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Involvement of Neurohypophyseal Peptides in Drug-Mediated Adaptive Responses

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VAN REE, J. M. AND D. DE WIED. *Involvement of neurohypophyseal peptides in drug-mediated adaptive responses.* PHARMAC. BIOCHEM. BEHAV. **13**: Suppl. 1, 257–263, 1980.—Neurohypophyseal hormones and their fragments affect learning and memory processes in animals and men. Such processes have been implicated in development of tolerance to and physical dependence on as well as acquisition of self-administering behavior with psychoactive drugs, e.g. morphinomimetics and ethanol. The data reviewed indicate that these phenomena are modulated by neuropeptides related to neurohypophyseal hormones in a rather subtle and specific manner. This may be of significance for the underlying mechanism of drug dependence.

Morphine tolerance Morphine dependence Ethanol tolerance Ethanol dependence
Morphine physical dependence Ethanol physical dependence Heroin self-administration
Neurohypophyseal peptides Vasopressin Oxytocin PLG

EVIDENCE is accumulating to date that neurohypophyseal principles are concerned in adaptive processes in both experimental animals and men. The first report on this matter demonstrated that removal of the posterior pituitary in rats facilitated extinction of two way avoidance behavior [11]. This behavioral disturbance could be restored by treatment with purified lysine^8-vasopressin (LVP) [11,12]. In addition, it was observed that the deficits in learning and memory abilities due to hypophysectomy could be restored by treatment with LVP [12]. Subsequent studies in intact rats showed that a single subcutaneous injection of LVP results in a long term resistance to extinction of one way active avoidance behavior [13]. A similar effect was observed with desglycinamide^9-lysine^8-vasopressin (DG-LVP), which is practically devoid of classical endocrine activities displayed by the whole LVP molecule [16]. thus, a dissociation with respect to recognition sites mediating biological activity can be made between peripheral and central effects of vasopressin. Further experimentation in animals revealed that vasopressin promotes consolidation of acquired information and additionally plays a role in retrieval processes or in the expression of stored information [12,57]. Structure-activity relationships show that particularly the ring structure of vasopressin and oxytocin (Fig. 1) is important for consolidation processes, while the C-terminal part seems to be more concerned with retrieval processes [7,57]. The whole oxytocin molecule may act to block consolidation processes and to repress reproduction of recent information [6,7], and therefore may be regarded as an amnestic peptide. Thus, neurohypophyseal hormones may be involved in processes which enable the organism to adapt adequately to environmental changes and in addition may serve as precursor molecules for neuropeptides which selectively modulate brain mechanisms to consolidate, to retrieve and to repress

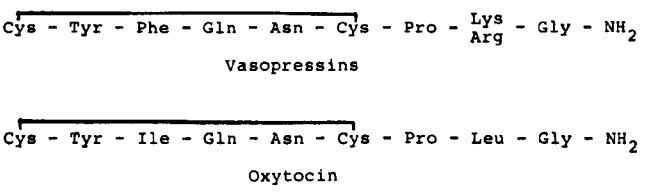

FIG. 1. Amino acid sequence of the neurohypophyseal hormones.

recently acquired information. The action of neurohypophyseal hormones and their fragments on memory processes can also be derived from studies showing that these neuropeptides affect experimentally induced amnesia [30, 42, 63].

The physiological significance of the neurohypophyseal hormones in this respect was demonstrated using three experimental models. First, the already-mentioned ablation of the posterior or the whole pituitary, resulting in behavioral disturbances which could be corrected by vasopressin treatment. Second, rats of the Brattleboro strain, which have hereditary diabetes insipidus, lack the ability to synthesize vasopressin. These rats have difficulties in acquiring and maintaining responses in active and passive avoidance behavioral procedures [4,18]. Treatment of these rats with vasopressin restored normal avoidance behavior. Third, temporary blockade of vasopressin or oxytocin activity in the brain by the intracerebroventricular injection of, respectively, vasopressin or oxytocin antiserum. Such a treatment, administered immediately after the animal had acquired the information, repressed (in the case of vasopressin antiserum) or facilitated (in the case of oxytocin antiserum) consolidation processes [5,58].

