ELSEVIER

European Journal of Pharmacology 321 (1997) R11–R13

ejp

Rapid communication

Potentiation of fluoxetine-induced penile erections by combined blockade of 5-HT_{1A} and 5-HT_{1B} receptors

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Received 9 January 1997; accepted 14 January 1997

Abstract

The serotonin reuptake inhibitor, fluoxetine (10.0 mg/kg, s.c.), elicited penile erections in rats. Selective blockade of 5-HT_{1A} autoreceptors with WAY 100,635 ((*N*-{2-[4-(2-methoxyphenyl)-1-piperazinyl]ethyl}-*N*-(2-pyridinyl) cyclo-hexanecarboxamide) (0.16 mg/kg, s.c.), or of 5-HT_{1B} autoreceptors with GR 127,935 (*N*-[4-methoxy-3-(4-methylpiperazin-1-yl)phenyl]-2′-methyl-4′-(5-methyl-1,2,4-oxadiazol-3-yl)biphenyl-4-carboxamide) (2.5 mg/kg, s.c.), slightly (1.5- to 2-fold) increased fluoxetine-induced penile erections. However, conjoint administration of WAY 100,635 and GR 127,935 markedly (5-fold) potentiated induction of penile erections by fluoxetine. Penile erections were abolished by the novel 5-HT_{2C} receptor antagonist, SB 206,553 (5 methyl-1-(3-pyridil-carbamoyl)-1,2,3,5-tetrahydropyrrolo[2,3-*f*]indole). These data provide functional evidence for redundancy in autoreceptor control of 5-HT release. Combined blockade of 5-HT_{1A} and 5-HT_{1B} autoreceptors markedly enhances the actions of serotonin reuptake inhibitors.

Keywords: 5-HT_{2C} receptor; Fluoxetine; Antidepressant

An increase in postsynaptic, extracellular 5-hydroxytryptamine (5-HT, serotonin) levels contributes to the antidepressant properties of fluoxetine and other serotonin reuptake inhibitors (Artigas et al., 1996). However, serotonin reuptake inhibitors simultaneously increase 5-HT levels at inhibitory dendritic 5-HT_{1A} autoreceptors and terminal 5-HT_{1B} receptors, actions which may compromize their clinical efficacy (Artigas et al., 1996). The progressive desensitization of these autoreceptors may underlie the delay to their onset of action, and the 5-HT_{1A} receptor antagonist, (−)-pindolol, increases the antidepressant actions of serotonin reuptake inhibitors in man by mimicking this process of 5-HT_{1A} autoreceptor inactivation (Artigas et al., 1996). Further, in rats, the (separate) blockade of 5-HT_{1A} autoreceptors with (−)-pindolol or WAY 100,635 ((*N*-{2-[4-(2-methoxyphenyl)-1-piperazinyl]ethyl}-*N*-(2-pyridinyl) cyclo-hexanecarboxamide) (a selective antagonist), or of 5-HT_{1B} autoreceptors with GR 127,935 (*N*-[4-methoxy-3-(4-methylpiperazin-1-yl)phenyl]-2′-methyl-4′-(5-methyl-1,2,4-oxadiazol-3-yl)biphenyl-4-carboxamide) (a selective antagonist), enhances the influence of serotonin reuptake inhibitors upon postsynaptic 5-HT levels (see Artigas et al., 1996). However, their influence is limited, probably owing to mutual redundancy in the roles of 5-HT_{1A} and 5-HT_{1B} autoreceptors. Indeed, conjoint blockade of both 5-HT_{1A} and 5-HT_{1B} autoreceptors by combined application of WAY 100,635 and GR 127,935 synergistically potentiates the influence of fluoxetine upon 5-HT levels (Gobert et al., 1997). However, functional evidence for a potentiation in the actions of serotonin reuptake inhibitors is, to date, lacking. The present study addressed this issue employing penile erections, a response reflecting activation of postsynaptic 5-HT_{2C} receptors (Berendsen et al., 1990).

