Available online at www.sciencedirect.com
SCIENCE DIRECT
European Journal of Pharmacology 540 (2006) 115–120
www.elsevier.com/locate/ejphar

Lack of efficacy of melanin-concentrating hormone-1 receptor antagonists in models of depression and anxiety

Ana M. Basso [a,*], Natalie A. Bratcher [a], Kelly B. Gallagher [a], Marlon D. Cowart [a], Chen Zhao [a], Minghua Sun [a], Timothy A. Esbenshade [a], Michael E. Brune [b], Gerard B. Fox [a], Martin Schmidt [a], Christine A. Collins [b], Andrew J. Souers [b], Rajesh Iyengar [b], Anil Vasudevan [b], Philip R. Kym [b], Arthur A. Hancock [a], Lynne E. Rueter [a]

[a] *Neuroscience, Abbott Laboratories, Global Pharmaceutical Research and Development, Abbott Laboratories, Abbott Park, IL 60064, USA*
[b] *Metabolic Diseases Research, Abbott Laboratories, Global Pharmaceutical Research and Development, Abbott Laboratories, Abbott Park, IL 60064, USA*

Received 2 March 2006; received in revised form 20 April 2006; accepted 28 April 2006
Available online 6 May 2006

Abstract

The aim of this study was to validate melanin-concentrating hormone (MCH)-1 receptor antagonism as a potential treatment of mood disorders. We attempted to replicate the effects previously reported with SNAP-7941 and expanded the investigation to three other orally bioavailable MCH-1 receptor antagonists with good brain penetration. SNAP-7941 (3–30 mg/kg, i.p.) and T-226296 (5–60 mg/kg, p.o.) (± racemate), were evaluated in the rat forced swim and mouse tail suspension tests. (+)SNAP-7941 (3–10 mg/kg, p.o.) was also tested in a modified 5-min rat forced swim protocol as previously reported. A-665798 (3–30 mg/kg, p.o.) and A-777903 (3–30 mg/kg, p.o.) were tested in mouse tail suspension and rat Vogel tests. None of the compounds showed meaningful efficacy in the paradigms tested. The lack of efficacy with four structurally different MCH-1 receptor antagonists does not support a role for therapeutic treatment of depression/anxiety via this mechanism of action.

Keywords: MCH-1 receptor antagonists; Depression; Anxiety; (Rat); (Mouse)

1. Introduction

Melanin-concentrating hormone (MCH) regulates a variety of functions in mammalian brain, in particular, energy homeostasis and feeding behavior. However, additional physiological roles for this peptide remain unknown.

The MCH-1 receptor has been isolated from rodent and human and bind MCH with high affinity (Saito et al., 1999). The localization of MCH and MCH-1 receptors in the brain, suggests a functional role in the regulation of arousal, stress and emotion (Hervieu, 2003). However, the effect of central administration of MCH in anxiety-related behaviors and neuroendocrine responses associated with stress is controversial and not clearly understood (Gonzalez et al., 1996; Kela et al., 2003; Kennedy et al., 2003; Ludwig et al., 1998; Monzon and De Barioglio, 1999).

The aim of this study was to test the hypothesis as to whether MCH-1 receptor antagonists might be useful for the treatment of mood disorders. We evaluated the efficacy of potent non-peptide MCH-1 receptor antagonists with unrelated chemical structures in well-established paradigms for the screening of antidepressant and anxiolytic compounds. We tested SNAP-7941, a selective and potent human (h) MCH-1 receptor antagonist (K_i=15±0.11 nM), reported to have anxiolytic, antidepressant and anorectic properties (Borowsky et al., 2002), and extended the analysis to three other orally bioavailable hMCH-1 receptor antagonists: T-226296 (IC_{50}=87±14 nM) (Takekawa et al., 2002), A-665798 (IC_{50}=2.0±0.9 nM) (Kym et al., 2005) and A-777903 (IC_{50}=16±7 nM) (Vasudevan et al., 2005). SNAP-7941 and

* Corresponding author. Tel.: +1 847 935 1061; fax: +1 847 938 0072.
E-mail address: Ana.basso@abbott.com (A.M. Basso).