Some reports concerning the influence of vasopressin on memory processes in men are available at present. Legros and associates [33,34] showed that intranasal application of LVP produces an improvement of several aspects of attention and memory in men aged from 50 to 65 years. Case reports describe a beneficial effect of vasopressin in patients suffering from post-traumatic amnesia [38] or amnesia due to alcoholism [32], although others question these findings [3,24]. Some pilot studies suggest that vasopressin may be beneficial in psychosis and depression [21, 40, 62]. Patients with diabetes insipidus appeared to be inferior in several memory tasks, as compared to healthy individuals, and the patients with diabetes insipidus improved after vasopressin treatment [29]. Children with Lesch-Nyhan disease, who are unable to learn a passive avoidance task, improved markedly in their learning abilities after treatment with a vasopressin analog [1]. Thus, vasopressin may also be implicated in learning and memory processes of both healthy individuals and patients with disturbances in these processes.

TOLERANCE AND PHYSICAL DEPENDENCE

A decrease in drug response is characterized as tolerance when the effect of a fixed dose of the drug diminishes with repeated exposure, and when this reduced effect is overcome by increasing the dose. Thus, tolerance is due to adaptive changes in the organism, which alter the dose-response curve [2, 5, 26]. The development of tolerance is certainly not based on one single process. In fact, many adaptive changes induced by drug administration may contribute more or less to the ultimate degree of tolerance. Various concepts have been formulated in order to explain the underlying mechanisms of tolerance development [25]. One of these concerns learning and memory processes in adaptive changes during tolerance development. Two aspects of this concept have been presented. First, the display of tolerance seems to be at least partly dependent upon environmental stimuli associated with drug administration, which stresses the importance of Pavlovian conditioning principles in tolerance [45]. Second, the process of tolerance development can be regarded as cellular learning and memory processes. The cellular functions altered by drug administration adapt, as it were, to a new environment which includes the continuous presence of the drug, and this adaptation leads to a more or less correction of drug-induced changes. The cells 'remember' the experience with the drug since the response to the drug is changed by repeated exposure. Support for the 'learning' concept of tolerance is obtained from studies showing that certain manipulations can attenuate both the consolidation of conditioned behavior and the development of tolerance. These manipulations include treatment with protein synthesis inhibitors or with electroconvulsive shock, and stimulation of certain brain areas (for ref. see [49, 53, 57]).

Physical dependence can be defined as an altered state induced by repeated administration of a drug, which is recognized by a specific pattern of disturbances on withdrawal of the drug. Most of the concepts used to explain tolerance suggest that the development of physical dependence covaries with that of tolerance, although exceptions are present [8,25]. It should be emphasized thet the similarity between tolerance and physical dependence concerns almost exclusively the development of these phenomena and not the expression of these processes like withdrawal symptoms, indicating that physical dependence is present.

Neurohypophyseal Hormones and Morphine Tolerance and Physical Dependence

The involvement of neurohypophyseal hormones in learning/memory processes and the learning hypothesis of tolerance development, stimulated research on the influence of these peptides on morphine tolerance and physical dependence. Krivoy and associates [28] studied the influence of DG-LVP on the development of tolerance to the antinociceptive action of morphine in mice. It was found that this neuropeptide, administered after morphine injection, facilitated development of tolerance. Accordingly, DG-LVP injected directly into the nucleus linearis intermedius raphe accelerated the development of tolerance to morphine-induced behavioral changes in freely moving cats [10]. Two series of experiments were performed to study the physiological role of vasopressin in tolerance development. First, it was shown that in hereditary diabetes insipidus rats the development of tolerance to the antinociceptive effect of morphine was delayed. Treatment of these rats with vasopressin or DG-LVP normalized this delayed development of tolerance [15]. Second, vasopressin antiserum was injected into the cerebrospinal fluid of rats which were subjected to repeated injection of morphine. It appeared that vasopressin antiserum inhibited the development of tolerance to the analgesic action of morphine as assessed with the electric foot-shock procedure. The antiserum was effective whether it was applied after the morphine injection or prior to the injection at which time the development of tolerance was determined [59,60]. Thus, endogenous vasopressin may be physiologically involved in the development of tolerance to morphine at the level of storage as well as retrieval of information. The action of neurohypophyseal hormones and their fragments was also studied on development of physical dependence. The first report in this respect concerns the facilitatory influence of both DG-AVP (desglycinamide^{9}-arginine^{8}-vasopressin) and oxytocin on development of physical dependence on morphine [49,50]. The degree of physical dependence was measured by the body weight loss and the hypothermia induced by naloxone treatment of rats which had been repeatedly injected with morphine, and by the increased sensitivity to naloxone in antagonizing morphine antinociception following morphine pretreatment. It appeared that the process of development, rather than the expression of physical dependence was affected by these neuropeptides, since omitting peptide treatment on the test-day did not substantially influence the results. Structure-activity relationship studies revealed that the covalent ring of vasopressin and oxytocin did not influence development of physical dependence on morphine. However, the C-terminal part of these hormones was as active as the parent molecules. Both oxytocin and its C-terminal part prolyl-leucyl-glycinamide (PLG) appeared to be approximately 5 times more active than DG-AVP and prolyl-arginyl-glycinamide (PAG). The observation that particularly PLG was effective in facilitating development of physical dependence on morphine, has led to a number of investigations. The data so far are somewhat conflicting, although the different test procedures used may, at least partly, account for the observed discrepancies. In contrast to development of tolerance to and physical dependence on morphine, the ultimate degree of these phenomena induced by pellet implantation or multiple injection with morphine, was not affected by vasopressin or oxytocin treatment [44, 49, 50]. Although the effectiveness of PLG in one of the above-mentioned tests

