Drugs of the Future 2004, 29(2): 137-147

CCC: 0377-8282/2004

Review Article

Brain neuropeptide Y (NPY) system: a candidate target for treatment of anxiety, depression and alcohol dependence

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CONTENTS

Abstract

Neuropeptide Y (NPY), the prototypical member of the NPY-like peptide family, antagonizes behavioral consequences of stress through actions within the brain. Behavioral antistress actions of NPY are noteworthy in that their magnitude surpasses that of other endogenous compounds, and they are produced across a wide range of animal models, normally thought to reflect different aspects of emotionality. These findings suggest that NPY acts with a high potency on a common core mechanism of emotionality and behavioral stress responses. Behavioral studies in genetically modified animals support this hypothesis. Increased emotionality and increased alcohol intake is seen upon inactivation of NPY transmission, while the opposite is found when NPY signaling is made overactive. Together, available data point to the potential of the NPY system as a target for novel pharmacological treatments of stress-related disorders, including anxiety and depression. Recent data additionally point to a role of NPY in the regulation of alcohol intake, and alcohol dependence emerges as a novel potential indication for compounds targeting the NPY system.

Introduction: basic biology of the central NPY system

Neuropeptide Y and the NPY family of peptides

Neuropeptide Y (NPY) is a 36 amino acid (aa) peptide with a C-terminal amide group. It belongs to a family of peptides among which pancreatic polypeptide (PP) was first discovered as a biproduct of insulin isolation (1). PP-like immunoreactivity in the brain was then found to represent the related peptide NPY, named so because of its exclusively neuronal expression and its terminal tyrosine (Y in the 1-letter aa code) (2, 3). Due to higher conservation during evolution, this peptide family is more appropriately called the NPY family (4, 5). Sequence comparison indicates that NPY is one of the most highly conserved neuroendocrine peptides known (4). This remarkable conservation of NPY implies an important functional role or roles. NPY-like peptides all consist of an N-terminal polyproline helix (residues 1-8) and an amphiphilic α-helix (residues 15-30), connected with a β-turn, creating a hairpin-like loop (6).

The NPY gene

A single open reading frame predicts a transcript which gives rise to a simple precursor for NPY, consisting of 97 amino acids (7). The precursor contains a 28 aa signal peptide, required for entry into the endoplasmic reticulum (ER) and secretion. The signal peptide is cleaved, resulting in a 69 aa prohormone where mature NPY (36 aa) is flanked at its C-terminus by 33 amino acids, three of which are a motif necessary for NPY amidation, critical for virtually all actions of NPY. The peptide formed by the remaining 30 amino acids of the precursor has been named CPON (C flanking peptide of NPY). CPON is conserved to a degree that is lower than NPY but still significant (4). Despite this, its function remains unknown.

NPY expression in brain areas involved in emotionality

NPY-positive neurons are abundant in the central nervous system, including brain areas involved in emotionality (8-10). Within the cerebral cortex and forebrain nuclei, NPY is mainly present in inhibitory interneurons. Coexistence of NPY with somatostatin and nitric oxide

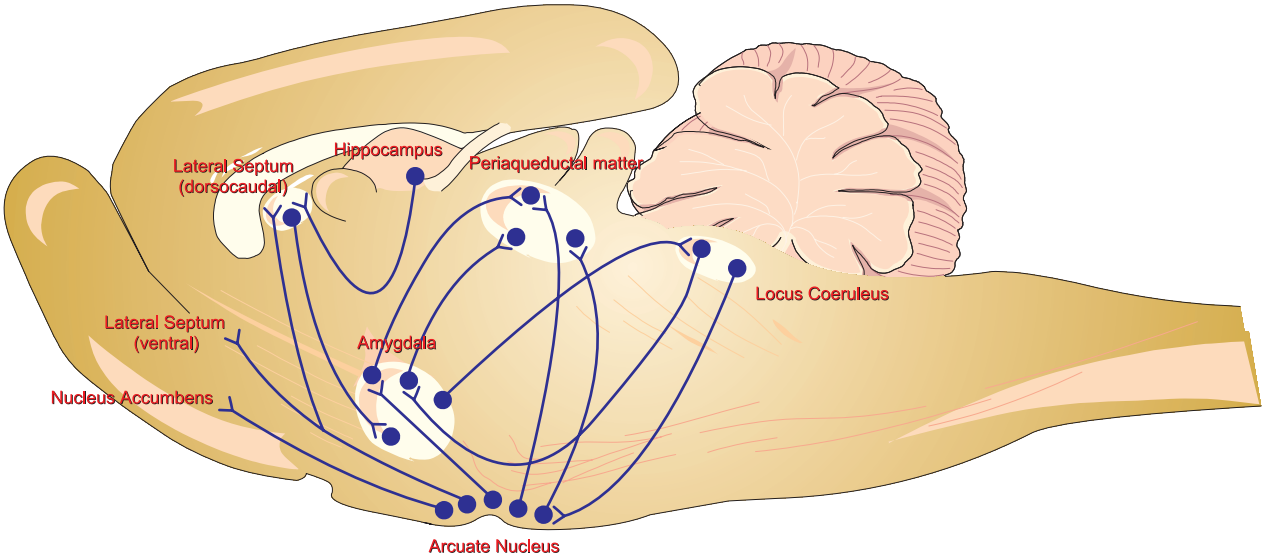

Fig. 1. Distribution of major NPY circuits in the rat brain involved in emotionality.

synthase is common in cortex and striatum. In addition, within the cortex and amygdala, NPY is extensively colocalized with GABA (10).

NPY-positive neurons in the hypothalamus, brainstem and spinal cord are more heterogeneous. An important NPY-ergic pathway from the arcuate nucleus to the ipsilateral paraventricular neucleus of the hypothatlamus has been described (11) and is involved in feeding effects of NPY. More recently, a coexistence was also reported of NPY and agouti gene-related protein (AGRP) in arcuate hypothalamic neurons. Among these, numerous cells project to telencephalic brain areas involved in emotionality, such as the amygdala complex (12) (Fig. 1).

NPY acts through heterogeneous receptors

The nomenclature Y_1 and Y_2 was introduced to denote the receptor that required the entire NPY (or PYY) molecule for activation (Y_1), and a subtype selectively stimulated by the long C-terminal NPY fragments (Y_2), respectively (13). Soon thereafter, a similar heterogeneity was found for brain-mediated actions of NPY (14). Molecular cloning has revealed additional diversity of the NPY receptor family. NPY receptors cloned to date all belong to the superfamily of G-protein-coupled receptors. The NPY Y_1 receptor, one of the subtypes implicated i.a. in feeding effects of NPY (15-17), requires the intact NPY sequence for recognition and activation and appears to be the subtype mediating antistress actions of NPY (18-22). The Y_2 receptor subtype is also activated by C-terminal fragments of NPY such as NPY_{13-36} (23, 24). A high number of Y_2 sites is found within the hippocampus. Activation of Y_2 receptors within this structure has been shown to suppress hippocampal glutamatergic transmission through presynaptic mechanisms (25, 26). Behavioral consequences of Y_2 signaling in this area are unclear. Although the existence of a Y_3 receptor has been postulated on the basis of pharmacological experiments (27), this has not been confirmed by molecular studies (28). Furthermore, a receptor termed Y_4 has been cloned, but appears to preferentially bind PP, and is therefore more appropriately referred to as a PP receptor (29, 30). Finally, a Y_5 receptor with restricted hypothalamic expression has been cloned and postulated to mediate the profound effects of NPY on feeding (31). Subsequent work indicates that the Y_5 receptor probably shares this role with the Y_1 subtype.