0014-2999/$ - see front matter © 2006 Elsevier B.V. All rights reserved.
doi:10.1016/j.ejphar.2006.04.043

T-226296 (±, racemate) were tested in the rat forced swim and mouse tail suspension tests and A-665798 and A-777903 in the mouse tail suspension and rat Vogel conflict tests. Based on the potential relevance of MCH in stress-related behaviors and the previous report by Borowsky et al. (2002), we also attempted to replicate the data following the same experimental procedure and using the active enantiomer (+)SNAP-7941.

2. Materials and methods

2.1. Animals

Male Sprague Dawley rats (forced swim), male Wistar rats (Vogel, both 250–350 g, unless otherwise specified) and male Balb/c mice (tail suspension and locomotor activity, 20–25 g), were purchased from Charles River Laboratories. Rats and mice were group housed (5 and 10/cage, respectively) and maintained in a controlled environment on a 12:12 h light-dark schedule with free access to food and water, unless otherwise described. Protocols were approved by Abbott Laboratories Institutional Animal Care and Use Committee, according to the guidelines of the Association for the Assessment and Accreditation of Laboratory Animals Care (AAALAC).

2.2. Rat forced swim test

Each experiment consisted of a pre- and a test-swim. Naïve rats were placed inside cylinders containing 25 cm of water at 23–25 °C for 15 min. After 24 h, the rats were replaced in the cylinder for 5 min (test-swim), and the total duration of immobility and escape behaviors was measured. The active enantiomer (+)SNAP-7941, was tested using a non-standard protocol where the rats (150–180 g) were placed only once in a cylinder with 30 cm of water at 23–25 °C for a 5 min test-swim session (no pre-swim) (Borowsky et al., 2002). Test-swims were videotaped and assessed for the following behaviors: immobility, climbing and swimming (Detke et al., 1995).

2.3. Mouse tail suspension test

Following the pretreatment time, a piece of tape was wrapped around the tail 20 mm from the tip. The mouse was hung by the tape from a hook attached to a transducer, which communicated information about duration of movements to a computer. Immobility time was recorded for 6 min.

2.4. Vogel conflict test

Rats were water-deprived for 48 h before testing. After 24 h of deprivation, rats were habituated to the testing chambers and allowed to drink for 15 min, with an additional 15 min drinking in their home cages. Water deprivation then continued for another 24 h. On the test day, rats were placed in the chambers with access to the water spigot for 5 min. Every 20 licks, they received one shock of 0.5 mA, 1 s duration delivered through the drinking tube. The number of punished responses was recorded.

2.5. Locomotor activity

Mice were habituated to the testing room under dim lighting for 1 h. Locomotor activity was recorded for 90 min by individually placing the mice into activity chambers (42 cm × 42 cm × 30 cm) after the drug administration. Activity was detected by infrared photo beam sensors and recorded as total distance moved (cm) during 90 min.

2.6. Drugs

SNAP-7941 [methyl (4 S)-3-[(3-4-[3-(acetylamino) phenyl]-1-piperidinyl propyl) amino] carbonyl-4-(3,4-difluorophenyl)-6-(methoxymethyl)-2-oxo-1,2,3,4-tetrahydro-5-pyrimidinecarboxylate hydrochloride] (racemate and active enantiomer), T-226296 [(±)*N*-[6-(dimethylamino)-methyl]-5,6,7,8-tetrahydro-2-naphthalenyl]-4′–fluoro[1,1′–biphenyl]-4-carboxamide] (racemate), A-665798 [4-(1-Benzo[1,3]dioxol-5-ylmethyl-piperidin-4-ylamino)-6-chloro-chromen-2-one] (designated as compound 7 in Kym et al., 2005) and A-777903 [(1-Benzo[1,3] dioxol-5-ylmethyl-piperidin-4-yl)-(5-chloro-1H-indazol-3-yl)-amine] (designated as compound 19 in Vasudevan et al., 2005), were synthesized at Abbott Laboratories. (±)SNAP-7941 was solubilized in 1% dimethylsulfoxide in water with methanesulfonic acid 1 N, pH=4.5–5.5 and (+)SNAP-7941 in lactic acid solution, pH=5–6. T-226269 was suspended in 0.5% methylcellulose in water. A-665798 and A-777903 were suspended in 1% Tween 80 in water. Imipramine and chlordiazepoxide hydrochloride (Sigma, St. Louis, MO, USA) were dissolved in water.