could not be replicated by others [36], it has been confirmed that PLG facilitated development of morphine tolerance in both rats and mice and that of physical dependence in mice [9,46]. In fact, the data obtained in mice accord very well with our results in rats. The influence of PLG in this respect is not limited to morphine, since the development of tolerance to the antinociceptive action of intracerebroventricularly injected β-endorphin was facilitated by the subcutaneous administration of PLG as well [55]. Replacement of an L amino acid residue in a neuropeptide by its D-enantiomer may yield peptides with decreased or increased activity, or with effects opposite to those of the original entity [17]. Considering this principle, the dipeptide Z-Pro-D-Leu was prepared and tested on development of tolerance to and physical dependence on morphine. Indeed, it was found that this modified peptide attenuated tolerance development [64]. However, these authors, using the same test procedure, found that PLG also inhibited development of morphine tolerance and physical dependence, while oxytocin had a facilatory effect [2,65]. Thus, neurohypophyseal hormones and their fragments modulate adaptive processes concerned in the development of morphine tolerance and physical dependence. It is not clear at present whether a common neuronal mechanism mediates the action of neurohypophyseal principles on both tolerance and physical dependence. The data obtained in diabetes insipidus rats and with vasopressin antiserum suggest a physiological role for vasopressin in tolerance development, and PLG so far appears to be the most potent peptide with respect to modulation of development of physical dependence. It must be kept in mind that the ultimate degree of tolerance and physical dependence, as determined in the experimental situation, is the result of several neuronal processes. Thus, one of these processes may be under the control of vasopressin, while other processes could be preferentially affected by PLG. Accordingly, it has been suggested that the action of PLG is mediated by its suppressive effect on α-MSH release [46] but such a mode of action is very unlikely for vasopressin.

Neurohypophyseal Hormones and Ethanol Tolerance and Physical Dependence

A parallel between tolerance and learning has also been suggested in the case of ethanol. It appeared that tolerance is acquired more rapidly when animals can perform a task under the influence of ethanol as compared to animals receiving ethanol after the task. This phenomenon has been defined as behavioral augmentation of tolerance (for ref. see [31]). It was argued that association of the presence of ethanol with the behavioral task facilitated the rate of tolerance development rather than the ultimate degree of tolerance. In view of the influence of neurohypophyseal hormones on both learning and memory processes and on development of tolerance to and physical dependence on opioids, experiments were performed to test whether these hormones are also involved in ethanol tolerance and physical dependence. The first report on this matter from Hoffman and coworkers [22] showed that treatment with arginine-vasopressin (AVP) attenuated the disappearance rate of tolerance to ethanol in mice. This disappearance rate of tolerance after cessation of ethanol administration via a liquid diet, was evaluated by measuring the hypothermic response and the duration of loss of the righting reflex induced by a test dose of ethanol. The attenuating effect of AVP occurred by treatment either during and after or only after the ethanol consuming period. Oxytocin appeared to be ineffective in this respect [23]. Neither the ethanol metabolism nor the withdrawal symptomatology was effected by vasopressin or oxytocin treatment. Using a different test procedure, in which mice were continuously exposed to ethanol vapours, it was found that residual tolerance to the hypothermic effect of ethanol was enhanced by DG-AVP. This effect was observed when the peptide was continuously administered either during the ethanol exposure phase or during and after this phase, but not when the administration of DG-AVP was restricted to the period of testing for residual tolerance [43]. Similar effects were noted when withdrawal convulsions were measured. These convulsions were exacerbated in animals treated with DG-AVP throughout the period of physical dependence induction and to a minor degree when treatment was started after the exposure to ethanol. Oxytocin appeared to mimic at least some of the effects of DG-AVP in this test [41]. Thus, DG-AVP seems to facilitate development of tolerance to and physical dependence on ethanol, which agrees well with the findings on opioids. Also the decay of ethanol tolerance and physical dependence was found to be more or less attenuated by vasopressin. As in the case of morphine, the development of ethanol tolerance and physical dependence may preferentially be influenced by vasopressin. Also with ethanol, a variety of processes may be involved in the development of tolerance to and physical dependence on this drug. These processes may be differentially influenced by the same neuropeptide. This may explain the preliminary finding that DG-AVP and PLG inhibited the development of behaviorally augmented tolerance to ethanol [27].