Antistress and antianxiety actions of NPY

Sedative effects of central NPY

Consistent with initial EEG findings (32), early behavioral studies demonstrated that i.c.v. administration of high central NPY doses (1.0-5.0 nmol) produced behavioral sedation. Of interest from a dependence point of view, no apparent tolerance was seen upon repeated administration. Effects of a single i.c.v. dose lasted up to 3 days but were still fully reversible (33). In another similarity with sedative drugs, i.c.v. pretreatment with NPY largely prevented the formation of gastric erosions normally produced by cold water immersion stress (34). These findings indicated shared properties between NPY and several classes of sedative compounds, including alcohol, benzodiazepines and barbiturates. This similarity was subsequently confirmed by EEG experiments in awake animals, where i.c.v. NPY produced a pattern resembling that produced by benzodiazepines (35).

Observations suggesting similarities between these classes of compounds have more recently been extended by findings of anticonvulsant actions of NPY (36, 37), which possibly provide a mechanism for the anticonvulsant state seen following electroconvulsive treatment (38-40). Evidence for shared properties between NPY and sedative compounds was also provided by mutual substitution of NPY and EtOH with regard to electrophysiological effects (41) and potentiation by NPY of barbiturate induced sleep (42). The latter study mapped out NPY effects on sedation to the posterior hypothalamus, an area involved in the regulation of sleep-wake cycles.

Initial studies suggested that sedative NPY effects are mediated by Y_1 receptors (43). Supporting the original conclusion, Y_1 mediation of NPY-induced sedation was demonstrated using Y_1 receptor knockouts (42). In contrast, anticonvulsant actions of NPY appear to be mediated through Y_5 receptors (44).

NPY is active in a wide range of anxiety models

It has been debated whether different animal models in the anxiety area reflect different aspects of clinical anxiety disorders, different disease entities, or perhaps are simply limited in their ability to model the relevant clinical disease (45-48). Against this background, a remarkable feature of NPY is its ability to produce antistress/antianxiety effects in all models tested thus far, suggesting that it acts at a core process underlying fear- and stress-related behaviors and affect, common to different models and disorders.

The Vogel (49) and Geller-Seifter (50) tests, commonly referred to as “conflict tests”, are the classical industry screening tests for benzodiazepine-like antianxiety action. In both tests, benzodiazepines reverse fear suppression of behavior. So does i.c.v. administration of NPY, with a high potency (51, 52). Control experiments (51) demonstrate that antinociceptive effects of NPY (53-56) do not contribute to these effects. The classical conflict tests do not probe the natural behavioral repertoire of the animal. The elevated plus maze (57) and the social interaction test (58) are more attractive in that respect. In both models, classical antianxiety compounds counteract fear suppression of the respective spontaneous exploratory behavior, and so does NPY, with high potency and specificity (21, 22, 51). A similar observation has been reported in the light-dark compartment test, which is conceptually related to the elevated plus maze (59). Finally, the fear potentiated startle model differs from most other anxiety paradigms in that it is based on fear potentiation rather than inhibition of behavior. It is less sensitive to effects on motor performance or motivation (60). NPY effectively reverses the potentiation of the acoustic startle response which accompanies conditioned fear, without affecting basal, unconditioned startle (21). Consistent actions of NPY both in models based on response inhibition and response facilitation indicate that the observed behavioral actions are indeed specifically related to emotionality.

Receptor pharmacology of NPY’s antistress actions

Full-length NPY peptide as well as [$Leu^{31}Pro^{34}$]NPY (61) potently and dose-dependently produce an anxiolytic-like effect in the elevated plus maze, Vogel test (51) and Geller-Seifter punished responding test (18), while the C-terminal, presumably Y_2-selective fragment NPY_{13-36}, does not reproduce this action. This is consistent with a “non-Y_2” profile, within which early antisense data pointed to Y_1 receptors (19). An involvement of Y_4 receptors seems to have been excluded using PP (62). Based on receptor distribution, it has been suggested that, similar to regulation of food intake, Y_5 receptors may be involved in emotionality in concert with Y_1 receptors (63). Under basal conditions, the selective Y_5 antagonist CGP-71683A does not influence anxiety-related behaviors in the social interaction test, the elevated plus maze or the open field (64), but Y_5 receptors in basolateral amygdala area are clearly capable of mediating anxiolytic actions (65).

The more recent arrival of subtype-selective, nonpeptide NPY receptor antagonists has allowed a more refined analysis of this issue. The first member in this class of compounds, BIBP-3226 acts as a Y_1 antagonist both *in vitro* and *in vivo*, while being devoid of affinity for Y_2, Y_4 and Y_5 receptors (66). In agreement with the above studies, BIBP-3226 was reported to suppress plus maze exploration following both i.c.v. (67) and localized site injections (68-70). BIBP-3226 has limited solubility, and is also clearly capable of producing nonreceptor-mediated effects (71). Furthermore, it has recently been reported that BIBP-3226 has a moderate but non-negligible affinity for NPFF receptors (72). Taken together, these observations raise some concern regarding findings obtained using this tool, in particular since anxiety-related behaviors are highly sensitive to stressful side effects of drugs. The arrival of the structurally related Y_1 antagonist BIBO-3304 has helped resolve these issues, as this compound has increased solubility, higher affinity for Y_1 receptors and appears to lack the nonspecific side effects (73). Studies using this compound confirm the results obtained using agonists, antisense oligonucleotides and BIBP-3226. For instance, anxiolytic-like effects of NPY in the social interaction test are blocked by intra-amygdala administration of BIBO-3304 (22), and this compound also increased defecation during exploratory behavior in the open field, a variable normally correlated with the level of emotionality (74).

Y_2 receptors may also play an important role in the regulation of emotionality and, in fact, offer the most attractive target for drug development efforts, as discussed below. NPY Y_2 receptors are located presynaptically on NPY-ergic neurons, and control the release of endogenous NPY (75, 76). Antagonism at these receptors would thus be expected to potentiate endogenous NPY release, and through this mechanism offer an “NPY mimetic” without the requirement for developing a Y_1 agonist. Available data in an alcohol self-administration model provide support for this notion (77), as does the

recent report of a consistent, anxiolytic-like phenotype in a Y_2 receptor null mutant (78).

Anatomical substrates of NPY's antistress actions

The amygdala is crucial for emotional learning, and coordinates behavioral, autonomic and endocrine fear responses (79-81), making it an obvious candidate for mediating anxiolytic-like actions of NPY. Indeed, site-specific injections of NPY and NPY analogs into the amygdala reproduced the antianxiety actions of NPY administered i.c.v. at approximately 10-fold lower doses, while other injection sites were ineffective in this respect (18, 22). Amygdala injections have also helped separate anxiolytic-like from appetitive effects of NPY, since they reproduce anxiolytic actions of NPY administered i.c.v., without affecting feeding. Central amygdala constitutes an output relay for the functional consequences of amygdala activation by fearful stimuli (79-82) and was initially reported to be the amygdalar compartment within which anxiolytic-like actions are produced by amygdala injections of NPY (18). However, subsequent work employing microinjections of smaller volumes has prompted a reevaluation of the data, suggesting that the lateral/basolateral complex in fact mediates antistress effects of NPY within the amygdala (22).

The periaqueductal gray matter (PAG) is involved in the behavioral output of fear responses, with subcompartments differentially involved in defensive behaviors (80, 83). Its dorsolateral compartment (DPAG) has been suggested to tonically inhibit the amygdala. Microinjections of the Y_1-selective nonpeptide antagonist BIBP-3226 within the DPAG have been reported to produce anxiogenic-like effects in the elevated plus maze, with behavioral specificity demonstrated by unaffected open field behavior following the same treatment (70). Similar effects were found in a separate report using the social interaction test (69). In the latter study, the di-peptide Y_1 antagonist 1229U91 mimicked the action of BIBP-3226, offsetting the limitations of BIBP-3226.

