Anxiolytic- and antidepressant-like effects of non-peptide vasopressin V_{1b} receptor antagonists

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Abstract. The observation that the nonapeptide vasopressin is critical for adaptation of the hypothalamo-pituitary-adrenal axis during stress through its ability to potentiate the stimulatory effect of corticotropin-releasing factor, taken together with the identification of vasopressin receptors (e.g. V_{1b}) in limbic structures has led to the idea that this peptide may provide a good opportunity for pharmacological treatment of stress-related disorders. The availability of the first orally active non-peptide V_{1b} receptor antagonist, SSR149415, has allowed us to verify this hypothesis. Studies in rodents showed that SSR149415 is able to attenuate stress-related behaviors. While the antidepressant-like activity of the compound was comparable to that of reference antidepressants, the overall profile displayed in anxiety tests was different from that of classical anxiolytics, such as the benzodiazepines. While the latter were active in a wide range of anxiety models, the vasopressin receptor antagonist showed clear-cut effects only in particularly stressful situations. In line with these findings, SSR149415 blocked several endocrine

(i.e. ACTH release), neurochemical (i.e. noradrenaline release) and autonomic (i.e. heart rate) responses following acute stress exposure. Moreover, the drug exhibited anti-aggression effects in mice and hamsters. Experiments using local infusion of SSR149415 in brain areas known to be involved in the modulation of emotionality showed that the lateral septum and the central nucleus of the amygdala play a predominant role in the antidepressant-like effects of the drug. It is noteworthy that SSR149415 was devoid of central effects not related to emotionality. These findings suggest that blockade of central V_{1b} receptors may represent a new therapeutic strategy for the treatment of depression and certain forms of anxiety disorders.

1. Introduction

The treatment of stress-related disorders remains an active area of research. Recent efforts have begun to focus on the development of pharmacological agents that can attenuate the stress response itself, rather than the symptoms associated with stress. It is well established that hyperactivity of the corticotropin-releasing factor (CRF) neuronal systems is thought to play a causal role in the etiology and symptomatology of affective disorders [1]. However, there is an accumulating body of evidence suggesting that the vasopressin system may play an equal role in the regulation of the stress response, and that vasopressin receptor antagonists may be of potential therapeutic benefit. The availability of SSR149415 [2], the first selective antagonist for the vasopressin V_{1b} receptor has allowed us to evaluate this hypothesis.

2. Evidence that vasopressin is a stress hormone and neurotransmitter

Vasopressin is a nine amino acid mammalian peptide that functions as a hormone in the blood and as a neurotransmitter/neuromodulator in the brain [3]. It is well known for its

role on fluid metabolism, but there is also clear evidence that the peptide plays an important role as a regulator of pituitary adrenocorticotropin (ACTH) secretion [4-6]. Vasopressin is critical for adaptation of the hypothalamo-pituitary-adrenal (HPA) axis during stress through its ability to potentiate the stimulatory effect of CRF. Stressors such as social defeat and forced swimming are potent stimuli to trigger release of vasopressin from the median eminence into the pituitary portal circulation and increase expression of the peptide in parvocellular neurons (for a recent review, see [7]). Increased levels of vasopressin were found in the hypothalamus of rats with high innate anxiety [8]. Studies in genetically vasopressin-deficient Brattleboro rats have provided substantial evidence for the role of the peptide in stress, as the ACTH response to several stimuli is impaired in these rats (for a recent review, see [9]). Abnormalities in vasopressin levels have been detected in depression [10-12], and there is evidence suggesting that HPA axis dysregulation in depression may be associated with a shift towards increased vasopressinergic control of the axis [13,14]. A recent clinical finding in healthy volunteers showed that vasopressin levels were significantly elevated after anxiogenic drug challenge [15]. Interestingly, volunteers with the highest levels of vasopressin also showed higher levels of respiratory distress and cognitive anxiety.