2.7. Experimental procedure

(±)SNAP-7941 was administered i.p. 24, 5 and 0.5 h before the test-swim session (1 ml/kg), with the first administration 15 min after the pre-swim session. For the tail suspension test, (±)SNAP-7941 was administered 0.5 h before the test (10 ml/kg). Doses of (±)SNAP-7941 in both models were 3, 10 and 30 mg/kg. (+)SNAP-7941 was tested in the rat forced swim test as described by Borowsky et al., 2002, being administered acutely at 3 and 10 mg/kg, p.o. (1 ml/kg), 1 h before the 5 min test-swim session.

T-226296 was administered p.o., 24, 5 and 1 h before the forced swim (5 ml/kg) and 1 h before the tail suspension (10 ml/kg). Doses of T-226296 were 20, 40 and 60 mg/kg for the rat forced swim test and 5, 10, 15, 20, 40 and 60 mg/kg for the tail suspension test. A-665798 and A-777903 were administered p.o., 1 h before Vogel and 45 min before the tail suspension at 3, 10 and 30 mg/kg (1 ml/kg for Vogel and 10 ml/kg for tail suspension). T-226296 and A-777903 were tested in locomotor activity to better understand the results in tail suspension. Route of administration and pre-treatment times were selected in accordance with pharmacokinetic data obtained in house, showing acceptable plasma and brain