Although more experimentation is needed to clarify some seeming discrepancies, the present data suggest that tolerance to and physical dependence on both opioids and ethanol are modulated by neurohypophyseal hormones and their fragments. Thus, adaptive changes in response to invading stimuli, resulting in a more or less adjustment to the new environment—the presence of the drug—may be modulated by these neuropeptides.

DRUG DEPENDENCE

Several psychoactive drugs, including morphinomimetics and ethanol, possess dependence-creating properties. The (self-)administration of these drugs may lead to a state of drug dependence, which is characterized by the drug user performing behavior leading specifically to further administration of the drug [26]. Severe degrees of dependence are commonly labelled as drug addiction. The dependence creating properties can be analyzed reliably in self-administration experiments in animals as well as humans. Such procedures in particular test the reinforcing capacity of the drug, which is the common denominator of the occurrence of dependence with various drugs [26,48]. Additionally, procedures using self-administration techniques in animals predict reliably about human abuse potential of a particular drug. Learning and memory processes play an important role in the initial phase of a new behavioral pattern. Thus, neuropeptides related to neurohypophyseal hormones may be involved in acquisition and maintenance of drug seeking behavior as well.

Neurohypophyseal Hormones and Heroin Self-Administration

Acquisition of intravenous heroin self-administration in

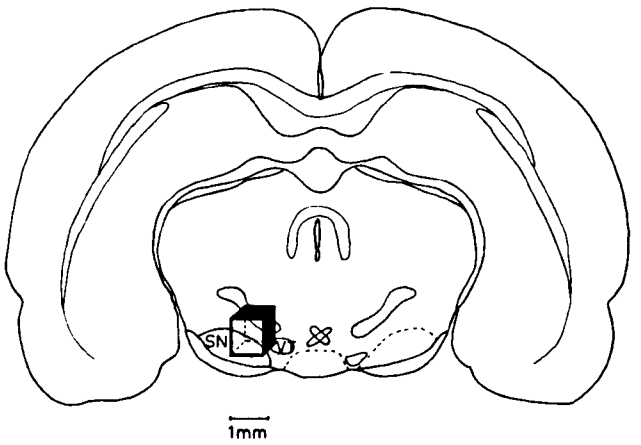

FIG. 2. Position of the cannulae for fentanyl self-administration. Only the data of rats which have the cannula in the indicated area, as verified by histological examination, were used in the analysis. VT=ventral tegmental area; SN=substantia nigra. Drawing is taken from Pellegrino and Cushman [39].

rats was attenuated by daily treatment with DG-AVP. This effect was clearly present after some days of testing and appeared to be long-lasting [52]. Vasopressin may be physiologically involved in acquisition of heroin self-administration, since intracerebroventricularly applied vasopressin antiserum enhanced the rate of acquisition [51] Structure-activity relationship studies revealed that the effect of vasopressin is located mainly in the ring structure of this hormone [52,53]. Interestingly, PLG facilitated acquisition of heroin self-administration, thus exhibiting an effect opposite to that of DG-AVP. Since the amount of drug taken can serve as an index of the reinforcing efficacy of the reinforcer, i.e. drug injection [56], it was postulated that DG-AVP attenuates and PLG enhances the reinforcing efficacy of heroin. This hypothesis is supported by data on intracranial self-stimulating (ICSS) behavior. This behavior is widely used to explore the significance of certain brain structures with respect to reward [66]. It was found that DG-AVP attenuated and that PLG enhanced ICSS elicited via electrodes implanted in the ventral tegmental-medial substantia nigra area, which contains the cell bodies of the mesolimbic and mesocortical dopaminergic pathways [19]. Thus, these peptides may influence heroin self-administration by interfering with transmission in mesolimbic dopaminergic systems, which may be consistent with the role of dopamine in reward and the reinforcing effects of opiates [67] and with the interference of these neuropeptides with dopaminergic activity in the brain [57,61]. To test this possibility an experiment was carried out with fentanyl self-administration into the ventral tegmental-medial substantia nigra area (Fig. 2). It was observed that self-administering behavior developed relatively fast, in that the ceiling level of behavior was obtained already on the 4 day of testing (Fig. 3). Peripheral treatment with DG-AVP attenuated, while treatment with PLG enhanced, fentanyl self-administration into this area (Fig. 3). These data may support the involvement of the mesolimbic dopaminergic pathway in the interaction between neuropeptides related to neurohypophyseal hormones and opiate self-administration.