Extrahypothalamic vasopressin-containing neurons have been characterized in the rat, notably in the medial amygdala and the bed nucleus of the stria terminalis, which innervate limbic structures such as the lateral septum and the ventral hippocampus [16-18] (Figure 1). In these latter structures, the biological effects of vasopressin are mediated by activation of V_{1a} and V_{1b} receptors that activate phospholipases via $G_{q/11}$ proteins [19-21]. The V_{1a} receptors are ubiquitously located in the brain. V_{1b} receptor immunoreactivity has been described in the pituitary gland, hypothalamus, amygdala,

cerebellum and in areas close to the circumventricular organs devoid of a blood-brain barrier [19,22,23].

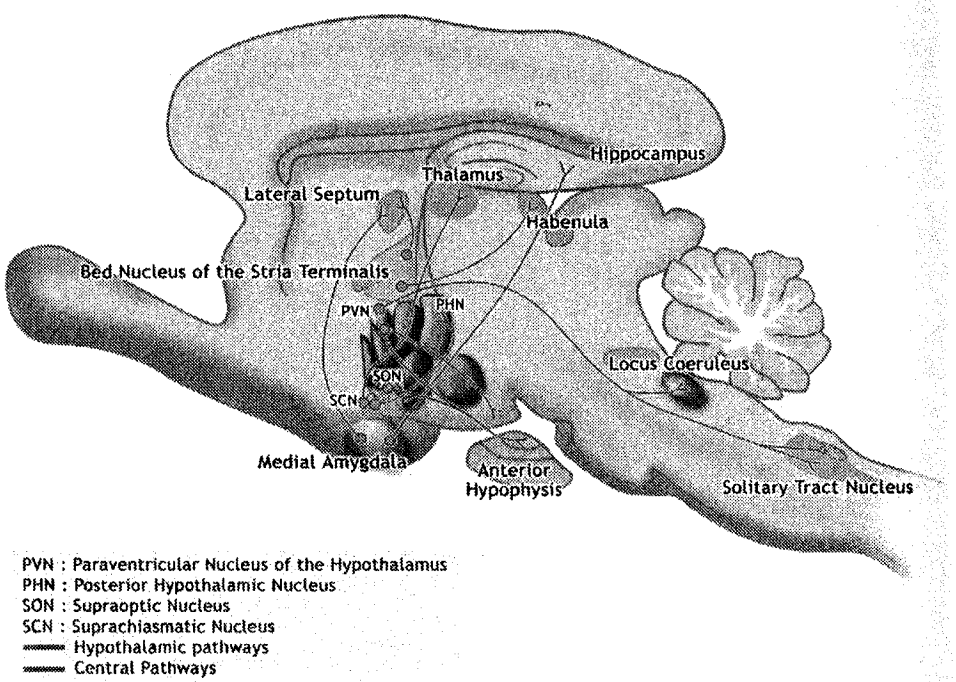


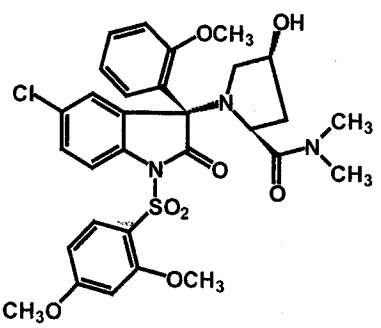
Figure 1: The main vasopressinergic pathways in the rat brain.

The presence of this vasopressin network suggests a modulator role of the peptide in limbic functioning. Initial studies on avoidance behavior in the early 1960s provided the first clue that vasopressin was involved in learning and memory processes. It is now recognized that centrally released vasopressin is involved in a variety of roles, including social, reproductive and feeding behavior [3,24-28]. The neuroanatomical distribution of vasopressin and its receptors has also prompted speculation about their functional role in emotional processes leading to studies that investigated the behavioral action of

central infusion of peptide vasopressin and V_1 receptor antagonists in animal models of anxiety. These studies showed that intracerebroventricular infusion of vasopressin produced anxiogenic-like activity in rats [29]. In contrast, the intra-septal application of the mixed $V_{1a/b}$ receptor antagonist $d(CH_2)_5$Tyr(Et)VAVP was found to produce anxiolytic-like effects [30]. Together, these findings suggest that vasopressin receptor antagonists may represent potential agents for the treatment of stress-related disorders.