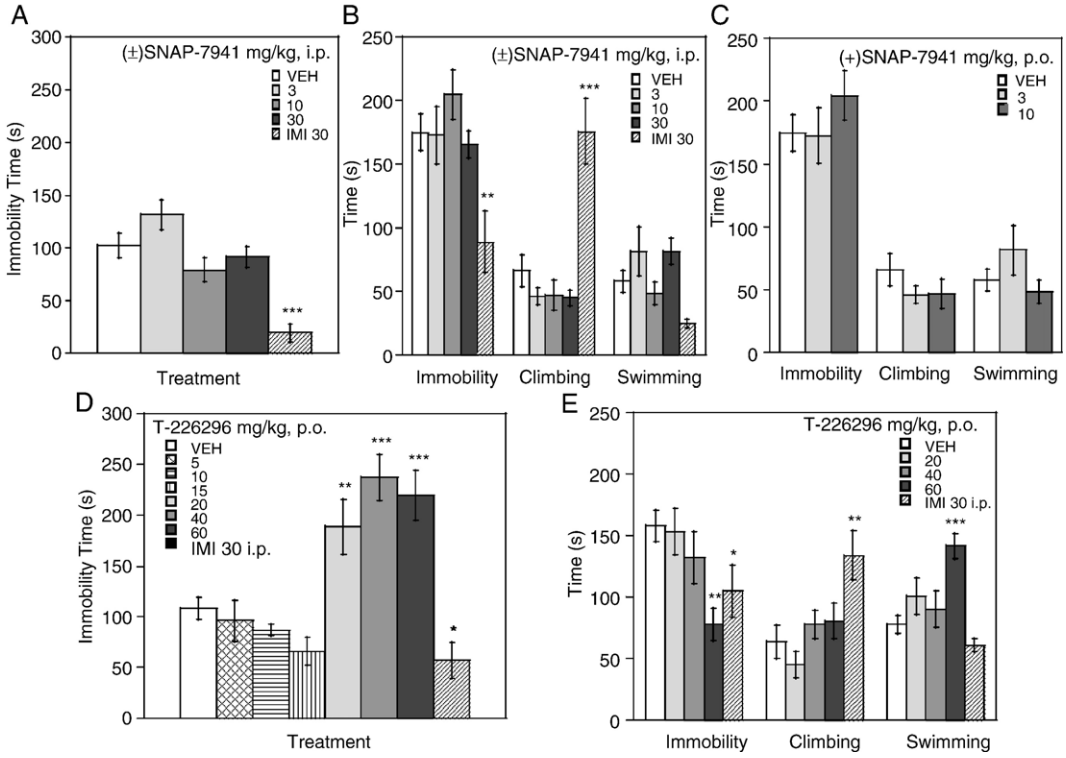

Fig. 1. Effects of (±)SNAP-7941 on A) mouse tail suspension and B) rat forced swim test; effects of (+)SNAP-7941 on C) rat forced swim test; and effects of T-226296 on D) mouse tail suspension and E) rat forced swim test. Graphs represent mean ± S.E.M. ★$P<0.05$, ★★$P<0.01$ and ★★★$P<0.001$ compared to vehicle (Fisher test). IMI = imipramine.

concentrations at the time of testing. Doses of the compounds were selected according to demonstrated efficacy in previous in vivo experiments (Borowsky et al., 2002; Kym et al., 2005; Vasudevan et al., 2005).

Each experimental group was $n=9–10$, except for (+)SNAP-7941 in rat forced swim ($n=12–13$), T-226296 in tail suspension ($n=10–22$) and A-777903 in Vogel test ($n=8$). Positive control compounds were tested in parallel. Animals were used only once in a single experiment.

2.8. Statistical analyses

Data were analyzed using a one-way analysis of variance (ANOVA) for treatment groups including the data for the reference compound. When the ANOVA revealed a significant effect, post hoc analysis for individual group comparisons (Fisher PLSD test) followed the overall ANOVA. Data are expressed as the mean ± S.E.M. Significance was defined at least as $P<0.05$.

3. Results

3.1. Effect of SNAP-7941 and T-226296 on the mouse tail suspension test and the rat forced swim test

(±)SNAP-7941 did not show antidepressant-like properties at the doses tested in either of the two animal models used: tail suspension test and rat forced swim test. The reference compound imipramine, but not (±)SNAP-7941, induced a significant decrease in immobility in the tail suspension [(ANOVA $F_{4,\ 45}=13.177$, $P<0.001$) ($P<0.001$ Fisher test)] (Fig. 1A).

Data for the forced swim test indicated that imipramine elicited a significant effect decreasing immobility [(ANOVA $F_{4,\ 42}=3.483$, $P<0.05$) ($P<0.01$ Fisher test)], and increasing climbing [(ANOVA $F_{4,\ 42}=15.287$, $P<0.001$) ($P<0.001$ Fisher test)]. (±)SNAP-7941 failed to induce a significant effect in the rat forced swim test (Fig. 1B). The active enantiomer (+)SNAP-7941, did not show antidepressant-like efficacy in the rat forced swim test when administered acutely 1 h before the 5-min swim session. ANOVA revealed a non-significant treatment effect for immobility ($F_{2,34}=0.233$, n.s.), climbing ($F_{2,34}=0.5468$, n.s.) and swimming ($F_{2,34}=1.131$, n.s.) (Fig. 1C).

T-226296 did not show antidepressant-like activity in the tail suspension test. ANOVA revealed a significant treatment effect for immobility ($F_{7,\ 103}=13.747$, $P<0.001$) that was related with the efficacy of imipramine ($P<0.05$). Moreover, doses of 20, 40 and 60 mg/kg of T-226296 induced a significant increase in immobility time, the opposite effect than that expected for an antidepressant-like profile (Fig. 1D).

ANOVA for the forced swim test with T-226296 indicated a significant treatment effect for immobility ($F_{4,\ 45}=3.567$, $P<0.05$), climbing ($F_{4,\ 45}=5.330$, $P<0.01$), and swimming ($F_{4,\ 45}=7.453$, $P<0.001$). T-226296 at the highest dose of 60 mg/kg induced a significant decrease in immobility ($P<0.01$)

and an increase in swimming time ($P<0.001$). As expected, imipramine decreased immobility ($P<0.05$), and increased climbing ($P<0.01$) (Fig. 1E).

3.2. Effect of A-665798 and A-777903 on the mouse tail suspension test and Vogel conflict test in rat

A-665798 and A-777903 did not show antidepressant- or anxiolytic-like effects in the tail suspension test and Vogel test.