The striking similarities between the effectiveness of vasopressin in memory consolidation and in heroin self-administration [14,52], might suggest that vasopressin and related neuropeptides are only effective during acquisition of heroin self-administration or when this behavior is changed in response to variations in the reinforcement or environmental cues. Thus, the effectiveness of these neuropeptides may depend on the degree of reinforcement control over behavior. This assumption may explain the findings showing that DG-AVP interacts with ICSS at low but not at high current intensities [19], and that this neuropeptide does not reduce morphine self-administration in well-trained monkeys, physically dependent on morphine and with a long history of self-administration [35]. The influence of DG-AVP was also studied in human heroin addicts in an outpatient clinic. Since the effectiveness of DG-AVP may be most pronounced in situations in which the behavior is changed in response to alterations in the reinforcement, the effect of DG-AVP on drug intake was investigated during the initial

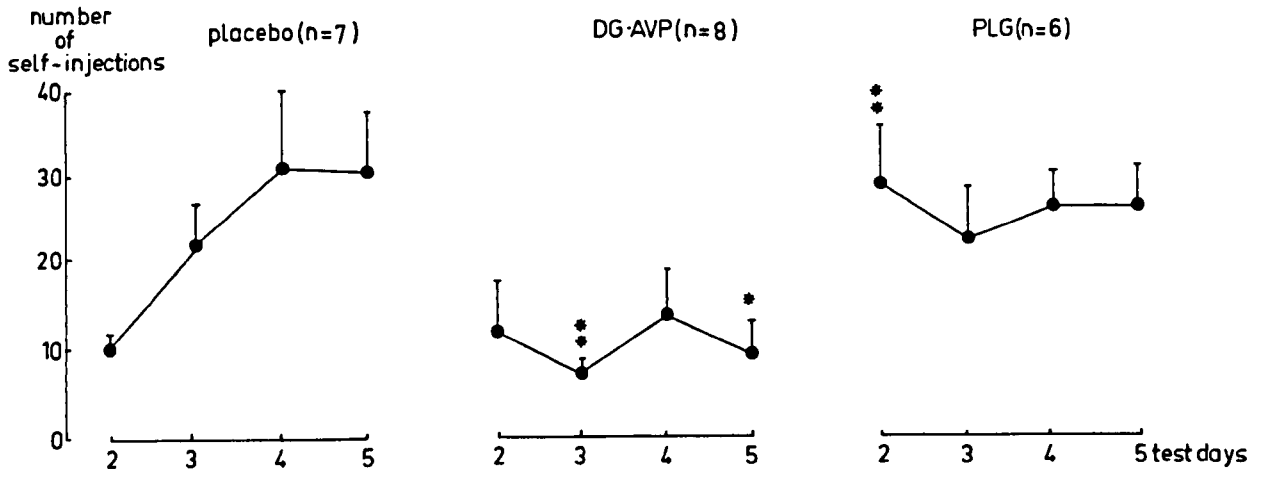

FIG. 3. Fentanyl self-administration via cannula implanted into the ventral tegmental area (see Fig. 2). Animals were allowed to self-administer fentanyl (2.5 ng/0.5 μl/injection) on a continuous reinforcement schedule by pressing a lever in a 5 day, 6 hours per day, test procedure as described in detail previously [52]. The mean (±SEM) of self-injections per day of a group of rats subcutaneously treated every day 1 hr before the experimental session with either placebo (0.5 ml saline) or 1 μg DG-AVP or 1 μg PLG are presented. n=number of animals. *different from placebo treatment (*$p<0.02$, **$p<0.01$).

phase of the methadon detoxification therapy. The outcome of this pilot experiment indicates that sublingual application of DG-AVP facilitated the methadon detoxification of heroin addicts as was inferred from the longer time course of attending the clinic and from the lower percentage of urine samples with detectable morphine in patients treated with DG-AVP as compared to those receiving placebo [47]. This decrease of heroin intake in patients treated with DG-AVP may support the postulate that this neuropeptide attenuated the reinforcing efficacy of heroin which may be critically involved in acquisition and maintenance of heroin seeking behavior.