3. SSR149415: the first non-peptide antagonist at vasopressin V_{1b} receptors

The first non-peptide antagonist at the V_{1b} receptor, SSR149415 (-)-(2*S*,4*R*)-1-[(3*R*)-5-chloro-1-[(2,4-dimethoxylphenyl) sulfonyl] –3-(2-methoxyphenyl)-2-oxo-2,3-dihydro-1*H* – indol – 3 – yl]- 4 – hydroxy – *N*,*N* – dimethyl – 2- pyrrolidine carboxamide, has been described recently [2] (Figure 2). It belongs to the [1H] – 2,3 dihydroindol-2 one family of vasopressin antagonists.



SSR149415

Figure 2: Chemical structure of SSR149415 ((-)-(2*S*,4*R*)-1-[(3*R*)-5-chloro-1-[(2,4-dimethoxylphenyl) sulfonyl] –3-(2-methoxyphenyl)-2-oxo-2,3-dihydro-1*H* – indol – 3 – yl]- 4 – hydroxy – *N*,*N* – dimethyl – 2- pyrrolidine carboxamide).

The compound was obtained by convergent synthesis in 8 steps from 1,3-dimethoxybenzene, Z-Hyp-OH and 5-chloroisatin (Figure 3). II was obtained from Z-Hyp-OH using a standard amidification reaction, followed by the removal of the carbobenzyloxy-protecting group. 1,3 – Dimethoxybenzene III was reacted in a two steps sequence to yield the crystalline sulfonylchloride analog IV in 30% overall yield. Treatment of 5-chloroisatin V with excess 2-methoxyphenylmagnesiumbromide gave the alcohol VI, which upon treatment with SOCl2 in pyridine yielded VII as a stable solid after chromatographic purification and crystallization. Mixing VII as a suspension in a 1/1 mixture of methylene chloride/tetrahydrofuran, with II (1.1 eq) and N-Ethyldiisopropylamine (1.1eq) for 4 days at R.T led to a clean conversion into a 1/1 mixture of the two stereoisomers VIII and IX. They were purified by fractional crystallization using suitable solvents and/or chromatography. VIII was obtained in a 33% yield from VII. Due to the presence of 2 asymetric carbon atoms of well know configuration in the pyrrolidine ring, the chirality at C-3 of the indolinone ring could be deduced unambiguously. Thus the dextro and highly insoluble stereoisomer IX was submitted to a single crystal X-Ray structural determination that showed that the absolute configuration at C-3 was 3-S. Consequently, the chirality at C-3 in the levo-stereoisomer VIII was deduced as being 3-R. Conversion of VIII to X (SSR149415; active stereoisomer) or IX to XI (SR 149424, inactive stereoisomer) was run under argon at 0°C in dimethylformamide using NaH as a base and a slight excess of IV, the proline 4-OH group being left unprotected, in a minimum yield of 70%. SSR149415 is a white crystalline solid, it has a molecular weight of 630,1 g. mole-1 and a number of O/N H-bond-acceptors exceeding the value of 10, so SSR149415 violates Lipinski's rule of 5. However its trans epithelial permeability in Caco2 /TC-7 cells (Papp = 106 x

10-7 cm.sec –1) and its transendothelial permeability (BBB) (Pe = 4x10-3 cm.min-1) should ensure SSR149415 good oral absorption and excellent penetration into the brain.