ANOVA for A-665798 in the tail suspension revealed a significant treatment effect for immobility ($F_{4,\ 44}=6.455$, $P<0.001$), related with the antidepressant-like effect of imipramine ($P<0.01$, Fisher test). No significant effect was elicited by A-665798 (Fig. 2A).

None of the doses of A-665798 showed anxiolytic-like activity in Vogel test. ANOVA revealed a significant treatment effect for the number of punished responses ($F_{4,\ 42}=10.015$, $P<0.001$), which was associated with the efficacy of chlordiazepoxide ($P<0.001$) (Fig. 2B).

ANOVA for A-777903 in the tail suspension test revealed a significant effect for immobility ($F_{4,\ 44}=24.116$, $P<0.001$). Imipramine, but not A-777903, decreased immobility time thereby demonstrating an antidepressant-like effect ($P<0.01$). The highest dose of A-777903, 30 mg/kg, elicited a robust increase in immobility, a pattern opposite to an antidepressant profile, which may be reflective of changes in activity (Fig. 2C).

ANOVA for Vogel test, revealed a significant treatment effect for the number of punished responses ($F_{4,\ 31}=3.306$, $P<0.05$), associated with the anxiolytic-like activity of chlordiazepoxide ($P<0.001$), but not A-777903 (Fig. 2D).

3.3. Effect of T-226296 and A-777903 on locomotor activity in mice

T-226296 and A-777903 decreased spontaneous locomotor activity in mice at most of the doses tested ($F_{5,\ 54}=14.167$, $P<0.001$ for T-226296 and $F_{3,\ 36}=13.75$, $P<0.001$ for A-777903). Fisher post hoc revealed a decrease in distance moved at 5 mg/kg ($P<0.01$), 10, 20, 40 and 60 mg/kg of T-226296 ($P<0.001$), as well as at 10 and 30 mg/kg of A-777903 ($P<0.05$ and $P<0.001$, respectively). Spontaneous locomotor activity supports the data from tail suspension test showing higher immobility time in the model (data not shown).

4. Discussion

In the present study we attempted to validate the efficacy of MCH-1 receptor antagonists for the potential treatment of affective disorders as proposed by a previous report (Borowsky et al., 2002). Our study was not able to demonstrate efficacy for SNAP-7941 in two models of depression: mouse tail suspension test and rat forced swim test. These findings led us to study other MCH-1 receptor antagonists with diverse chemical structures. T-226296 was tested in the mouse tail suspension and the rat forced swim tests, and two novel MCH-1 receptor antagonists, A-665798 and A-777903 (Kym et al., 2005; Vasudevan et al., 2005), were evaluated in the mouse tail suspension and the Vogel tests. None of the compounds showed efficacy in animal models of depression and/or anxiety.

SNAP-7941 has been reported as having efficacy in models of antidepressant and anxiolytic activity. Borowsky