Neurohypophyseal Hormones and Ethanol Self-Administration

Acquisition and retention of ethanol drinking was studied in rats using a procedure in which the ethanol concentration in the drinking water was varied according to the fluid consumption of each individual animal. It was reported that hypophysectomized animals rejected lower ethanol concentrations than did sham operated controls [20]. This may implicate pituitary hormones in the aversiveness of ethanol to rats. This is consistent with the findings that oral morphine and quinine intake behavior are suppressed after removal of the pituitary [54]. Detailed analysis of this effect suggested that the influence of corticosteroids on the threshold for taste may be important in this respect. The ethanol consumption of hypophysectomized rats was not substantially affected by treatment with DG-LVP [20]. However, DG-LVP treatment during the learning period enhanced the final ethanol concentration in sham operated controls, suggesting that this neuropeptide facilitated the acquisition of forced ethanol drinking behavior. The influence of DG-LVP was of a long term nature, because it was also present in the period after treatment, when the rats could choose between water and an ethanol solution. Subsequent studies showed that PLG, like DG-LVP, augmented the time-related increase in the concentration of ethanol which rats accepted in their drinking water [37]. The influence of the neuropeptides emerged after some days of the forced ethanol consumption period. The peptides appeared to be active when treatment was started from the first day of ethanol consumption, but not when DG-LVP injections were given to rats already drinking at the maximal level of acceptance concentration of ethanol.

In view of the similarities and dissimilarities between the effects of DG-LVP and PLG on ethanol consumption and the action of these neuropeptides on morphine and ethanol tolerance and physical dependence as well as heroin self-administration, it may be argued that ethanol consumption is stimulated by these peptides via facilitation of tolerance development, although other modes of actions can not be excluded as yet [37].

CONCLUDING REMARKS

The data reviewed here suggest that neuropeptides related to neurohypophyseal hormones affect drug-induced changes of homeostatic mechanisms and responses in the central nervous system. These peptides may be physiologically implicated in adaptive processes elicited by psychoactive drugs. This concerns the development of tolerance and physical dependence as well as drug seeking behavior. As yet, the available information in this respect is practically limited to morphinomimetics and ethanol, and even concerning these drugs the amount of data is too limited to draw definite conclusions as to the influence and mode of action of the neurohypophyseal hormones. However, the possible implication of these and other neuropeptide systems in drug dependence justifies detailed analysis of the activity of these systems during acquisition, maintenance and extinction of drug seeking behavior in experimental animals as well as human addicts.

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REFERENCES

1. Anderson, L. T., R. David, K. Bonnet and J. Dancis. Passive avoidance learning in Lesch-Nyhan disease: effect of l-desamino-8-arginine-vasopressin. *Life Sci.* **24**: 905–910, 1979.
2. Bhargava, H. N., R. Walter and R. F. Ritzmann. Development of narcotic tolerance and physical dependence: effects of Pro-Leu-Gly-NH_2 and cyclo (Leu-Gly). *Pharmac. Biochem. Behav.* **12**: 73–77, 1980.
3. Blake, D. R., M. J. Dodd and J. Grimley Evans. Vasopressin in amnesia. *Lancet* **1**: 608, 1978.
4. Bohus, B., Tj. B. van Wimersma Greidanus and D. de Wied. Behavioral and endocrine responses of rats with hereditary hypothalamic diabetes insipidus (Brattleboro Strain). *Physiol. Behav.* **14**: 609–615, 1975.
5. Bohus, B., I. Urban, Tj. B. van Wimersma Greidanus and D. de Wied. Opposite effects of oxytocin and vasopressin on avoidance behavior and hippocampal theta rhythm in the rat. *Neuropharmacology* **17**: 239–247, 1978.
6. Bohus, B., G. L. Kovács and D. de Wied. Oxytocin, vasopressin and memory: opposite effects on consolidation and retrieval processes. *Brain Res.* **157**: 414–417, 1978.
7. Bohus, B., G. L. Kovács, D. de Wied and H. M. Greven. Structural requirements for the opposite effects of oxytocin and vasopressin on memory consolidation. *Pharmac. Biochem. Behav.*, submitted.
8. Clouet, D. H. and K. Iwatsubo. Mechanisms of tolerance to and dependence on narcotic analgesic drugs. *A. Rev. Pharmac.* **15**: 49–71, 1975.
9. Contreras, P. C. and A. E. Takemori. The effects of prolyl-leucyl-glycinamide on morphine tolerance and dependence. *Fedn Proc.* **39**: 845, 1980.
10. Cools, A. R., C. L. E. Broekkamp, L. C. M. Gieles, A. Megens and H. J. G. M. Mortiaux. Site of action of development of partial tolerance to morphine in cats. *Psychoneuroendocrinology* **2**: 17–33, 1977.
11. de Wied, D. The influence of the posterior and intermediate lobe of the pituitary and pituitary peptides on the maintenance of a conditioned avoidance response in rats. *Int. J. Neuropharmac.* **4**: 157–167, 1965.
12. de Wied, D. Effects of peptide hormones on behavior. In: *Frontiers in Neuroendocrinology*, edited by W. F. Ganong and L. Martini. New York: Oxford University Press, 1969, pp. 97–140.
13. de Wied, D. Long term effect of vasopressin on the maintenance of a conditioned avoidance response in rats. *Nature* **232**: 58–60, 1971.
14. de Wied, D. Behavioral effects of intraventricularly administered vasopressin and vasopressin fragments. *Life Sci.* **19**: 685–690, 1976.