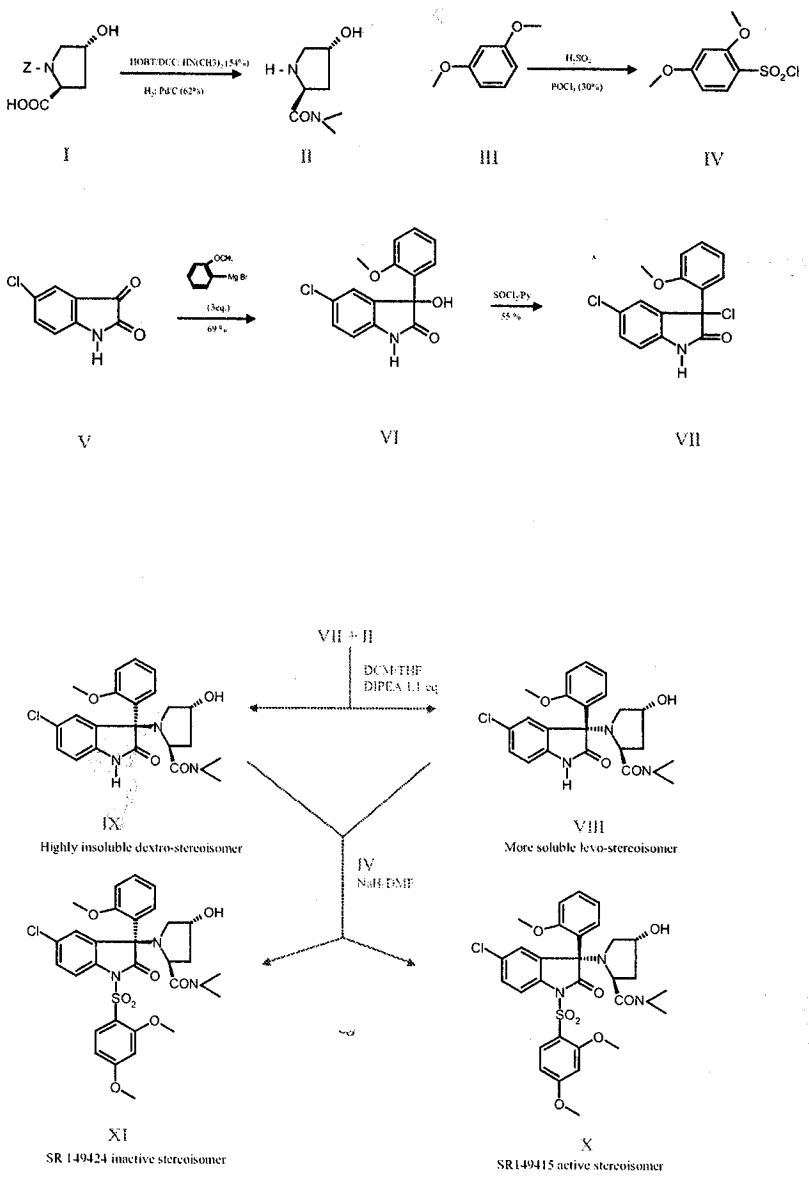


Figure 3: Synthesis of SSR149415

The compound displays high affinities for both native and recombinant human and rat V_{1b} receptors (human: K_i = 4.2 and 1.5 nM, respectively; rat: K_i = 3.7 and 1.3 nM, respectively), 60- and 800-fold selectivity for human and rat V_{1b} as compared to V_{1a} receptor, displayed weak affinity at V_2 and oxytocin (a closely vasopressin-related peptide) receptors, and was inactive in more than 90 binding assays for neurotransmitters and peptides. It is a potent antagonist at the V_{1b} receptor as shown by its ability to inhibit vasopressin-induced Ca^{2+} increase in Chinese hamster ovary cells expressing the human or rat V_{1b} receptor (K_i = 1.26 and 0.73 nM, respectively), and vasopressin-induced ACTH secretion in corticotroph cells in rats.