Septo-hippocampal circuits are important for fear-related behaviors, consistent with observations that dorsal hippocampus is an important component of neuronal circuitry controlling anxiety-related behaviors and stress responses (84, 85). NPY microinjections into the lateral septum reproduced anxiolytic-like actions of i.c.v. administered NPY and reversed the anxiogenic action of corticotropin-releasing factor (CRF). The anxiolytic-like action of NPY was clearly Y_1 receptor-mediated, as it was blocked by the highly selective, nontoxic Y_1 receptor antagonist BIBO-3304 (86). This study also demonstrated that NPY injections into the cholinergic medial septal nucleus were ineffective, contradicting the previous suggestion that anxiolytic-like actions of NPY might be produced through interactions with cholinergic afferents to the hippocampus which originate from this structure (87).

Finally, the locus coeruleus has long been implicated in anxiety disorders and stress (88, 89), although its involvement may be restricted to symptom domains of pathological arousal and vigilance seen in these disorders, as a reflection of the normal physiology of this structure (90). As mentioned above, anxiolytic-like actions have been reported in the social interaction test after local injections of 10 pmol NPY into the locus coeruleus (68). Based on the ligand profile of this effect, and the inactivity of BIBP-3226 in this region, it was suggested that NPY Y_2 receptors in this area mediate these actions. However, an alternative interpretation which needs to be considered is whether this reflects actions at presynaptic Y_2 receptors on A6 neurons in which NPY and norepinephrine are colocalized, ultimately inhibiting noradrenergic transmission in projection areas of these neurons.

Expression of NPY and behavioral stress responses

On the basis of the pharmacological observations above and early expression studies (91, 92), we proposed that an upregulation of NPY expression may contribute to successful behavioral adaptation to stress. This extends a previously introduced hypothesis that NPY may act to "buffer" behavioral effects of stress-promoting signals such as CRF (93). Our hypothesis predicted that upregulated expression of NPY should render a subject less sensitive to anxiety-promoting effects of stress, a prediction potentially possible to test in a transgenic system. The generation of an NPY transgenic rat offered an attractive model for our studies. In summary, results obtained in the NPY transgenic rat model have supported the intial hypothesis. No anxiety-related phenotype was observed in the elevated plus maze under basal, unstressed conditions. However, a marked behavioral insensitivity to stress was found when stressful manipulations were introduced. In nontransgenic littermate controls, exposure to an established stressor (1 h of restraint) 1 h prior to behavioral testing gave rise to an expected and marked anxiogenic effect on the plus maze, manifested as a profound decrease of the percentage of time spent exploring the open arms of the maze and of the number of entries into the open arms. This behavioral consequence of stress was completely absent in transgenic subjects. Furthermore, in the markedly stressful Vogel test, response inhibition is normally seen due to the delivery of electric shock upon drinking, and this was indeed observed in nontransgenic littermates. In contrast, and similar to benzodiazepine-treated animals, response inhibition in this test was completely absent in NPY transgenic subjects (94).

NPY and depression

The anxiety-depression spectrum

Symptoms of anxiety and depression commonly coexist, and both disorders are thought to reflect maladaptive changes in stress responsive systems (95). In fact, a

latent class analysis of clinical symptoms has suggested that the present classification of depressive and anxiety disorders may be artificial and suggested the existence of a category labeled "major depression-generalized anxiety disorder" (96). In agreement with this notion, genetic factors which confer increased vulnerability for both of these disorders are largely the same (97). Given the extensive evidence for an involvement of central NPY in stress and anxiety, it is therefore not unexpected that this system has also been implicated in depressive disorder, although less extensive data are available in this area.

Effects of antidepressant treatments on the NPY system

Inconsistent effects of treating experimental animals with clinically effective antidepressants have been reported, possibly due to different dosing, treatment intervals and pharmacokinetics (98-101). A region-specific regulation of NPY and Y_1 receptor expression was then reported following chronic treatment with the long-acting serotonin-selective reuptake inhibitor (SSRI) fluoxetine, both in the Flinders Sensitive Line (FSL), a genetic model of depression (102), and the corresponding control Flinders Resistant Line (FRL) (103, 104). On the basis of these findings, an involvement of NPY was suggested in the antidepressant effect of fluoxetine.

Another established and effective antidepressant treatment, electroconvulsive shock (ECS), has been much more consistent in upregulating brain NPY levels, with the hippocampus as a seemingly central target. This was originally reported independently by two different groups, both of which also demonstrated elevation of hippocampal NPY levels after repeated, but not single ECS, paralleling the requirements for clinical effect in depressed subjects (105, 106). These findings have subsequently been replicated and extended (107-111).

Finally, similar to what has been reported with ECS and some pharmacological antidepressant treatments, administration of the clinically established affective stabilizer lithium also leads to an upregulation of hippocampal NPY synthesis (107).

NPY in animal models of depression

Further evidence for an involvement of NPY in depression comes from the findings of differential NPY expression in two different genetic animal model of depression, the FSL described above (103, 104, 112), and the Fawn Hooded rat (109, 113). The hippocampus appears to be a consistent candidate structure for a possible functional involvement.

Olfactory bulbectomy in rats produces behavioral consequences which have been interpreted as indicative of a depression-like phenotype (114). It is therefore of interest that some of these, normally interpreted as signs of increased irritability during open field exploration, are counteracted by subchronic administration of NPY (115), while in the long run (2-4 weeks) NPY expression was upregulated in piriform cortex and the dentate hippocampal gyrus by olfactory bulbectomy, possibly as a compensatory mechanism (116).

Early postnatal separation of rat pups induces a long-term phenotype characterized by increased responsiveness of the HPA axis (117). Although not extensively validated pharmacologically as a model for detecting antidepressant drug action, this model is attractive in that it closely mimics dysregulations of the HPA axis thought to be at the core of pathophysiology in depressed humans (95). It is therefore of interest that maternal separation has been shown to reduce NPY (118, 119). Reductions of NPY-like immunoreactivity in this model were found within the hippocampus, consistent with the findings of increased hippocampal NPY following several types of antidepressant treatments.

Consistent with the observations reviewed above, NPY given i.c.v. produces antidepressant-like effects in the forced swimming test, both in rats (120) and mice (121), and a similar effect is seen as a result of a Y_2 null mutation thought to potentiate NPY signaling (78). A common final pathway may mediate effects of conventional antidepressant drugs and NPY, since, at least *in vitro*, NPY shares the ability of the former to stimulate hippocampal neurogenesis (122).

Is there a primary NPY disturbance in human depression?

An early study reported decreased levels of NPY in the cerebrospinal fluid (CSF) of patients with major depression, possibly reflecting decreased central availibility of NPY (123). Within this patient population, a more detailed analysis revealed an inverse correlation between NPY levels in the CSF and ratings of anxiety symptoms. Independently, lower levels of NPY in brain tissue were also reported in suicide victims (124). These findings indicate that the NPY system might be primarily affected in depressive syndromes and contribute to the clinical symptomatology. However, neither the finding of reduced NPY levels in CSF of depressed subjects (125) nor in frontal cortex of depressed subjecta (126) was replicated in subsequent studies. The reason for this is unclear, but may have been related to the limited understanding of peptide processing and assay specificity issues in that early era, in particular since mass spectrometry studies have since shown that processing of NPY may differ between depressed subjects and normals, resulting in a different fragment pattern (127). In a recent reexamination of this issue, we used a sample of therapy-refractory depressed patients of considerable size for this type of study (n=50), and analyzed CSF levels of monoamines, monoamine metabolites and several neuropeptides. NPY was analyzed using an assay which has been extensively characterized with regard to specificity and lacks cross-reactivity with C-terminal NPY fragments. The one robust difference between patients and controls using this

approach was a highly significant, 30% reduction of CSF (Heilig *et al*., in press). It remains to be established whether this reflects improved methodology, altered NPY signaling in subpopulations of depressed subjects only, or both. The latter possibility is supported by the demonstration of suppressed NPY expression in human postmortem brain tissue in bipolar, but not unipolar, affective disorder (128).