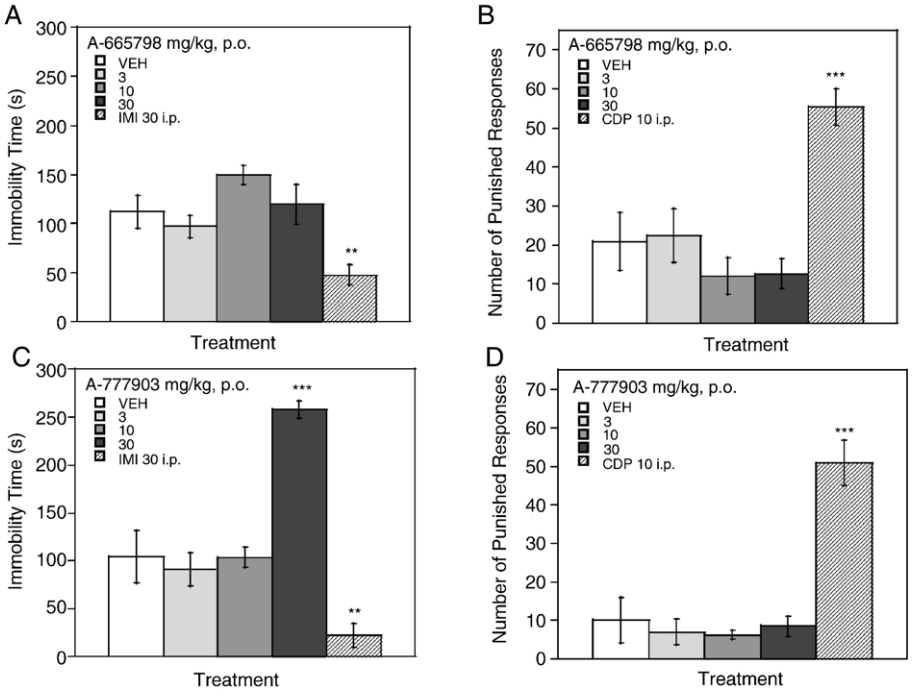

Fig. 2. Effects of A-665798 on A) mouse tail suspension and B) Vogel conflict test; and effects of A-777903 on C) mouse tail suspension and D) Vogel conflict test. Graphs represent mean±S.E.M. $^{\star\star}P<0.01$ and $^{\star\star\star}P<0.001$ compared to vehicle treated rats (Fisher test). CDP=chlordiazepoxide.

et al. (2002) tested the compound in only one model of depression, the rat forced swim test, using a different procedure than the standard protocol involving a pre-swim session followed 24 h later by a 5 min swim session, between which the compound is usually administered 3 times in a 24 h period (Detke et al., 1995; Porsolt et al., 1978). Their modified version included a single 5 min swim session after a single drug administration. Due to the differences in the methodology as well as the possible impact of using the racemate of SNAP-7941 instead of the active enantiomer, we did an additional study following exactly the experimental procedure described in Borowsky publication. The active enantiomer of SNAP-7941 did not show antidepressant-like efficacy in the rat forced swim when administered acutely before a single swim session. It has been proposed that exposure to the pre-swim session will elicit altered behavior in response to subsequent stressors (swim session), such as a more rapid adoption of the immobility posture and for a longer period than that seen in naïve animals, suggestive of a "depressive-like state". If animals have not been exposed to the pre-swim session, it becomes more likely that lower immobility times observed in those animals could be related to changes in spontaneous activity induced by the drug, rather than a specific antidepressant effect.

T-226296 showed efficacy in blocking the increase in food intake induced by MCH (Takekawa et al., 2002). T-226296 did not produce antidepressant-like activity in the tail suspension test. In addition, the compound elicited a significant increase in immobility, probably due to reduction of spontaneous activity as demonstrated in the mouse locomotor activity study. T-226296 showed no efficacy in the rat forced swim test at doses of 20 and 40 mg/kg. The highest dose of 60 mg/kg induced a reduction in immobility, an effect that could be due to the activity of the compound on other receptors. A receptor profiling study at CEREP revealed that T-226296 at 10 μM ($n=2$), produced a 98% inhibition of ligand binding to muscarinic receptors and to a lesser extent to serotoninergic and alpha-adrenergic receptors (unpublished data). Binding to muscarinic receptors could be implicated in this effect since tricyclic antidepressants are potent muscarinic antagonists and antimuscarinic agents show antidepressant-like activity in this test (Mancinelli et al., 1988).

It has been reported that intracerebral administration of an MCH-1 receptor antagonist to rats produces antidepressant-like effects in the forced swim test, similar to the behavioral profile observed in mice with a deletion in the MCH gene (Georgescu et al., 2005). Differences between the cited paper and our results could be explained by possible differences in effective concentrations of the compounds in specific areas of the brain following intracerebral or systemic administration. Another report indicated that MCH-1 receptor knockout mice are hyperactive when evaluated in their home-cages, making it difficult to conclude about a particular behavioral and/or antidepressant-like phenotype (Marsh et al., 2002). Null mutations of particular receptors have been developed to generate a genetic model for the phenotype of the receptor antagonist. Nevertheless, pharmacological antagonists not always induce the same effects observed in knockout mice phenotype, since developmental and plastic changes might be present to compensate for the mutation of a gene.

