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Anxiogenic-like effect of the neuropeptide Y Y_1 receptor antagonist BIBP3226: antagonism with diazepam

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Abstract

The effect of the novel non-peptide neuropeptide Y Y_1 receptor antagonist BIBP3226, N^2-(diphenylacetyl)-*N*-[(4-hydroxy-phenyl)methyl]-D-arginine amide, on exploratory behaviour of rats in the elevated plus-maze was studied. BIBP3226 (0.5 and 5 μg, i.c.v.) induced an anxiogenic-like effect at the higher dose tested. This effect was antagonised by diazepam (0.5 mg/kg). The anxiogenic-like effect of BIBP3226 was not related to a decrease in general locomotor activity. These findings support the hypothesis that neuropeptide Y Y_1 receptor subtype is involved in anxiety regulation.

Keywords: Neuropeptide Y Y_1 receptor; BIBP3226; Elevated plus-maze

Neuropeptide Y has been implicated in the regulation of feeding, cardiovascular regulation and emotional behaviour (Wahlestedt and Reis, 1991). Several behavioural studies have found that the effects of exogenously administered neuropeptide Y are similar to conventional anxiolytic drugs (Wahlestedt and Reis, 1991). Studies with different neuropeptide Y fragments (Broqua et al., 1995) and antisense oligodeoxynucleotides complementary to neuropeptide Y Y_1 receptor mRNA (Wahlestedt et al., 1993) lend support to the hypothesis that the anxiolytic-like effects of neuropeptide Y in animal models of anxiety are mediated through neuropeptide Y Y_1 receptors. While the anxiolytic-like effects of exogenously administered neuropeptide Y are well established, the involvement of endogenous neuropeptide Y in the regulation of exploratory behaviour has not been confirmed by pharmacological means. A few proposed neuropeptide Y receptor antagonists have been synthesised, but none of them has high potency and selectivity for neuropeptide Y Y_1 receptor (Wahlestedt and Reis, 1991). Recently, a novel non-peptide neuropeptide Y Y_1 receptor antagonist, BIBP3226 (N^2-(diphenylacetyl)-*N*-[(4-hydroxy-phenyl)methyl]-D-arginine amide), was shown to be highly selective for neuropeptide Y Y_1 receptors in binding studies and functional assays (Jacques et al., 1995). The aim of the present study was to investigate the effect of BIBP3226 on exploratory behaviour and to test the hypothesis that blockade of neuropeptide Y Y_1 receptors in vivo will lead to appearance of anxiety signs.

Male Wistar rats (270–350 g), purchased from Grindex, Latvia, were kept under standard laboratory conditions and implanted with chronic injection cannulae aimed at lateral ventricles. The behavioural experiments were started 1 week after surgery. BIBP3226 (Thomae, Germany) was injected i.c.v. 20 min and diazepam (0.5 mg/kg i.p.) 30 min before the test. The plus-maze and open field test design was as described elsewhere (Harro et al., 1990) but 1 cm ledges were added to open arms of the maze.

As shown in Table 1, BIBP3226 at the dose of 5 μg caused an anxiogenic-like effect while the lower dose was ineffective. In the second experiment we found that the dose of diazepam (0.5 mg/kg) that itself had no effect on exploratory parameters as a single treatment completely blocked the anxiogenic-like effect of the neuropeptide Y Y_1 receptor antagonist. Immediately after the plus-maze test the rats were subjected to an open-field test for 4 min. Since the overall locomotory activity was similar in all treatment groups (data not shown), the anxiogenic profile of BIBP3226 and its antagonism with diazepam cannot be interpreted as the changes in general locomotor activity

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Table 1
Effect of BIBP3226 on exploratory behaviour of rats in the elevated plus-maze

Drug/dose	Time spent in open part	Number of line crossings	Open arm entries	Ratio open/total arm entries (%)
Saline	108 ± 14	18.6 ± 2.4	2.1 ± 0.3	29.9 ± 1.8
BIBP3226 0.5 μg i.c.v.	110 ± 14	18.9 ± 2.8	2.9 ± 0.6	36.1 ± 5.7
BIBP3226 5 μg i.c.v.	66 ± 12	11.3 ± 2.4	0.7 ± 0.4	10.3 ± 5.7 [a]
Saline + saline	114 ± 16	26.2 ± 3.5	3.6 ± 0.8	35.2 ± 6.3
Saline + BIBP3226 5 μg i.c.v.	43 ± 12 [a]	8.6 ± 2.9 [a]	0.4 ± 0.3 [a]	4.3 ± 2.8 [a]
Diazepam 0.5 mg/kg + saline	115 ± 17	26.2 ± 5.1	2.6 ± 0.7	25.4 ± 5.9
Diazepam 0.5 mg/kg + BIBP 5 μg i.c.v.	94 ± 25	28.0 ± 9.0 [b]	4.0 ± 1.4 [b]	37.7 ± 11.3 [b]