Human findings – periphery

In attempts to link NPY to psychiatric disorders, studies of the more easily accessible peripheral compartment have also been carried out. Presently available evidence suggests that peripheral NPY is a marker of sympathetic nervous system activity, unrelated to central NPY signaling of importance for emotionality and mood. However, it has been suggested that a functional polymorphism in the preproneuropeptide Y gene leads to altered NPY expression and/or release (129), and this kind of mechanism might affect central and peripheral NPY levels in a similar manner, making the latter a marker of the former. In this context, peripheral NPY has been reported to be lowered in recent suicide attempters (130) and to correlate with personality variables in this patient population (131). In a separate line of study, lowered peripheral NPY levels, both at baseline and upon stimulation of sympathetic neurons with the presynaptic autoreceptor blocker yohimbine, have been found in combat survivors with posttraumatic stress disorder (132). In an interesting experimental follow-up of these findings, NPY was analyzed during exposure to interrogation, presumably a highly stressful experience, in special force as well as nonspecial force soldiers. The former had generally lower levels of plasma NPY. Furthermore, across both groups, a range of responses to this stressful situation was inversly correlated with plasma NPY. On the basis of these findings, it was suggested that peripheral NPY is a marker of stress resiliance (133), possibly through a correlation with personality traits (134). It remains to be established whether these findings reflect altered patterns of sympathetic activity, altered central NPY signaling, or both.

NPY and alcohol dependence

A link between NPY signaling and regulation of alcohol consumption was simultaneously suggested by two groups. One group demonstrated that transgenic overexpression of NPY in mice leads to reduced self-administration of alcohol, while homologous recombination knock-out of NPY produces the opposite result (135); the other group based the connection on EEG analysis (41). The former group has subequently also shown that deletion of the Y_1 gene increases alcohol intake. Consistent with these data, a quantitative trait locus (QTL) identified in the alcohol preferring P line spans the NPY gene (136, 137). P rats, and the genetically selected alcohol preferring AA rat line differ from nonpreferring rats in NPY, and, in the latter case, NPY Y_2 receptor expression in several brain regions. The pattern may point to the amygdala as a region mediating the role of NPY in this context (138-140).

In humans, an association between a functional Leu^7Pro polymorphism in the NPY gene and high alcohol consumption has been reported (141, 142). A report also found an association between this marker and alcohol dependence (143), but this finding has not been supported by subsequent analysis (144).

Whether primarily involved in the pathogenesis of human alcoholism or not, the NPY system is likely to offer an attractive target for treatment of alcohol dependence. In normal rats, direct i.c.v. administration of NPY does not affect alcohol intake (145) or operant alcohol self-administration (140). However, a marked suppression of ethanol intake is seen in conditions of excessive intake, such as in the genetically selected, ethanol preferring P line (146), the alcohol preferring HAD line (147) as well as in genetically heterogenous subjects with a history of dependence (Thorsell *et al*., in preparation). Furthermore, i.c.v. administration of the selective Y_2 antagonist BIIE-0246 suppresses operant self-administration of NPY (77), presumably by blocking presynaptic autoreceptors for NPY and thereby facilitating release of endogenous NPY (75, 76), and its subsequent action at Y_1 receptors. This action is much more pronounced in subjects in whom a history of dependence has led to increased alcohol intake (Thorsell *et al*., in preparation).

Conclusions

In the 20 years since its isolation, numerous actions have been discovered for NPY. Several of these, most notably its blood pressure effects and effects on feeding, gave rise to hopes that compounds targeting the NPY system could be developed for the treatment of clinical disorders. With the perspective of the last two decades, the conditions where this hope not only persists, but has in fact been strengthened, are psychiatric disorders, most notably anxiety disorders, major depression and alcohol dependence. To realize these hopes, development of treatments potentiating Y_1 receptor signaling appears to be necessary. The most promising strategy is the development of Y_2 receptor antagonists aimed at potentiating endogenous NPY signaling.

References

1. Kimmel, J.R., Hayden, L.J., Pollock, H.G. *Isolation and characterization of a new pancreatic polypeptide hormone.* J Biol Chem 1975, 250: 9369-76.

2. Tatemoto, K. *Neuropeptide Y: Complete amino acid sequence of the brain peptide.* Proc Natl Acad Sci USA 1982, 79: 5485-9.

3. Tatemoto, K., Carlquist, M., Mutt, V. *Neuropeptide Y – a novel brain peptide with structural similarities to peptide YY and pancreatic polypeptide.* Nature 1982, 296: 659-60.

4. Larhammar, D., Söderberg, C., Blomqvist, A.G. *Evolution of the neuropeptide Y family of peptides.* In: The Biology of Neuropeptide Y and Related Peptides. Colmers, W.F., Wahlestedt, C. (Eds.). Humana Press: Totowa 1993, 1-41.

5. Wraith, A., Tornsten, A., Chardon, P. et al. *Evolution of the neuropeptide Y receptor family: Gene and chromosome duplications deduced from the cloning and mapping of the five receptor subtype genes in pig.* Genome Res 2000, 10: 302-10.

6. Schwartz, T.W., Fuhlendorff, J., Kjems, L.L. et al. *Signal epitopes in the three-dimensional structure of neuropeptide Y. Interaction with Y_1, Y_2, and pancreatic polypeptide receptors.* Ann NY Acad Sci 1990, 611: 35-47.

7. Minth, C.D., Bloom, S.R., Polak, J.M., Dixon, J.E. *Cloning, characterization, and DNA sequence of a human cDNA encoding neuropeptide tyrosine.* Proc Natl Acad Sci USA 1984, 81: 4577-81.

8. De Quidt, M.E., Emson, P.C. *Distribution of neuropeptide Y-like immunoreactivity in the rat central nervous system–I. Radioimmunoassay and chromatographic characterisation.* Neuroscience 1986, 18: 527-43.

9. De Quidt, M.E., Emson, P.C. *Distribution of neuropeptide Y-like immunoreactivity in the rat central nervous system–II. Immunohistochemical analysis.* Neuroscience 1986, 18: 545-618.

10. Hendry, S.H. *Organization of neuropeptide Y neurons in the mammalian central nervous system.* In: The Biology of Neuropeptide Y and Related Peptides. Colmers, W.F., Wahlestedt, C. (Eds.). Humana Press: Totowa 1993, 65-156.

11. Bai, F.L., Yamano, M., Shiotani, Y. et al. *An arcuato-paraventricular and -dorsomedial hypothalamic neuropeptide Y-containing system which lacks noradrenaline in the rat.* Brain Res 1985, 331: 172-5.

12. Broberger, C., Johansen, J., Johansson, C., Schalling, M., Hokfelt, T. *The neuropeptide Y/agouti gene-related protein (AGRP) brain circuitry in normal, anorectic, and monosodium glutamate-treated mice.* Proc Natl Acad Sci USA 1998, 95: 15043-8.

13. Wahlestedt, C., Yanaihara, N., Håkanson, R. *Evidence for different pre- and post-junctional receptors for neuropeptide Y and related peptides.* Regul Pept 1986, 13: 307-1.

14. Heilig, M., Vecsei, L., Widerlöv, E. *Opposite effects of centrally administered neuropeptide Y (NPY) on locomotor activity of spontaneously hypertensive (SH) and normal rats.* Acta Physiol Scand 1989, 137: 243-8.

15. Eva, C., Keinanen, K., Monyer, H., Seeburg, P., Sprengel, R. *Molecular cloning of a novel G protein-coupled receptor that may belong to the neuropeptide receptor family.* FEBS Lett 1990, 271: 81-4.

16. Herzog, H., Hort, Y.J., Ball, H.J., Hayes, G., Shine, J., Selbie, L.A. *Cloned human neuropeptide Y receptor couples to two different second messenger systems.* Proc Natl Acad Sci USA 1992, 89: 5794-8.

17. Larhammar, D., Blomqvist, A.G., Yee, F., Jazin, E., Yoo, H., Wahlestedt, C. *Cloning and functional expression of a human neuropeptide Y/peptide YY receptor of the Y_1 type.* J Biol Chem 1992, 267: 10935-8.