Penile erections were measured as previously (Jenck et al., 1993) in male Wistar rats (140–160 g) individually placed in Plexiglas cages behind which a mirror was placed to facilitate observation. Penile erections were measured over 40 min commencing immediately following treatment with fluoxetine. Rats were pretreated 20 min before fluoxetine with the antagonist. Drugs were dis-

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0014-2999/97/$17.00
PII S0014-2999(97)00050-2

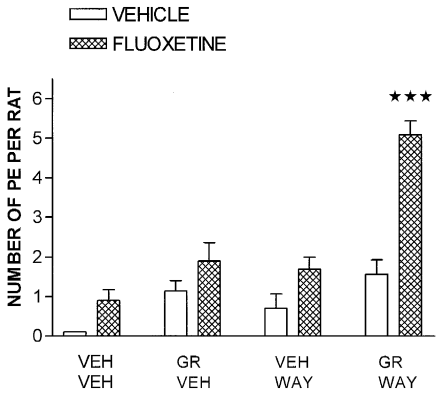

Fig. 1. Influence of the 5-HT_{1A} autoreceptor antagonist, WAY 100,635 (0.16 mg/kg, s.c.), and of the 5-HT_{1B} autoreceptor antagonist, GR 127,935 (2.5 mg/kg, s.c.), upon the induction of penile erections by fluoxetine (10.0 mg/kg, s.c.). Data are means ± S.E.M. $n = 7–10$ per column. ANOVA as follows: $F(7,67) = 22.9$, $P < 0.001$. Asterisks indicate the significance of differences of WAY 100,635/GR 127,935/fluoxetine to vehicle/vehicle/fluoxetine and to WAY 100,635/GR 127,935/vehicle values in the Newman-Keuls test, $P < 0.001$. Chi analysis of numbers of rats per treatment group showing ≥ 1 penile erections yielded the following significant differences ($P < 0.05$): vehicle/vehicle/fluoxetine vs. vehicle/vehicle/vehicle; vehicle/GR 127,935/vehicle and WAY 100,635/GR 127,935/vehicle vs. vehicle/vehicle/vehicle and WAY100,635/GR 127,935/fluoxetine vs. vehicle/vehicle/fluoxetine, WAY 100,635/vehicle/fluoxetine and vehicle/GR 127,935/fluoxetine and WAY 100,635/GR 127,935/vehicle.

solved in sterile water and injected s.c. Doses are in terms of the base.

Fluoxetine elicited a significant penile erection response (Fig. 1). WAY 100,635 and GR 127,935, injected either alone or together, also provoked a mild penile erection response (Fig. 1). Administered separately, WAY 100,635 and GR 127,935 tended (though not significantly) to potentiate the action of fluoxetine. However, their co-administration resulted in a marked (5-fold) facilitation of the action of fluoxetine. The 5-HT_{2C} receptor antagonist, SB 206,553 (5 methyl-1-(3-pyridil-carbamoyl)-1,2,3,5-tetrahydropyrrolo[2,3-*f*]indole) (2.5 mg/kg, s.c., – 30 min) abolished these penile erections: vehicle/WAY 100,635/GR 127,935/fluoxetine, $n = 8$, 3.8 ± 0.5 penile erections vs. SB 206,553/WAY 100,635/GR 127,935/fluoxetine, $n = 8$, 0.1 ± 0.1, $P < 0.001$, Student's two-tailed *t*-test. Induction of penile erections by the selective 5-HT_{2C} agonist, RO 60,0175 ((*S*)-2-(6-chloro-5-fluoroindol-1-yl)-1-methylethylamine) fumarate) (0.63 mg/kg, s.c.) was unaffected by WAY 100,635 (0.16 mg/kg, s.c.) and GR 127,935 (2.5 mg/kg, s.c.): vehicle/vehicle/RO 60,0175, $n = 8$, 1.6 ± 0.4 penile erections vs. WAY 100,635/GR 127,935/fluoxetine, $n = 8$, 2.1 ± 0.6 ($P > 0.05$).

As reported previously, fluoxetine elicited penile erections in rats; this action likely reflects an increase in extracellular 5-HT levels and subsequent activation of postsynaptic 5-HT_{2C} receptors (Berendsen et al., 1990; Simon et al., 1993). The selective or combined administration of WAY 100,635 and/or GR 127,935 also elicited a modest penile erection response, presumably due to an increase in spontaneous 5-HT release by blockade of 5-HT_{1A} and/or 5-HT_{1B} autoreceptors (Artigas et al., 1996; Gobert et al., 1997). In a recent study, the 5-HT_{1A} autoreceptor antagonist, (−)-tertatolol, potentiated fluoxetine-induced penile erections (Simon et al., 1993). However, no biochemical evidence for an increase in synaptic levels of 5-HT was presented and the actions of (−)-tertatolol are difficult to interpret since it also behaves as a partial agonist at 5-HT_{1B} sites and as a potent antagonist at β-adrenoceptors (see Simon et al., 1993). Herein, WAY 100,635, which is highly selective for 5-HT_{1A} sites (see Artigas et al., 1996), only slightly potentiated the action of fluoxetine. Similarly, GR 127,935 only tended to enhance the action of fluoxetine. In line with these findings, in a parallel study (Gobert et al., 1997), each of these antagonists alone produced only a mild (approx. 2-fold) increase in the influence of fluoxetine upon extracellular 5-HT levels. However, co-administration of WAY 100,635 and GR 127,935 resulted in a marked (6-fold) increase in extracellular levels of 5-HT (Gobert et al., 1997). This observation corresponds remarkably well to the marked 5-fold increase in penile erections elicited by fluoxetine in the presence of WAY 100,635 and GR 127,935 (Fig. 1). Further, the novel 5-HT_{2C} receptor antagonist, SB 206,553 (Kennett et al., 1996), blocked these penile erections, confirming their mediation by 5-HT_{2C} receptors. Inasmuch as WAY 100,635 and GR 125,935 did not modify induction of penile erections by the selective 5-HT_{2C} agonist RO 60,0175 (Martin et al., 1995), an interaction between postsynaptic 5-HT_{2C} and 5-HT_{1A}/5-HT_{1B} receptors is not involved in the potentiation of fluoxetine-induced penile erections. Further, fluoxetine behaves as a weak antagonist at 5-HT_{2C} receptors (Pälvimäki et al., 1996).