3.1. Evidence that SSR149415 attenuates acute stress-induced anxiety responses in animals

In traditional screening tests for anxiolytics, such as conflict paradigms (e.g. punished drinking procedure in rats and four-plate test in mice), exploratory- (e.g. elevated plus-maze in rats and light/dark choice task in mice) and social interaction-based models, SSR149415 elicited anxiolytic-like activity following acute peripheral administration (Table 1) [2,31,32]. The magnitude of the anxiolytic-like action of SSR149415 in these models was generally less than that of the benzodiazepine (BZ) anxiolytic diazepam, which was used as a positive control. Whether this may indicate a less efficacious anxiolytic-like potential of V_{1b} receptor antagonists compared to BZs, or suggests that this type of compound may have a different spectrum of therapeutic activity in anxiety disorders compared to BZs remains to be determined. Results obtained with SSR149415 in atypical models, such as the mouse defense test battery (MDTB) and procedures

based on behavioral changes produced by traumatic events (social defeat, separation or unavoidable electric shocks) may, however, be relevant to this issue (Table 1).

In the MDTB, SSR149415 failed to modify significantly risk assessment, a behavior which has been shown to be particularly sensitive to BZs, but it produced clear-cut effects on defensive aggression, a behavior which is claimed to be associated with certain aspects of stress disorders following traumatic events [33], thereby suggesting that SSR149415 may be useful in these conditions [32]. In the social defeat and conditioned fear stress-induced anxiety paradigms in mice, SSR149415 attenuated the heightened emotionality produced by prior (stressful) exposure to an aggressive isolated resident or an unavoidable foot shock, respectively. Finally, SSR149415 decreased sonic and ultrasonic vocalizations of guinea pig and rat pups, respectively, removed from their litter and separated from their mother [31].

To characterize further the anti-stress profile of SSR149415, the drug was evaluated on its ability to attenuate stress-induced activation of the HPA axis, and the central and sympathetic nervous systems (Table 1). Results showed that the drug prevented restraint stress-induced elevation of ACTH levels and heart rate, and reduced the evoked noradrenaline release following tail pinch stress [34]. Moreover, SSR149415 attenuated rebound paradoxical sleep (PS) following prior stressful deprivation of PS in rats (unpublished data). Altogether, these latter findings show clear-cut effects of the V_{1b} receptor antagonist in all models tested, with comparable efficacy to reference compounds, thereby strengthening the idea that a V_{1b} receptor antagonist may be useful in conditions associated with exposure to acute traumatic events.

Table 1: Summary of the effects of the V_{1b} receptor antagonist, SSR149415, in animal models of anxiety and stress. Comparison with the benzodiazepine, diazepam.

	MED, mg/kg, po, (ip) or §sc	
Tests	SSR149415	Diazepam
Drinking conflict test in rats	(3) [++]	(1) [+++]
Elevated plus-maze in rats	10 [+]	3 [+++]
Light/dark test in mice	(1) [+]	(1) [+++]
Four-plate test in mice	3 [+]	1 [+++]
Social interaction in gerbils	10 [+++]	(0.1) [+++]
Risk assessment in the MDTB	Inactive	(0.5) [+++]
Flight in the MDTB	30 [+]	(Inactive)
Defensive aggression in the MDTB	1 [+++]	(1) [+++]
Social defeat stress in mice	0.3 [+++]	4 [+++]
Conditioned fear stress in mice	10 [+]	(2) [++]
Distress vocalizations in rat pups§	10 [++]	1 [+++]
Distress vocalizations in guinea pig pups	(20) [+++]	NT
Restraint-induced physiological changes in rats	30 [++]	(2) [+++]
Restraint stress-induced ACTH release in rats	(10) [+++]	NT
Tail pinch stress-induced NE release in rats	(10) [+++]	NT
Swim stress-induced PS rebound in rats	30 [++]	NT

MDTB = mouse defense test battery; MED = minimal effective dose; [+++] = produced clear anxiolytic-like effects; [++] = produced significant anxiolytic-like effects; [+] = demonstrated weak anxiolytic-like effects; NT = not tested. Data are from [2,31,32], or unpublished.

3.2. Evidence that SSR149415 has antidepressant potential

The potential antidepressant-like properties of SSR149415 were examined in several procedures, including the forced-swimming test in rats [35], the stress-induced tonic immobility paradigm in gerbils [36], the chronic mild stress model in mice [37,38] and the chronic subordination stress paradigm in rats [39] (Table 2). Results from the forced-swimming and tonic immobility tests showed that SSR149415 produced dose-dependent antidepressant-like activity [32,36]. These effects were comparable to those observed with the reference antidepressant, fluoxetine.