15. de Wied, D. and W. H. Gispen. Impaired development of tolerance to morphine analgesia in rats with hereditary diabetes insipidus. *Psychopharmacologia* 46: 27–29, 1976.
16. de Wied, D., H. M. Greven, S. Lande and A. Witter. Dissociation of the behavioral and endocrine effects of lysine vasopressin by tryptic digestion. *Br. J. Pharmac.* 45: 118–122, 1972.
17. de Wied, D., A. Witter and H. M. Greven. Behaviorally active ACTH analogues. *Biochem. Pharmac.* 24: 1463–1468, 1975.
18. de Wied, D., B. Bohus and Tj. B. van Wimersma Greidanus. Memory deficit in rats with hereditary diabetes insipidus. *Brain Res.* 85: 152–156, 1975.
19. Dorsa, D. M. and J. M. van Ree. Modulation of substantia nigra self-stimulation by neuropeptides related to neurohypophyseal hormones. *Brain Res.* 172: 367–371, 1979.
20. Finkelberg, F., H. Kalant and A. E. LeBlanc. Effect of vasopressin-like peptides on consumption of ethanol by the rat. *Pharmac. Biochem. Behav.* 9: 453–458, 1978.
21. Gold, Ph. W., H. Weingarten, J. C. Ballenger, F. K. Goodwin and R. M. Post. Effects of DDAVP on behaviour and cognition in primary affective disorder. *Lancet* II: 992–994, 1979.
22. Hoffman, P. L., R. F. Ritzmann, R. Walter and B. Tabakoff. Arginine vasopressin maintains ethanol tolerance. *Nature* 276: 614–616, 1978.
23. Hoffman, P. L., R. F. Ritzmann and B. Tabakoff. The influence of arginine vasopressin and oxytocin on ethanol dependence and tolerance. In: *Currents in Alcoholism, Vol. V*, edited by M. Galanter. New York: Grune and Stratton Inc., 1979, pp. 5–16.
24. Jenkins, J. S., H. M. Mather, A. K. Coughlan and D. G. Jenkins. Desmopressin in post-traumatic amnesia. *Lancet* II: 1245–1246, 1979.
25. Kalant, H. Behavioral criteria for tolerance and physical dependence. In: *The Bases of Addiction*, edited by J. Fishman. Berlin: Dahlem Konferenzen, 1978, pp. 199–220.
26. Kalant, H., J. A. Engel, L. Goldberg, R. R. Griffiths, J. H. Jaffe, N. A. Krasnegor, N. K. Mello, J. H. Mendelson, T. Thompson and J. M. van Ree. Behavioral aspects of addiction. In: *The Bases of Addiction*, edited by J. Fishman. Berlin: Dahlem Konferenzen, 1978, pp. 463–496.
27. Kalant, H., R. F. Mucha and R. Niesink. Effects of vasopressin and oxytocin fragments on ethanol tolerance. *Proc. Can. Fedn Biol. Soc.* 21: 71, 1978.
28. Krivoy, W. A., E. Zimmermann and S. Lande. Facilitation of development of resistance to morphine analgesia by desglycinamide^{9}-lysine vasopressin. *Proc. natn. Acad. Sci. U.S.A.* 71: 1852–1856, 1974.
29. Laczi, F., Zs. Valkusz, F. A. László, A. Wagner, T. Járdánházy, A. Szász, J. Szilárd and G. Telegdy. Effects of lysine-vasopressin and l-deamino-8-D-arginine-vasopressin on the memory in healthy individuals and diabetes insipidus patients. Submitted.
30. Lande, S., J. B. Flexner and L. B. Flexner. Effect of corticotropin and desglycinamide^{9}-lysine vasopressin on suppression of memory by puromycin. *Proc. natn. Acad. Sci. U.S.A.* 69: 558–560, 1972.
31. LeBlanc, A. E., C. X. Poulos and H. D. Cappell. Tolerance as a behavioral phenomenon: evidence from two experimental paradigms. In: *Behavioral Tolerance: Research and Treatment Implications*, edited by N. A. Krasnegor. NIDA Research Monograph 18, 1978, pp. 72–89.