The present observations provide, then, a functional correlate to our dialysis studies in freely moving rats (Gobert et al., 1997) in demonstrating that the actions of fluoxetine are more powerfully reinforced by the simultaneous as compared to separate blockade of 5-HT_{1A} and 5-HT_{1B} autoreceptors. This difference likely reflects the fact that, when 5-HT_{1A} sites are blocked, 5-HT_{1B} receptors remain available to 5-HT and vice versa. Further, although a minor population of dendritic 5-HT_{1D} receptors may also inhibit the activity of serotoninergic neurones, these are also susceptible to blockade by GR 127,935 (see Artigas et al., 1996; Gobert et al., 1997). Interestingly, actions at 5-HT_{2C} receptors, which mediate penile erections, have been implicated in the actions of serotonin reuptake inhibitors and other antidepressant drugs (Jenck et al., 1993).

In conclusion, the present data provide the first functional evidence that combined blockade of 5-HT_{1A} and 5-HT_{1B} autoreceptor actions markedly potentiates the actions of serotonin reuptake inhibitors. Conjoint antagonism of 5-HT_{1A} and 5-HT_{1B} autoreceptors may offer an attractive strategy for reinforcing the antidepressant properties of serotonin reuptake inhibitors.

References

Artigas, F., Romero, L., De Montigny, C., Blier, P., 1996. Acceleration of the effect of selected antidepressant drugs in major depression by 5-HT_{1A} receptor antagonists. Trends Pharmacol. Sci. 19, 378–383.

Berendsen, H.H.G., Jenck, F., Broekkamp, C.L.E., 1990. Involvement of 5-HT_{1C} receptors in drug-induced penile erections in rats. Psychopharmacology 101, 57–66.

Gobert, A., Rivet, J.-M., Cistarelli, L., Millan, M.J., 1997. Potentiation of the fluoxetine-induced increase in dialysate levels of serotonin (5-HT) in the frontal cortex of freely-moving rats by combined blockade of 5-HT_{1A} and 5-HT_{1B} receptors with WAY 100,635 and GR 127,935. J. Neurochem. 68 (in press).

Jenck, F., Moreau, J.-L., Mutel, V., Martin, J.R., Haefely, W.E., 1993. Evidence for a role of 5-HT_{1C} receptors in the antiserotonergic properties of some antidepressant drugs. Eur. J. Pharmacol. 231, 223–226.

Kennett, G.A., Wood, M.D., Bright, F., Cilia, J., Piper, D.C., Gager, T., Thomas, D., Baxter, G.S., Forbes, I.T., Ham, P., Blackburn, T.P, 1996. In vitro and in vivo profile of SB 206,553, a potent 5-HT_{2C} /5-HT_{2B} receptor antagonist with anxiolytic-like properties. Br. J. Pharmacol. 117, 427–434.

Martin, J.R., Bös, M., Jenck, F., Köhler, C., Moreau, J.-L., Sleight, A.J., Stadler, H., Wichmann, J., 1995. 5-HT_{2C} receptor agonists and antagonists in animal models of anxiety. Eur. Neuropsychopharmacol. Abstr. 5, S4–17.

Pälvimäki, E.P., Roth, B.L., Majasuo, H., Laakso, A., Kuoppamäki, M., Syvälahti, E., Hietala, J., 1996. Interactions of selective serotonin reuptake inhibitors with the serotonin 5-HT_{2C} receptor. Psychopharmacology 126, 234–240.

Simon, P., Bertrand, J., Costentin, J., 1993. 5-HT_{1A} receptor blockade increases penile erections induced by indirect serotonin agonists. NeuroReport 5, 229–230.