18. Heilig, M., McLeod, S., Brot, M. et al. *Anxiolytic-like action of neuropeptide Y: Mediation by Y_1 receptors in amygdala, and dissociation from food intake effects.* Neuropsychopharmacology 1993, 8: 357-63.

19. Wahlestedt, C., Pich, E.M., Koob, G.F., Yee, F., Heilig, M. *Modulation of anxiety and neuropeptide Y-Y_1 receptors by antisense oligodeoxynucleotides.* Science 1993, 259: 528-31.

20. Heilig, M. *Antisense inhibition of neuropeptide Y (NPY)-Y_1 receptor expression blocks the anxiolytic like action of NPY in amygdala and paradoxically increases feeding.* Regul Pept 1995, 59: 201-5.

21. Broqua, P., Wettstein, J.G., Rocher, M.N., Gauthier-Martin, B., Junien, J.L. *Behavioral effects of neuropeptide receptor agonists in the elevated plus-maze and fear-potentiated startle procedure.* Behav Pharmacol 1995, 6: 215-22.

22. Sajdyk, T.J., Vandergriff, M.G., Gehlert, D.R. *Amygdalar neuropeptide Y Y-1 receptors mediate the anxiolytic-like actions of neuropeptide Y in the social interaction test.* Eur J Pharmacol 1999, 368: 143-7.

23. Sheikh, S.P., Håkansson, R., Schwartz, T.W. *Y_1 and Y_2 receptors for neuropeptide Y.* FEBS Lett 1989, 245: 209-14.

24. Gerald, C., Walker, M.W., Vaysse, P.J., He, C., Branchek, T.A., Weinshank, R.L. *Expression cloning and pharmacological characterization of a human hippocampal neuropeptide Y/peptide YY Y_2 receptor subtype.* J Biol Chem 1995, 270: 26758-61.

25. McQuiston, A.R., Colmers, W.F. *Neuropeptide Y_2 receptors inhibit the frequency of spontaneous but not miniature EPSCs in CA3 pyramidal cells of rat hippocampus.* J Neurophysiol 1996, 76: 3159-68.

26. Qian, J., Colmers, W.F., Saggau, P. *Inhibition of synaptic transmission by neuropeptide Y in rat hippocampal area CA1: Modulation of presynaptic Ca^{2+} entry.* J Neurosci 1997, 17: 8169-77.

27. Glaum, S.R., Miller, R.J., Rhim, H. et al. *Characterization of Y_3 receptor-mediated synaptic inhibition by chimeric neuropeptide Y-peptide YY peptides in the rat brainstem.* Br J Pharmacol 1997, 120: 481-7.

28. Jazin, E.E., Yoo, H., Blomqvist, A.G. et al. *A proposed bovine neuropeptide Y (NPY) receptor cDNA clone, or its human homologue, confers neither NPY binding sites nor NPY responsiveness on transfected cells.* Regul Pept 1993, 47: 247-58.

29. Bard, J.A., Walker, M.W., Branchek, T.A., Weinshank, R.L. *Cloning and functional expression of a human Y_4 subtype receptor for pancreatic polypeptide, neuropeptide Y, and peptide YY.* J Biol Chem 1995, 270: 26762-5.

30. Lundell, I., Blomqvist, A.G., Berglund, M.M. et al. *Cloning of a human receptor of the NPY receptor family with high affinity for pancreatic polypeptide and peptide YY.* J Biol Chem 1995, 270: 29123-8.

31. Gerald, C., Walker, M.W., Criscione, L. et al. *A receptor subtype involved in neuropeptide-Y-induced food intake.* Nature 1996, 382: 168-71.

32. Fuxe, K., Agnati, L.F., Härfstrand, A. et al. *Central administration of neuropeptide Y induces hypotension bradypnea and*

EEG synchronization in the rat. Acta Physiol Scand 1983, 118: 189-92.

33. Heilig, M., Murison, R. *Intracerebroventricular neuropeptide Y suppresses open field and home cage activity in the rat.* Regul Pept 1987, 19: 221-31.

34. Heilig, M., Murison, R. *Intracerebroventricular neuropeptide Y protects against stress-induced gastric erosion in the rat.* Eur J Pharmacol 1987, 137: 127-9.

35. Ehlers, C.L., Somes, C., Lopez, A., Kirby, D., Rivier, J.E. *Electrophysiological actions of neuropeptide Y and its analogs: New measures for anxiolytic therapy?* Neuropsychopharmacology 1997, 17: 34-43.

36. Woldbye, D.P., Larsen, P.J., Mikkelsen, J.D., Klemp, K., Madsen, T.M., Bolwig, T.G. *Powerful inhibition of kainic acid seizures by neuropeptide Y via Y_5-like receptors.* Nat Med 1997, 3: 761-4.

37. Baraban, S.C., Hollopeter, G., Erickson, J.C., Schwartzkroin, P.A., Palmiter, R.D. *Knock-out mice reveal a critical antiepileptic role for neuropeptide Y.* J Neurosci 1997, 17: 8927-36.

38. Bolwig, T.G., Woldbye, D.P., Mikkelsen, J.D. *Electroconvulsive therapy as an anticonvulsant: a possible role of neuropeptide Y (NPY).* J ECT 1999, 15: 93-101.

39. Mikkelsen, J.D., Woldbye, D., Kragh, J., Larsen, P.J., Bolwig, T.G. *Electroconvulsive shocks increase the expression of neuropeptide Y (NPY) mRNA in the piriform cortex and the dentate gyrus.* Brain Res Mol Brain Res 1994, 23: 317-22.

40. Woldbye, D.P., Madsen, T.M., Larsen, P.J., Mikkelsen, J.D., Bolwig, T.G. *Neuropeptide Y inhibits hippocampal seizures and wet dog shakes.* Brain Res 1996, 737: 162-8.

41. Ehlers, C.L., Somes, C., Cloutier, D. *Are some of the effects of ethanol mediated through NPY?* Psychopharmacology 1998, 139: 136-44.

42. Naveilhan, P., Canals, J.M., Valjakka, A., Vartiainen, J., Arenas, E., Ernfors, P. *Neuropeptide Y alters sedation through a hypothalamic Y_1-mediated mechanism.* Eur J Neurosci 2001, 13: 2241-6.

43. Heilig, M., Wahlestedt, C., Widerlöv, E. *Neuropeptide Y (NPY)-induced suppression of activity in the rat: Evidence for NPY receptor heterogeneity and for interaction with alpha-adrenoceptors.* Eur J Pharmacol 1988, 157: 205-13.

44. Marsh, D.J., Baraban, S.C., Hollopeter, G., Palmiter, R.D. *Role of the Y_5 neuropeptide Y receptor in limbic seizures.* Proc Natl Acad Sci USA 1999, 96: 13518-23.

45. Rodgers, R.J. *Animal models of 'anxiety': Where next?* Behav Pharmacol 1997, 8: 477-96.

46. Hogg, S. *A review of the validity and variability of the elevated plus-maze as an animal model of anxiety.* Pharmacol Biochem Behav 1996, 54: 21-30.

47. McCreary, A.C., McBlane, J.W., Spooner, H.A., Handley, S.L. *5-HT systems and anxiety: Multiple mechanisms in the elevated X-maze.* Pol J Pharmacol 1996, 48: 1-12.

48. File, S.E. *The interplay of learning and anxiety in the elevated plus-maze.* Behav Brain Res 1993, 58: 199-202.

49. Vogel, J.R., Beer, B., Clody, D.E. *A simple and reliable conflict procedure for testing anti-anxiety agents.* Psychopharmacologia 1971, 21: 1-7.

50. Geller, I., Seifter, J. *The effects of meprobamate, barbiturates, d-amphetamine and promazine on experimentally induced conflict in the rat.* Psychopharmacologia 1960, 1: 482-92.