The antidepressant potential of SSR149415 was confirmed in the chronic mild stress model in mice. This procedure consists of the sequential application of a variety of mild stressors for several weeks. It leads to a degradation of the physical state, increased emotionality and a reduced ability to cope with aversive situations. These behavioral effects are accompanied by a severe reduction in the rate of newborn cell proliferation in the hippocampus leading to a suppression of neurogenesis. Repeated administration of SSR149415 for 4 weeks reversed the degradation of the physical state, anxiety, despair and the loss of coping behavior produced by stress [32]. It is noteworthy that at the end of the stress period, animals treated with SSR149415 displayed a comparable physical state as non-stressed controls. Moreover, SSR149415 significantly reversed the suppression of cell proliferation produced by chronic stress, and prevented the dramatic reduction of granule cell neurogenesis 30 days after the end of the stress period [40].

In the chronic subordination stress model, SSR149415 was administered repeatedly to subordinate male rats, which are strongly stressed by intense fighting with a dominant rat. This procedure leads to behavioral changes in subordinates which are

broadly isomorphic to many of the symptoms of depression [39]. SSR149415 treatment reduced defensiveness in the presence of the dominant rat and behavioral inhibition such as mounting of females in the presence of the dominant [41]. These observations suggest that some aspect of the behavior of SSR149415-treated subordinates was unusually provocative of attack by the dominant. Plasma ACTH levels were reduced in vehicle subordinates compared to dominants. SSR149415-treated rats showed much higher plasma ACTH levels relative to vehicle subordinates, suggesting normalization of this HPA axis parameter change [34]. Overall, the effects of SSR149415 and fluoxetine, which was tested in parallel, were comparable, confirming the antidepressant-like potential of the V_{1b} receptor antagonist.

Table 2: Summary of the effects of the V_{1b} receptor antagonist, SSR149415, in animal models of depression. Comparison with the selective 5-HT reuptake inhibitor, fluoxetine.

	MED, mg/kg, po or (ip)	
Tests	SSR149415	Fluoxetine
Forced-swimming in rats	10 [+++]	10 [+++]
Pinch stress-induced tonic immobility in gerbils	10 [+++]	(7.5) [+++]
Chronic mild stress-induced behavioral deficits in mice	(10) [+++]	(10) [+++]
Chronic mild stress-induced alteration in neurogenesis in mice	(30) [+++]	(10) [+++]
Chronic subordination stress in rats	10 [++]	10 [++]

MED = minimal effective dose; [+++] = produced clear effects; [++] = produced significant effects. Data are from [32,36,40,41].

3.3. SSR149415 reduces offensive aggression in animals

Vasopressin has been consistently implicated in the aggressive behaviors of a variety of species, including humans [42]. In rodents, vasopressin infusions into the hypothalamus or amygdala enhance aggression in hamsters and rats, while vasopressin receptor antagonist infusions reduce aggression [43-46]. A recent study [47] of V_{1b} receptor knockout mice has reported reductions in aggressive behavior in these animals, suggesting that V_{1b} receptor antagonists may be capable of modulating aggressive behavior. We used the resident-intruder aggression paradigms in mice and hamsters to assess the potential antiaggressive properties of SSR149415. Results showed that the V_{1b} receptor antagonist significantly reduced the duration of fighting between isolated and intruder mice [31]. In hamsters, SSR149415 exhibited a consistent anti-aggression effect, reducing chase and lateral attack behaviors, and flank marking, which is an important component of dominance behavior (manuscript in preparation).