The results are presented as mean values ± S.E.M.
[a] $P < 0.05$ BIBP3226 vs. saline.
[b] $P < 0.05$ diazepam + BIBP3226 vs. saline + BIBP3226-treated rats, Fisher's PLSD after significant one-way ANOVA..

due to, e.g., a neurotoxic effect. The overall activity of the animals was higher in the second experiment when i.p. injection preceded i.c.v. injection. Such an increase in exploratory behaviour may occur when animals are handled or exposed to other stimuli before the test (Hogg, 1996).

Our findings support the hypothesis that endogenous neuropeptide Y could reduce neophobia/anxiety by activating neuropeptide Y Y_1 receptors. The benzodiazepine diazepam, a clinically effective anti-anxiety agent, counteracted the anxiogenic-like effect of BIBP3226. This indicates that the neuropeptide Y-ergic neurotransmission might be closely coupled to the GABA-ergic system. Neuroanatomically this interaction may involve cortical areas since many cortical neurons immunoreactive for neuropeptide Y also contain GABA (Hendry et al., 1984).

It has been proposed that neuronal release of neuropeptide Y occurs during high bursts of firing (Lundberg et al., 1986) which occurs when animals are exposed to a novel environment. Neuropeptide Y itself or other endogenous ligands acting at neuropeptide Y Y_1 receptors might be essential for an adaptation to a new situation and for expression of normal exploratory behaviour. Therefore, it is possible that blockade of neuropeptide Y Y_1 receptors with BIBP3226 disturbed the adaptational processes and led to expression of anxiety.

In conclusion, these results confirm the involvement of neuropeptide Y Y_1 receptor in the regulation of exploratory behaviour and (or) anxiety in the rat. BIBP3226 appears to be a useful compound to study the role of neuropeptide Y Y_1 receptors in emotional processes.

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References

Broqua, P., J.G. Wettstein, M.N. Rocher, B. Gauthier-Martin and J.L. Junien, 1995, Behavioral effects of neuropeptide Y receptor agonists in the elevated plus-maze and fear-potentiated startle procedures, Behav. Pharmacol. 6, 215.

Harro, J., A. Lang and E. Vasar, 1990, Long-term diazepam treatment produces changes in cholecystokinin receptor binding in rat brain, Eur. J. Pharmacol. 180, 77.

Hendry, S.H.C., E.G. Jones, J. DeFelipe, D. Schmechel, C. Brandon and P. Emson, 1984, Neuropeptide-containing neurons of the cerebral cortex are also GABAergic, Proc. Natl. Acad. Sci. USA 81, 6526.

Hogg, S., 1996, A review of the validity and variability of the elevated plus-maze as an animal model of anxiety, Pharmacol. Biochem. Behav. 54, 21.

Jacques, D., A. Cadieux, Y. Dumont and R. Quirion, 1995, Apparent affinity and potency of BIBP3226, a non-peptide neuropeptide Y receptor antagonist, on purported neuropeptide Y Y_1, Y_2 and Y_3 receptors, Eur. J. Pharmacol. 278, R3.

Lundberg, J.M., A. Rudehill, E. Sollevi, E. Theodorsson-Norheim and B. Hamberger, 1986, Frequency- and reserpine dependent chemical coding of sympathetic transmission: differential release of noradrenaline and neuropeptide Y from pig spleen, Neurosci. Lett. 63, 96.

Wahlestedt, C. and D.J. Reis, 1991, Neuropeptide Y-related peptides and their receptors – are the receptors potential therapeutic drug targets?, Annu. Rev. Pharmacol. Toxicol. 32, 309.

Wahlestedt, C., E.M. Pich, G.F. Koob, F. Yee and M. Heilig, 1993, Modulation of anxiety and neuropeptide Y-Y_1 receptors by antisense oligonucleotides, Science 259, 528.