51. Heilig, M., Söderpalm, B., Engel, J.A., Widerlöv, E. *Centrally administered neuropeptide Y (NPY) produces anxiolytic- like effects in animal anxiety models.* Psychopharmacology 1989, 98: 524-9.

52. Heilig, M., McLeod, S., Koob, G.F., Britton, K.T. *Anxiolytic-like effect of neuropeptide Y (NPY), but not other peptides in an operant conflict test.* Regul Pept 1992, 41: 61-9.

53. Hua, X.Y., Boublik, J.H., Spicer, M.A., Rivier, J.E., Brown, M.R., Yaksh, T.L. *The antinociceptive effects of spinally administered neuropeptide Y in the rat: Systematic studies on structure-activity relationship.* J Pharmacol Exp Ther 1991, 258: 243-8.

54. Mellado, M.L., Gibert-Rahola, J., Chover, A.J., Mico, J.A. *Effect on nociception of intracerebroventricular administration of low doses of neuropeptide Y in mice.* Life Sci 1996, 58: 2409-14.

55. Wang, J.Z., Lundeberg, T., Yu, L. *Antinociceptive effects induced by intra-periaqueductal grey administration of neuropeptide Y in rats.* Brain Res 2000, 859: 361-3.

56. Broqua, P., Wettstein, J.G., Rocher, M.N. et al. *Antinociceptive effects of neuropeptide Y and related peptides in mice.* Brain Res 1996, 724: 25-32.

57. Pellow, S., Chopin, P., File, S.E., Briley, M. *Validation of open:closed arm entries in an elevated plus-maze as a measure of anxiety in the rat.* J Neurosci Methods 1985, 14: 149-67.

58. File, S.E. *The use of social interaction as a method for detecting anxiolytic activity of chlordiazepoxide-like drugs.* J Neurosci Methods 1980, 2: 219-38.

59. Pich, E.M., Agnati, L.F., Zini, I., Marrama, P., Carani, C. *Neuropeptide Y produces anxiolytic effects in spontaneously hypertensive rats.* Peptides 1993, 14: 909-12.

60. Davis, M., Rainnie, D., Cassell, M. *Neurotransmission in the rat amygdala related to fear and anxiety.* Trends Neurosci 1994, 17: 208-14.

61. Fuhlendorff, J., Gether, U., Aakerlund, L. *[Leu31, Pro34]neuropeptide Y: A specific Y_1 receptor agonist.* Proc Natl Acad Sci USA 1990, 87: 182-6.

62. Asakawa, A., Inui, A., Ueno, N., Fujimiya, M., Fujino, M.A., Kasuga, M. *Mouse pancreatic polypeptide modulates food intake, while not influencing anxiety in mice.* Peptides 1999, 20: 1445-8.

63. Parker, R.M., Herzog, H. *Regional distribution of Y-receptor subtype mRNAs in rat brain.* Eur J Neurosci 1999, 11: 1431-48.

64. Kask, A., Vasar, E., Heidmets, L.T., Allikmets, L., Wikberg, J.E. *Neuropeptide Y Y(5) receptor antagonist CGP71683A: The effects on food intake and anxiety-related behavior in the rat.* Eur J Pharmacol 2001, 414: 215-24.

65. Sajdyk, T.J., Schober, D.A., Gehlert, D.R. *Neuropeptide Y receptor subtypes in the basolateral nucleus of the amygdala modulate anxiogenic responses in rats.* Neuropharmacology 2002, 43: 1165-72.

66. Rudolf, K., Eberlein, W., Engel, W. et al. *The first highly potent and selective non-peptide neuropeptide Y Y_1 receptor antagonist: BIBP3226.* Eur J Pharmacol 1994, 271: R11-R13.

67. Kask, A., Rago, L., Harro, J. *Anxiogenic-like effect of the neuropeptide Y Y-1 receptor antagonist BIBP3226 – antagonism with diazepam.* Eur J Pharmacol 1996, 317: R3-R4.

68. Kask, A., Eller, M., Oreland, L., Harro, J. *Neuropeptide Y attenuates the effect of locus coeruleus denervation by DSP-4 treatment on social behaviour in the rat.* Neuropeptides 2000, 34: 58-61.

69. Kask, A., Rago, L., Harro, J. *NPY Y_1 receptors in the dorsal periaqueductal gray matter regulate anxiety in the social interaction test.* Neuroreport 1998, 9: 2713-6.

70. Kask, A., Rago, L., Harro, J. *Anxiogenic-like effect of the NPY Y_1 receptor antagonist BIBP3226 administered into the dorsal periaqueductal gray matter in rats.* Regul Pept 1998, 75-76: 255-62.

71. Doods, H.N., Wieland, H.A., Engel, W. et al. *BIBP3226, the first selective neuropeptide Y_1 receptor antagonist: A review of its pharmacological properties.* Regul Pept 1996, 65: 71-7.

72. Mollereau, C., Gouarderes, C., Dumont, Y. et al. *Agonist and antagonist activities on human $NPFF_2$ receptors of the NPY ligands GR231118 and BIBP3226.* Br J Pharmacol 2001, 133: 1-4.

73. Wieland, H.A., Engel, W., Eberlein, W., Rudolf, K., Doods, H.N. *Subtype selectivity of the novel nonpeptide neuropeptide Y Y_1 receptor antagonist BIBO 3304 and its effect on feeding in rodents.* Br J Pharmacol 1998, 125: 549-55.

74. Kask, A., Harro, J. *Inhibition of amphetamine- and apomorphine-induced behavioural effects by neuropeptide Y Y_1 receptor antagonist BIBO 3304*. Neuropharmacology 2000, 39: 1292-302.

75. King, P.J., Widdowson, P.S., Doods, H.N., Williams, G. *Regulation of neuropeptide Y release by neuropeptide Y receptor ligands and calcium channel antagonists in hypothalamic slices.* J Neurochem 1999, 73: 641-6.

76. King, P.J., Williams, G., Doods, H., Widdowson, P.S. *Effect of a selective neuropeptide Y Y_2 receptor antagonist, BIIE0246 on neuropeptide Y release.* Eur J Pharmacol 2000, 396: R1-R3.

77. Thorsell, A., Rimondini, R., Heilig, M. *Blockade of central neuropeptide Y (NPY) Y_2 receptors reduces ethanol self-administration in rats.* Neurosci Lett 2002, 332: 1-4.

78. Tschenett, A., Singewald, N., Carli, M. et al. *Reduced anxiety and improved stress coping ability in mice lacking NPY-Y_2 receptors.* Eur J Neurosci 2003, 18: 143-8.

79. Davis, M. *Are different parts of the extended amygdala involved in fear versus anxiety?* Biol Psychiatry 1998, 44: 1239-47.

80. Fendt, M., Fanselow, M.S. *The neuroanatomical and neurochemical basis of conditioned fear.* Neurosci Biobehav Rev 1999, 23: 743-60.

81. LeDoux, J.E. *Emotion circuits in the brain.* Annu Rev Neurosci 2000, 23: 155-84.

82. Swanson, L.W., Petrovich, G.D. *What is the amygdala?* Trends Neurosci 1998, 21: 323-31.

83. Brandao, M.L., Cardoso, S.H., Melo, L.L., Motta, V., Coimbra, N.C. *Neural substrate of defensive behavior in the midbrain tectum.* Neurosci Biobehav Rev 1994, 18: 339-46.

84. Andrews, N., File, S.E., Fernandes, C., Gonzalez, L.E., Barnes, N.M. *Evidence that the median raphe nucleus-dorsal hippocampal pathway mediates diazepam withdrawal-induced anxiety.* Psychopharmacology 1997, 130: 228-34.

85. Gonzalez, L.E., File, S.E., Overstreet, D.H. *Selectively bred lines of rats differ in social interaction and hippocampal 5-HT_{1A} receptor function: A link between anxiety and depression?* Pharmacol Biochem Behav 1998, 59: 787-92.