3.4. Evidence that SSR149415 is devoid of behavioral effects not related to emotionality

SSR149415 was tested in a variety of standard procedures to evaluate possible unwanted effects (Table 3). Results showed that the drug was devoid of central depressant effects (ataxia, myorelaxation and sedation) as evidenced by a lack of activity in the rotarod, the traction test and in activity cages up to 100 mg/kg. Moreover, SSR149415 did not modify sleep patterns following electroencephalographic analysis. Vasopressin has frequently been implicated in learning and memory (for reviews, see

[3,48]). We used the Morris water maze task in mice and rats to investigate potential effects of SSR149415 on spatial memory, but found no effect on either the acquisition of the test or on recalling the platform position after removal. Overall, the findings of a lack of activity of SSR149415 in these models have a direct bearing on the issue of the behavioral selectivity of any changes observed in the stress models and indicate that the drug is devoid of central effects not related to emotionality.

Table 3: Summary of the side-effect profile of the V_{1b} receptor antagonist, SSR149415, in animal models. Comparison with the benzodiazepine, diazepam.

	MED or *ED_{50}, mg/kg, po or (ip)	
Tests	SSR149415	Diazepam
EEG in rats	>30	(1)
Locomotor activity	>100	10
Traction test in mice	>100	6*
Rotarod in mice	>100	9*
Morris water maze in mice	>30	1
Morris water maze in rats	>30	10

MED = minimal effective dose; EEG = electroencephalography. Data are from [31,32], or unpublished.

4. Conclusion

Growing evidence that several neuropeptides mediate the response to psychological stress identifies these systems as promising targets for the development of novel therapeutics for stress-related disorders. Among these, vasopressin has been the focus of

recent interest. The demonstration that repeated stress produces sustained elevations in vasopressin V_{1b} receptor mRNA in the pituitary [49] along with the observation that this receptor subtype may be upregulated in depression [14], provides the framework for a novel perspective on potential anti-stress drugs, namely that V_{1b} receptor antagonists may be of potential therapeutic benefit. This idea has now been strengthened by the findings described above showing that the selective V_{1b} receptor antagonist, SSR149415, is able to attenuate a variety of stress-related responses in animals confronted with aversive situations. It is noteworthy that unlike currently used anti-stress drug treatments, SSR149415 did not produce adverse effects, even when administered at doses ten times higher than the minimal effective dose in stress models. This clearly points to a more specific anti-stress action of V_{1b} receptor blockade compared to classical anxiolytics, such as BZs for example. Although these results might be therapeutically useful, they do not tell us where in the brain SSR149415 acts to reduce anxiety- or depressive-like behaviors. However, the finding that the BZ receptor antagonist flumazenil did not block the anxiolytic-like activity of SSR149415 [32] provides undisputed evidence that these effects are at least not mediated by an action at $GABA_A$/BZ receptors. Moreover, the fact that SSR149415 is still effective in hypophysectomized rats [32] indicates that the effects do not depend on blocking only the hypothalamic V_{1b} receptors. Brain structures that may be involved in these effects include the hippocampus, the lateral septum and the amygdala as they show high levels of expression of V_{1b} receptors [19,50]. To support this idea are recent findings, which demonstrated that infusion of SSR149415 into the lateral septum, or the central nucleus of the amygdala in rats yielded antidepressant-like activity, suggesting that these structures participate in the anti-stress action of SSR149415 [51]. In conclusion, these

findings suggest that blockade of central V_{1b} receptors may represent a new therapeutic strategy for the treatment of psychiatric disorders caused by dysfunction in the response to psychological trauma and stress.