Chaki et al. 2005, have described anxiolytic- and antidepressant-like effects for two hMCH-1 receptor antagonists ATC0065 and ATC0175, however these compounds also behave as h5-HT_{2B} receptor antagonists and 5-HT_{1A} receptor partial agonists, making difficult the differentiation of the involvement of MCH-1 receptor antagonism versus the involvement of 5-HT receptor activity in the behavioral effects of these compounds.

The compounds in the current study have adequate bioavailability and show good plasma and brain levels after systematic administration. In addition, they all decreased body weight and food intake in diet-induced obese mice, in agreement with the proposed role of MCH in feeding behavior (Kym et al., 2005; Vasudevan et al., 2005). Thus, pharmacokinetic properties or brain penetrability are not likely sources for the lack of efficacy of these compounds in the present study (data not shown).

The effect of central administration of MCH in anxiety-related behaviors is unclear. MCH induces anxiogenic-like effects (Gonzalez et al., 1996), as well as anxiolytic-like effects (Kela et al., 2003; Monzon and De Barioglio, 1999). The role of MCH modulating neuroendocrine responses associated with stress is also controversial, since there is evidence for both stimulatory and inhibitory action on the hypothalamic–pituitary–adrenal axis (Kennedy et al., 2003; Ludwig et al., 1998). Thus, the idea that MCH may have opposite actions in the neuroendocrine system and in stress-mediated behaviors requires further investigation.

In summary, in the present study we have not been able to demonstrate activity of SNAP-7941 and three additional, structurally distinct, non-peptide MCH-1 receptor antagonists in animal models of depression/anxiety. This suggests that the lack of efficacy is not related to the structure-activity mechanism of these compounds. Finally, our current findings do not support a role for MCH-1 receptor antagonists as a potential therapeutic treatment for depression and/or anxiety disorders.

References

Borowsky, B., Durkin, M.M., Ogozalek, K., Marzabadi, M.R., DeLeon, J., Lagu, B., Heurich, R., Lichtblau, H., Shaposhnik, Z., Daniewska, I., Blackburn, T.P., Branchek, T.A., Gerald, C., Vaysse, P.J., Forray, C., 2002. Antidepressant, anxiolytic and anorectic effects of a melanin-concentrating hormone-1 receptor antagonist. Nat. Med. 8, 825–830.

Chaki, S., Funakoshi, T., Hirota-Okuno, S., Nishiguchi, M., Shimazaki, T., Iijima, M., Grottick, A.J., Kanuma, K., Omodera, K., Sekiguchi, Y., Okuyama, S., Tran, T.A., Semple, G., Thomsen, W., 2005. Anxiolytic- and antidepressant-like profile of ATC0065 and ATC0175: nonpeptidic and orally active melanin-concentrating hormone receptor 1 antagonists. J. Pharmacol. Exp. Ther. 313, 831–839.

Detke, M.J., Rickels, M., Lucki, I., 1995. Active behaviors in the rat forced swimming test differentially produced by serotonergic and noradrenergic antidepressants. Psychopharmacology (Berl) 121, 66–72.

Georgescu, D., Sears, R.M., Hommel, J.D., Barrot, M., Bolanos, C.A., Marsh, D.J., Bednarek, M.A., Bibb, J.A., Maratos-Flier, E., Nestler, E.J., DiLeone, R.J., 2005. The hypothalamic neuropeptide melanin-concentrating hormone acts in the nucleus accumbens to modulate feeding behavior and forced-swim performance. J. Neurosci. 25, 2933–2940.

Gonzalez, M.I., Vaziri, S., Wilson, C.A., 1996. Behavioral effects of alpha-MSH and MCH after central administration in the female rat. Peptides 17, 171–177.

Hervieu, G., 2003. Melanin-concentrating hormone functions in the nervous system: food intake and stress. Expert Opin. Ther. Targets 7, 495–511.

Kela, J., Salmi, P., Rimondini-Giorgini, R., Heilig, M., Wahlestedt, C., 2003. Behavioural analysis of melanin-concentrating hormone in rats: evidence for orexigenic and anxiolytic properties. Regul. Pept. 114, 109–114.