86. Kask, A., Nguyen, H.P., Pabst, R., von Horsten, S. *Neuropeptide Y Y_1 receptor-mediated anxiolysis in the dorsocaudal lateral septum: Functional antagonism of corticotropin-releasing hormone-induced anxiety.* Neuroscience 2001, 104: 799-806.

87. Zaborszky, L., Duque, A. *Local synaptic connections of basal forebrain neurons.* Behav Brain Res 2000, 115: 143-58.

88. Nestler, E.J., Alreja, M., Aghajanian, G.K. *Molecular control of locus coeruleus neurotransmission.* Biol Psychiatry 1999, 46: 1131-9.

89. Sullivan, G.M., Coplan, J.D., Kent, J.M., Gorman, J.M. *The noradrenergic system in pathological anxiety: A focus on panic with relevance to generalized anxiety and phobias.* Biol Psychiatry 1999, 46: 1205-18.

90. Aston-Jones, G., Chiang, C., Alexinsky, T. *Discharge of noradrenergic locus coeruleus neurons in behaving rats and monkeys suggests a role in vigilance.* Prog Brain Res 1991, 88: 501-20.

91. Thorsell, A., Svensson, P., Wiklund, L., Sommer, W., Ekman, R., Heilig, M. *Suppressed neuropeptide Y (NPY) mRNA in rat amygdala following restraint stress.* Regul Pept 1998, 75-6: 247-54.

92. Thorsell, A., Carlsson, K., Ekman, R., Heilig, M. *Behavioral and endocrine adaptation, and up-regulation of NPY expression in rat amygdala following repeated restraint stress.* Neuroreport 1999, 10: 3003-7.

93. Heilig, M., Koob, G.F., Ekman, R., Britton, K.T. *Corticotropin-releasing factor and neuropeptide Y: Role in emotional integration.* Trends Neurosci 1994, 17: 80-5.

94. Thorsell, A., Michalkiewicz, M., Dumont, Y. et al. *Behavioral insensitivity to restraint stress, absent fear suppression of behavior and impaired spatial learning in transgenic rats with hippocampal neuropeptide Y overexpression.* Proc Natl Acad Sci USA 2000, 97: 12852-7.

95. Holsboer, F. *The corticosteroid receptor hypothesis of depression.* Neuropsychopharmacology 2000, 23: 477-501.

96. Sullivan, P.F., Kendler, K.S. *Typology of common psychiatric syndromes. An empirical study.* Br J Psychiatry 1998, 173: 312-9.

97. Kendler, K.S., Neale, M.C., Kessler, R.C., Heath, A.C., Eaves, L.J. *Major depression and generalized anxiety disorder. Same genes, (partly) different environments?* Arch Gen Psychiatry 1992, 49: 716-22.

98. Heilig, M., Wahlestedt, C., Ekman, R., Widerlöv, E. *Antidepressant drugs increase the concentration of neuropeptide Y (NPY)-like immunoreactivity in the rat brain.* Eur J Pharmacol 1988, 147: 465-7.

99. Widdowson, P.S., Halaris, A.E. *Chronic desipramine treatment reduces regional neuropeptide Y binding to Y_2-type receptors in rat brain.* Brain Res 1991, 539: 196-202.

100. Bellman, R., Sperk, G. *Effects of antidepressant drug treatment on levels of NPY or prepro-NPY-mRNA in the rat brain.* Neurochem Int 1993, 22: 183-7.

101. Heilig, M., Ekman, R. *Chronic parenteral antidepressant treatment in rats: Unaltered levels and processing of neuropeptide Y (NPY) and corticotropin-releasing hormone (CRH).* Neurochem Int 1995, 26: 351-5.

102. Overstreet, D.H., Pucilowski, O., Rezvani, A.H., Janowsky, D.S. *Administration of antidepressants, diazepam and psychomotor stimulants further confirms the utility of Flinders Sensitive Line rats as an animal model of depression.* Psychopharmacology (Berl) 1995, 121: 27-37.

103. Caberlotto, L., Fuxe, K., Overstreet, D.H., Gerrard, P., Hurd, Y.L. *Alterations in neuropeptide Y and Y_1 receptor mRNA expression in brains from an animal model of depression: Region specific adaptation after fluoxetine treatment.* Brain Res Mol Brain Res 1998, 59: 58-65.

104. Caberlotto, L., Jimenez, P., Overstreet, D.H., Hurd, Y.L., Mathe, A.A., Fuxe, K. *Alterations in neuropeptide Y levels and Y_1 binding sites in the Flinders Sensitive Line rats, a genetic animal model of depression.* Neurosci Lett 1999, 265: 191-4.

105. Wahlestedt, C., Blendy, J.A., Kellar, K.J., Heilig, M., Widerlöv, E., Ekman, R. *Electroconvulsive shocks increase the concentration of neocortical and hippocampal neuropeptide Y (NPY)-like immunoreactivity in the rat.* Brain Res 1990, 507: 65-8.

106. Stenfors, C., Theodorsson, E., Mathe, A.A. *Effect of repeated electroconvulsive treatment on regional concentrations of tachykinins, neurotensin, vasoactive intestinal polypeptide, neuropeptide Y, and galanin in rat brain.* J Neurosc Res 1989, 24: 445-50.

107. Husum, H., Mikkelsen, J.D., Hogg, S., Mathe, A.A., Mork, A. *Involvement of hippocampal neuropeptide Y in mediating the chronic actions of lithium, electroconvulsive stimulation and citalopram.* Neuropharmacology 2000, 39: 1463-73.

108. Mathe, A.A. *Neuropeptides and electroconvulsive treatment.* J ECT 1999, 15: 60-75.

109. Mathe, A.A., Jimenez, P.A., Theodorsson, E., Stenfors, C. *Neuropeptide Y, neurokinin A and neurotensin in brain regions of Fawn Hooded "depressed", Wistar, and Sprague Dawley rats. Effects of electroconvulsive stimuli.* Prog Neuropsychopharmacol Biol Psychiatry 1998, 22: 529-46.

110. Mathe, A.A., Gruber, S., Jimenez, P.A., Theodorsson, E., Stenfors, C. *Effects of electroconvulsive stimuli and MK-801 on neuropeptide Y, neurokinin A, and calcitonin gene-related peptide in rat brain.* Neurochem Res 1997, 22: 629-36.

111. Zachrisson, O., Mathe, A.A., Stenfors, C., Lindefors, N. *Limbic effects of repeated electroconvulsive stimulation on neuropeptide Y and somatostatin mRNA expression in the rat brain.* Brain Res Mol Brain Res 1995, 31: 71-85.

112. Jimenez, P., Salmi, P., Ahlenius, S., Mathe, A.A. *Neuropeptide Y in brains of the Flinders Sensitive Line rat, a model of depression. Effects of electroconvulsive stimuli and d-amphetamine on peptide concentrations and locomotion.* Behav Brain Res 2000, 111: 115-23.

113. Rezvani, A.H., Parsian, A., Overstreet, D.H. *The Fawn-Hooded (FH/Wjd) rat: A genetic animal model of comorbid depression and alcoholism.* Psychiatr Genet 2002, 12: 1-16.

114. Cairncross, K.D. *Olfactory bulbectomy as a model of depression.* In: Animal Models in Psychopathology. Academic Press Australia 1984, 99-128.

115. Song, C., Earley, B., Leonard, B.E. *The effects of central administration of neuropeptide Y on behavior, neurotransmitter, and immune functions in the olfactory bulbectomized rat model of depression.* Brain Behav Immun 1996, 10: 1-16.

116. Holmes, P.V., Davis, R.C., Masini, C.V., Primeaux, S.D. *Effects of olfactory bulbectomy on neuropeptide gene expression in the rat olfactory/limbic system.* Neuroscience 1998, 86: 587-96.

117. Plotsky, P.M., Meaney, M.J. *Early, postnatal experience alters hypothalamic corticotropin-releasing factor (CRF) mRNA, median eminence CRF content and stress-induced release in adult rats.* Brain Res Mol Brain Res 1993, 18: 195-200.