References

1 Hökfelt T et al (2000) *Neuropharm* 39, 1337-1356

2 Serradeil-Le Gal C et al (2002) *J Pharmacol Exp Ther* 300, 1122-1130

3 Engelmann M et al (1996) *Neurosci Biobehav Rev* 20, 341-358

4 McCann SM and Brobeck JR (1954) *Proc Natl Acad Sci USA* 87, 318-324

5 Antoni FA (1993) *Front Neuroendocrinol* 14, 76-122

6 Aguilera G (1994) *Front Neuroendocrinol* 15, 321-350

7 Aguilera G and Rabadan-Diehl C (2000) *Regul Pept* 96, 23-29

8 Keck ME et al (2002) *Neuropsychopharmacology* 26, 94-105

9 Carrasco GA and Van de Kar LDV (2003) *Eur J Pharmacol* 463, 235-272

10 Purba JS et al (1996) *Arch Gen Psychiatry* 53, 137-143

11 Zhou JN et al (2001) *Arch Gen Psychiatry* 58, 655-662

12 van Londen L et al (1997) *Neuropsychopharmacology* 17, 284-292

13 Holsboer F and Barden N (1996) *Endocrine Rev* 17, 187-205

14 Dinan TG et al (1999) *J Clin Endocrinol Metab* 84, 2238-2240

15 Abelson JL et al (2001) *Neuropsychopharmacology* 24, 161-169

16 Caffé AR et al (1987) *J Comp Neurol* 261, 237-252

17 De Vries GJ and Buijs RM (1983) *Brain Res* 273, 307-317

18 van Leeuwen FW and Caffé AR (1983) *Cell Tissue Res* 28, 525-534

19 Lolait SJ et al (1995) *Proc Natl Acad Sci USA* 92, 6783-6787

20 Young LJ et al (1999) *J Neuroendocrinol* 11, 291-297

21 Vaccari C et al (1998) *Endocrinol* 139, 5015-5033

22 Morel A et al (1992) *Nature* 356, 523-526

23 Tribollet E et al (1999) *Neuroendocr* 69, 113-120

24 De Wied D (1965) *Int J Neuropharmacol* 4, 157-167

25 De Wied D (1970) *J Endocrinol* 48, xlv-xlvi

26 Koob GF and Bloom FE (1982) *Annu Rev Physiol* 44, 571-582

27 Alescio-Lautier B et al (1993) *Rev Neurosci* 4, 239-266

28 Dantzer R and Bluthe RM (1992) *Crit Rev Neurobiol* 6, 243-255

29 Bhattacharya SK et al (1998) *Biog Amine* 14, 367-385

30 Liebsch G et al (1996) *Neurosci Lett* 217, 101-104

31 Griebel G et al (2002) *J Neuropsychopharmacol* 5 (Suppl. 1), S128

32 Griebel G et al (2002) *Proc Natl Acad Sci USA* 99, 6370-6375

33 Blanchard RJ et al (1997) *Neurosci Biobehav Rev* 21, 783-789

34 Griebel G, Simiand J, Serradeil-Le Gal C, Steinberg R (2003) In: *Handbook on Stress, Immunology and Behaviour* (T Steckler, N Kalin, JMHM Reul, eds), Elsevier Science B.V., The Netherlands (in press)

35 Porsolt RD et al (1977) *Nature* 266, 730-732

36 Simiand J et al (2003) *Behav Pharmacol* (in press)

37 Willner P et al (1992) *Neurosci Biobehav Rev* 16, 525-534

38 Kopp C et al (1999) *Behav Pharmacol* 10, 73-83

39 Blanchard DC et al (1993) *Behav Brain Res* 58, 113-121

40 Alonso R et al (2003) *Mol Psychiatr* (submitted)

41 Blanchard RJ et al (2002) *Program No 307 3 Abstract Viewer/Itinerary Planner Washington, DC: Society for Neuroscience, CD-ROM*

42 Coccaro EF et al (1998) *Arch Gen Psychiatry* 55, 708-714

43 Ferris CF et al (1988) *Eur J Pharmacol* 154, 153-159

44 Ferris CF et al (1986) *Physiol Behav* 38, 135-138

45 Ferris CF and Potegal M (1988) *Physiol Behav* 44, 235-239

46 Ferris CF and Delville Y (1994) *Psychoneuroendocr* 19, 593-601

47 Wersinger SR et al (2002) *Molecular Psychiatry* 7, 975-984

48 Van Wimersma Greidanus TB et al (1983) *Pharmacol Ther* 20, 437-458

49 Rabadan-Diehl C et al (1995) *J Neuroendocrinol* 7, 903-910

50 Hernando F et al (2001) *Endocrinol* 142, 1659-1668

51 Stemmelin J et al (2003) *Soc Neurosci Abstr* (in press)