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The NK-3 tachykinin receptor agonist senktide elicits 5-HT-mediated behaviour following central or peripheral administration in mice and rats

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The behavioural effects of the selective NK-3 tachykinin receptor agonist senktide were studied following intracisternal and subcutaneous administration in rodents. Behavioural manifestations of 5-hydroxytryptaminergic stimulation, including head twitches (mice), wet dog shakes (rats), forepaw treading, flat body posture, hindlimb splaying and Straub tail, were seen following the intracisternal (0.01–1.2 nmol) or subcutaneous (0.1–2.4 μmol kg^{-1}) administration of senktide in both species. We conclude that stimulation of NK-3 receptors stimulates 5-hydroxytryptaminergic pathways in rodent brain, and that senktide may cross the blood-brain barrier in biologically significant amounts.

Introduction Little is known about the behavioural consequences of interactions between substance P (SP) and 5-hydroxytryptamine (5-HT) in the central nervous system, although anatomical (Chan-Palay *et al.*, 1978; Hokfelt *et al.*, 1978; Neekers *et al.*, 1979; Magoul *et al.*, 1986) and pharmacological (Reubi *et al.*, 1978; Forchetti *et al.*, 1982; Reisine *et al.*, 1982) relationships have been recognized for some time. We have previously demonstrated (Stoessl *et al.*, 1987) that senktide, a synthetic peptide highly selective for NK-3 tachykinin receptors (Wormser *et al.*, 1986) elicits a 5-HTergic behavioural syndrome when administered intracisternally (i.c.s.) to the mouse. Senktide-induced head twitches were blocked by the 5-HT$_2$ antagonists ketanserin and ritanserin, while forepaw treading elicited by senktide was attenuated by the 5-HT$_1$ antagonist (−)-pindolol. The non-selective 5-HT antagonist methysergide blocked head twitches and forepaw treading (Stoessl *et al.*, 1987). In this communication, we describe the same behavioural effects in the mouse following subcutaneous (s.c.) administration, and also an analogous syndrome in the rat after s.c. or i.c.s. administration.

Methods Female CF-1 mice, 18–20 g, and male Sprague-Dawley rats, 230–260 g, were obtained from Bantin & Kingman, Hull. Animals were housed in groups of 5 on a 12 h light-dark cycle (lights on at 07 h 00 min) and allowed standard laboratory food pellets and tap water *ad libitum*.

Senktide (M_r 842) was synthesized, purified and characterized at Merck Sharp & Dohme Research Laboratories, Terlings Park. The solid was dissolved in a minimum quantity of dimethylsulphoxide (DMSO) and dilutions made in 0.9% saline. The maximum final concentration (v/v) of DMSO was 1% (intracisternal administration) or 5% (peripheral administration).

For intracisternal injection, rats were anaesthetized with halothane (4%) and nitrous oxide (70%) and then placed in a Kopf stereotaxic frame. With the head hyperflexed, the post-occipital groove was palpated and a cannula was introduced to a depth of 5 mm: 50 μl of senktide or vehicle were then infused manually from a 50 μl Hamilton syringe and the cannula was withdrawn. The animals were then allowed to recover from the anaesthetic and placed in perspex observation boxes. Behaviour was recorded for 60 min, with a BBC microcomputer system which generated latency, frequency and duration data for each behaviour of interest.

A range of concentrations of senktide or vehicle were also injected s.c. in the neck in a volume of 10 ml kg^{-1} (mice) or 1 ml kg^{-1} (rats). The animals were then immediately placed in perspex observation boxes and behaviour recorded for 30 min (mice) or 60 min (rats) as described above.

Results We have previously reported a dose-dependent head twitch response in mice, maximal at 0.1–0.6 nmol, following i.c.s. senktide (Stoessl *et al.*, 1987). At higher doses, head twitching declined and other behaviour, including forepaw treading, hindlimb splaying, tail rattling and immobility was recruited. The latency (mean ± s.e. mean) for the head twitch response was 94 ± 6 s. In rats, i.c.s. senktide elicited wet dog shakes (the behavioural equivalent of head twitches in mice, see Handley & Singh,

1986) with a latency of 189 ± 48 s. The maximum response rate was seen at 0.1 nmol. At higher doses, there was a marked attenuation of wet dog shakes, as other behaviour, including forepaw treading, chewing mouth movements, flat body posture, Straub tail and hindlimb splaying supervened (Figure 1). The degree of forepaw treading at 1.2 nmol was variable, with some animals showing almost continuous treading from 40 min after the injection, lasting for an hour or longer.