118. Jimenez-Vasquez, P.A., Mathe, A.A., Thomas, J.D., Riley, E.P., Ehlers, C.L. *Early maternal separation alters neuropeptide Y concentrations in selected brain regions in adult rats.* Brain Res Dev Brain Res 2001, 131: 149-52.

119. Husum, H., Termeer, E., Mathe, A.A., Bolwig, T.G., Ellenbroek, B.A. *Early maternal deprivation alters hippocampal levels of neuropeptide Y and calcitonin-gene related peptide in adult rats.* Neuropharmacology 2002, 42: 798-806.

120. Stogner, K.A., Holmes, P.V. *Neuropeptide Y exerts antidepressant-like effects in the forced swim test in rats.* Eur J Pharmacol 2000, 387: R9-R10.

121. Redrobe, J.P., Dumont, Y., Fournier, A., Quirion, R. *The neuropeptide Y (NPY) Y_1 receptor subtype mediates NPY-induced antidepressant-like activity in the mouse forced swimming test.* Neuropsychopharmacology 2002, 26: 615-24.

122. Howell, O.W., Scharfman, H.E., Herzog, H., Sundstrom, L.E., Beck-Sickinger, A., Gray, W.P. *Neuropeptide Y is neuroproliferative for post-natal hippocampal precursor cells.* J Neurochem 2003, 86: 646-59.

123. Widerlöv, E., Lindström, L.H., Wahlestedt, C., Ekman, R. *Neuropeptide Y and peptide YY as possible cerebrospinal fluid markers for major depression and schizophrenia, respectively.* J Psychiatr Res 1988, 22: 69-79.

124. Widdowson, P.S., Ordway, G.A., Halaris, A.E. *Reduced neuropeptide Y concentrations in suicide brain.* J Neurochem 1992, 59: 73-80.

125. Berrettini, W.H., Doran, A.R., Kelsoe, J., Roy, A., Pickar, D. *Cerebrospinal fluid neuropeptide Y in depression and schizophrenia.* Neuropsychopharmacology 1987, 1: 81-3.

126. Ordway, G.A., Stockmeier, C.A., Meltzer, H.Y., Overholser, J.C., Jaconetta, S., Widdowson, P.S. *Neuropeptide Y in frontal cortex is not altered in major depression.* J Neurochem 1995, 65: 1646-50.

127. Ekman, R., Juhasz, P., Heilig, M., Ågren, H., Costello, C.E. *Novel neuropeptide Y processing in human cerebrospinal fluid from depressed patients.* Peptides 1996, 17: 1107-11.

128. Caberlotto, L., Hurd, Y.L. *Reduced neuropeptide Y mRNA expression in the prefrontal cortex of subjects with bipolar disorder.* Neuroreport 1999, 10: 1747-50.

129. Kallio, J., Pesonen, U., Kaipio, K. et al. *Altered intracellular processing and release of neuropeptide Y due to leucine 7 to*

proline 7 polymorphism in the signal peptide of preproneuropeptide Y in humans. FASEB J 2001, 15: 1242-4.

130. Westrin, A., Ekman, R., Traskman-Bendz, L. *Alterations of corticotropin releasing hormone (CRH) and neuropeptide Y (NPY) plasma levels in mood disorder patients with a recent suicide attempt.* Eur Neuropsychopharmacol 1999, 9: 205-11.

131. Westrin, A., Engstom, G., Ekman, R., Traskman-Bendz, L. *Correlations between plasma-neuropeptides and temperament dimensions differ between suicidal patients and healthy controls.* J Affect Disord 1998, 49: 45-54.

132. Rasmusson, A.M., Hauger, R.L., Morgan, C.A., Bremner, J.D., Charney, D.S., Southwick, S.M. *Low baseline and yohimbine-stimulated plasma neuropeptide Y (NPY) levels in combat-related PTSD.* Biol Psychiatry 2000, 47: 526-39.

133. Morgan, C.A. III, Wang, S., Southwick, S.M. et al. *Plasma neuropeptide Y concentrations in humans exposed to military survival training.* Biol Psychiatry 2000, 47: 902-9.

134. Morgan, C.A. III, Wang, S., Rasmusson, A., Hazlett, G., Anderson, G., Charney, D.S. *Relationship among plasma cortisol, catecholamines, neuropeptide Y, and human performance during exposure to uncontrollable stress.* Psychosom Med 2001, 63: 412-22.

135. Thiele, T.E., Marsh, D.J., Ste, M., Bernstein, I.L., Palmiter, R.D. *Ethanol consumption and resistance are inversely related to neuropeptide Y levels.* Nature 1998, 396: 366-9.

136. Bice, P., Foroud, T., Bo, R.H. et al. *Genomic screen for QTLs underlying alcohol consumption in the P and NP rat lines.* Mamm Genome 1998, 9: 949-55.

137. Carr, L.G., Foroud, T., Bice, P. et al. *A quantitative trait locus for alcohol consumption in selectively bred rat lines.* Alcohol Clin Exp Res 1998, 22: 884-7.

138. Ehlers, C.L., Li, T.K., Lumeng, L. et al. *Neuropeptide Y levels in ethanol-naive, alcohol-preferring, and nonpreferring rats and in Wistar rats after ethanol exposure.* Alcohol Clin Exp Res 1998, 22: 1778-82.

139. Hwang, B.H., Zhang, J.K., Ehlers, C.L., Lumeng, L., Li, T.K. *Innate differences of neuropeptide Y (NPY) in hypothalamic nuclei and central nucleus of the amygdala between selectively bred rats with high and low alcohol preference.* Alcohol Clin Exp Res 1999, 23: 1023-30.

140. Caberlotto, L., Thorsell, A., Rimondini, R., Sommer, W., Hyytia, P., Heilig, M. *Differential expression of NPY and its receptors in alcohol-preferring AA and alcohol-avoiding ANA rats.* Alcohol Clin Exp Res 2001, 25: 1564-9.

141. Karvonen, M.K., Pesonen, U., Koulu, M. et al. *Association of a leucine^{7}-to-proline^{7} polymorphism in the signal peptide of neuropeptide Y with high serum cholesterol and LDL cholesterol levels.* Nat Med 1998, 4: 1434-7.

142. Kauhanen, J., Karvonen, M.K., Pesonen, U. et al. *Neuropeptide Y polymorphism and alcohol consumption in middle-aged men.* Am J Med Genet 2000, 93: 117-21.

143. Lappalainen, J., Kranzler, H.R., Malison, R. et al. *A functional neuropeptide Y Leu7Pro polymorphism associated with alcohol dependence in a large population sample from the United States.* Arch Gen Psychiatry 2002, 59: 825-31.

144. Zhu, G., Pollak, L., Mottagui-Tabar, S. et al. *NPY leu7pro and alcohol dependence in Finnish and Swedish populations.* Alcohol Clin Exp Res 2003, 27: 19-24.

145. Slawecki, C.J., Betancourt, M., Walpole, T., Ehlers, C.L. *Increases in sucrose consumption, but not ethanol consumption, following ICV NPY administration*. Pharmacol Biochem Behav 2000, 66: 591-4.

146. Badia-Elder, N.E., Stewart, R.B., Powrozek, T.A., Roy, K.F., Murphy, J.M., Li, T.K. *Effect of neuropeptide Y (NPY) on oral ethanol intake in Wistar, alcohol-preferring (P), and -nonpreferring (NP) rats.* Alcohol Clin Exp Res 2001, 25: 386-90.

147. Badia-Elder, N.E., Stewart, R.B., Powrozek, T.A., Murphy, J.M., Li, T.K. *Effects of neuropeptide Y (NPY) on ethanol intake and anxiety in high and low alcohol drinking (HAD1/LAD1) rats.* Alcohol Clin Exp Res 2000, 24: 82A.