on the basis of data showing the neuromodulator action of certain C-terminal fragments of peptide hormones and their role in homeostatic processes of behavior [1, 2] that various fragments of substance P, its possible hydrolysis products, may also participate in the regulation of the behavioral and monoaminergic processes of the brain.

In the investigation described below a comparative study was made of the behavioral and analgesic effects of substance P and of certain of its fragments, and also of their effect on the content of biogenic monoamines (BM) in the rat brain.

EXPERIMENTAL METHOD

The peptides — substance P (SP) (H-Arg-Pro-Lys-Pro-Gln-Gln-Phe-Phe-Gly-Leu-Met-NH_2) and its C-terminal fragments — a heptapeptide (SP 5-11), tetrapeptide (SP 8-11), and tripeptide (SP 9-11) — were synthesized at the Institute for the Study of Physiologically Active Substances (East Berlin); the dipeptide (SP 10-11) was synthesized at Warsaw University.

Laboratory of Molecular Biology and Pharmacology of Peptides, Institute of Organic Synthesis, Academy of Sciences of the Latvian SSR, Riga. Institute for the Study of Physiologically Active Substances, East Berlin. Warsaw University. (Presented by Academician of the Academy of Medical Sciences of the USSR A. D. Ado.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 92, No. 12, pp. 692-694, December, 1981. Original article submitted July 15, 1981.

0007-4888/81/9212-1665$07.50