Subcutaneously administered senktide (0.1–1.2 μmol kg^{-1}) produced head twitches in mice and wet dog shakes in rats, as well as forepaw treading in both species (Figure 1). In addition, the other behavioural manifestations seen following i.c.s. administration, i.e. tail rattling (mice), flat body posture and Straub tail (rats) and hindlimb splaying (mice and rats) were observed. In mice, the head twitch response was maximal at 0.6–1.2 μmol kg^{-1}, with a latency of 87 ± 14 s. Wet dog shakes in rats were maximal at 1.2 μmol kg^{-1}, with a latency of 168 ± 24 s.

Discussion These results demonstrate that centrally or peripherally administered senktide elicits 5-HTergic behaviours in both mice and rats. 5-HTergic mediation of senktide-induced behavioural responses is confirmed by our previous demonstration that they are attenuated by selective 5-HT antagonists (Stoessl *et al.*, 1987). The mechanism for this interaction is as yet undetermined. SP has been shown to elicit 5-HT release both *in vivo* and *in vitro* (Reubi *et al.*, 1978; Forchetti *et al.*, 1982; Reisine *et al.*, 1982), an effect possibly mediated by inhibition of 5-HT terminal autoreceptors (Mitchell & Fleetwood-Walker, 1981). Recently, the endogenous NK-3 tachykinin agonist neurokinin B has been shown to enhance 5-HT release from rat frontal cortical slices (Solti & Bartfai, 1987). This response was shown to occur at a site different from that involved in SP-induced 5-HT release. Finally, tachykinins may enhance the behavioural response to endogenous 5-HT by increasing the affinity of 5-HT binding (Agnati *et al.*, 1983).

It seems unlikely that the observed behavioural effects were peripherally mediated, since the syndrome observed following s.c. administration was very similar to that seen following central administration in the rat, and virtually identical in the mouse. Furthermore, other evidence (for review, see Curzon, 1988) suggests that these behaviours are mediated by activation of central 5-HT pathways. Our observations suggest that senktide probably crosses the blood-brain barrier to a biologically significant extent.

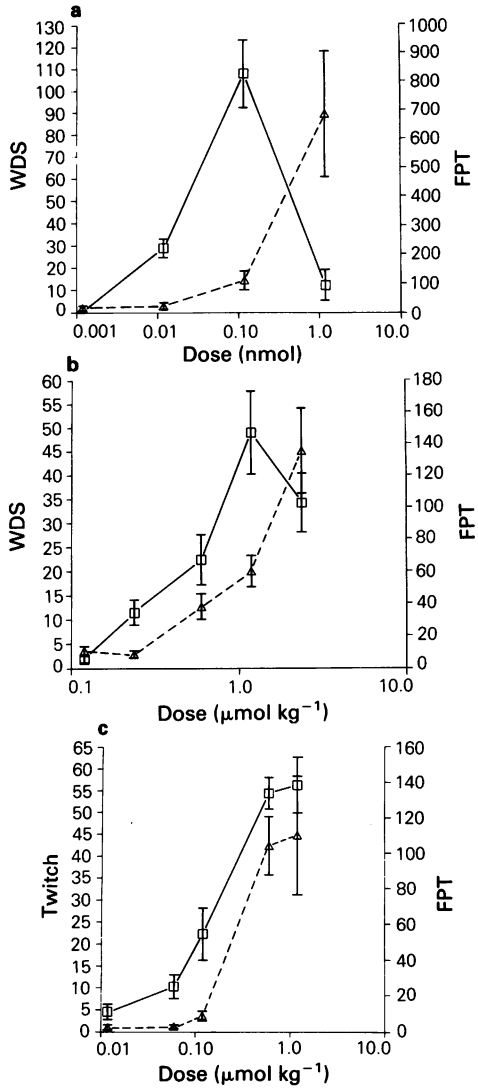

Figure 1 Dose-response curves for wet dog shakes (WDS, frequency, □) and forepaw treading (FPT, duration in seconds, △) occurring over a 60 min period following intracisternal (a) or subcutaneous (b) administration of senktide in the rat. Each point represents the mean of 5–10 animals; vertical lines show s.e. mean. (c) Dose-response curves for head twitches (frequency, □) and forepaw treading (FPT, duration in seconds, △) occurring over a 30 min period following subcutaneous administration of senktide in the mouse.

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