Kennedy, A.R., Todd, J.F., Dhillo, W.S., Seal, L.J., Ghatei, M.A., O'Toole, C.P., Jones, M., Witty, D., Winborne, K., Riley, G., Hervieu, G., Wilson, S., Bloom, S.R., 2003. Effect of direct injection of melanin-concentrating hormone into the paraventricular nucleus: further evidence for a stimulatory role in the adrenal axis via SLC-1. J. Neuroendocrinol. 15, 268–272.

Kym, P.R., Iyengar, R., Souers, A.J., Lynch, J.K., Judd, A.S., Gao, J., Freeman, J., Mulhern, M., Zhao, G., Vasudevan, A., Wodka, D., Blackburn, C., Brown, J., Che, J.L., Cullis, C., Lai, S.J., LaMarche, M.J., Marsilje, T., Roses, J., Sells, T., Geddes, B., Govek, E., Patane, M., Fry, D., Dayton, B.D., Brodjian, S., Falls, D., Brune, M., Bush, E., Shapiro, R., Knourek-Segel, V., Fey, T., McDowell, C., Reinhart, G.A., Preusser, L.C., Marsh, K., Hernandez, L., Sham, H.L., Collins, C.A., 2005. Discovery and characterization of aminopiperidinecoumarin melanin concentrating hormone receptor 1 antagonists. J. Med. Chem. 48, 5888–5891.

Ludwig, D.S., Mountjoy, K.G., Tatro, J.B., Gillette, J.A., Frederich, R.C., Flier, J.S., Maratos-Flier, E., 1998. Melanin-concentrating hormone: a functional melanocortin antagonist in the hypothalamus. Am. J. Physiol. 274, E627–E633.

Mancinelli, A., Borsini, F., d'Aranno, V., Lecci, A., Meli, A., 1988. Cholinergic drug effects on antidepressant-induced behaviour in the forced swimming test. Eur. J. Pharmacol. 158, 199–205.

Marsh, D.J., Weingarth, D.T., Novi, D.E., Chen, H.Y., Trumbauer, M.E., Chen, A.S., Guan, X.M., Jiang, M.M., Feng, Y., Camacho, R.E., Shen, Z., Frazier, E.G., Yu, H., Metzger, J.M., Kuca, S.J., Shearman, L.P., Gopal-Truter, S., MacNeil, D.J., Strack, A.M., MacIntyre, D.E., Van der Ploeg, L.H., Qian, S., 2002. Melanin-concentrating hormone 1 receptor-deficient mice are lean, hyperactive, and hyperphagic and have altered metabolism. Proc. Natl. Acad. Sci. U. S. A. 99, 3240–3245.

Monzon, M.E., De Barioglio, S.R., 1999. Response to novelty after i.c.v. injection of melanin-concentrating hormone (MCH) in rats. Physiol. Behav. 67, 813–817.

Porsolt, R.D., Anton, G., Blavet, N., Jalfre, M., 1978. Behavioural despair in rats: a new model sensitive to antidepressant treatments. Eur. J. Pharmacol. 47, 379–391.

Saito, Y., Nothacker, H.P., Wang, Z., Lin, S.H., Leslie, F., Civelli, O., 1999. Molecular characterization of the melanin-concentrating-hormone receptor. Nature 400, 265–269.

Takekawa, S., Asami, A., Ishihara, Y., Terauchi, J., Kato, K., Shimomura, Y., Mori, M., Murakoshi, H., Suzuki, N., Nishimura, O., Fujino, M., 2002. T-226296: a novel, orally active and selective melanin-concentrating hormone receptor antagonist. Eur. J. Pharmacol. 438, 129–135.

Vasudevan, A., Souers, A.J., Freeman, J.C., Verzal, M.K., Gao, J., Mulhern, M.M., Wodka, D., Lynch, J.K., Engstrom, K.M., Wagaw, S.H., Brodjian, S., Dayton, B., Falls, D.H., Bush, E., Brune, M., Shapiro, R.D., Marsh, K.C., Hernandez, L.E., Collins, C.A., Kym, P.R., 2005. Aminopiperidine indazoles as orally efficacious melanin concentrating hormone receptor-1 antagonists. Bioorg. Med. Chem. Lett. 15, 5293–5297.