32. LeBoeuf, A., J. Lodge and P. G. Eames. Vasopressin and memory in Korsakoff syndrome. *Lancet* II: 1370, 1978.
33. Legros, J. J., P. Gilot, X. Seron, J. Claessens, A. Adam, J. M. Moeglen, A. Audibert and P. Berchier. Influence of vasopressin on learning and memory. *Lancet* I: 41–42, 1978.
34. Legros, J. J. and P. Gilot. Vasopressin and memory in the human. In: *Brain Peptide: A New Endocrinology*, edited by A. M. Gotto, Jr., E. J. Peck, Jr. and A. E. Boyd, III. Amsterdam: Elsevier/North Holland Biomedical Press, 1979, pp. 347–363.
35. Mello, N. K. and J. H. Mendelson. Effects of the neuropeptide DG-AVP on morphine and food self-administration by dependent rhesus monkey. *Pharmac. Biochem. Behav.* 10: 415–419, 1979.
36. Mucha, R. F. and H. Kalant. Failure of prolyl-leucyl-glycinamide to alter analgesia measured by the Takemori test in morphine-pretreated rats. *J. Pharm. Pharmac.* 31: 572–573, 1979.
37. Mucha, R. F. and H. Kalant. Effects of desglycinamide$^{(9)}$-lysine$^{(8)}$-vasopressin and prolyl-leucyl-glycinamide on oral ethanol intake in the rat. *Pharmac. Biochem. Behav.* 10: 229–234, 1979.
38. Oliveros, J. C., M. K. Jandali, M. Timsit-Berthier, R. Remy, A. Benghezal, A. Audibert and J. M. Moeglen. Vasopressin in amnesia. *Lancet* I: 42, 1978.
39. Pellegrino, L. J. and A. J. Cushman. *A Stereotaxic Atlas of the Rat Brain*. New York: Appleton-Century-Crofts, 1967.
40. Raskind, M. A., R. E. Weitzman, H. Orenstein, D. A. Fisher and N. Courtney. Is antidiuretic hormone elevated in psychosis? A pilot study. *Biol. Psychiat.* 13: 385–390, 1977.
41. Rigter, H. and J. C. Crabbe. Neurohypophyseal peptides and ethanol. In: *Hormones and the Brain*, edited by D. de Wied and P. A. van Keep. MTP, 1980, in press.
42. Rigter, H., H. van Riezen and D. de Wied. The effects of ACTH- and vasopressin-analogues on CO_2-induced retrograde amnesia in rats. *Physiol. Behav.* 13: 381–388, 1974.
43. Rigter, H., H. Rijk and J. C. Crabbe. Tolerance to ethanol and severity of withdrawal in mice are enhanced by a vasopressin fragment. *Eur. J. Pharmac.* 64: 53–68, 1980.
44. Schmidt, W. K., J. W. Holaday, H. H. Loh and E. L. Way. Failure of vasopressin and oxytocin to antagonize acute morphine antinociception or facilitate narcotic tolerance development. *Life Sci.* 23: 151–158, 1978.
45. Siegel, S. Morphine analgesic tolerance: Its situation specificity supports a Pavlovian conditioning model. *Science* 193: 323–325, 1976.
46. Székely, J. I., E. Miglécz, Z. D. Kovács, I. Tarnawa, A. Z. Rónai, L. Gráf and S. Bajusz. Attenuation of morphine tolerance and dependence by α-melanocyte stimulating hormone (α-MSH). *Life Sci.* 24: 1931–1938, 1979.
47. van Beek-Verbeek, G., M. Fraenkel, P. J. Geerlings, J. M. van Ree and D. de Wied. Des-glycinamide-arginine-vasopressin in methadone detoxification of heroin addicts. *Lancet* II: 738–739, 1979.
48. van Ree, J. M. Reinforcing stimulus properties of drugs. *Neuropharmacology* 18: 963–969, 1979.
49. van Ree, J. M. and D. de Wied. Prolyl-leucyl-glycinamide (PLG) facilitates morphine dependence. *Life Sci.* 19: 1331–1340, 1976.
50. van Ree, J. M. and D. de Wied. Effect of neurohypophyseal hormones on morphine dependence. *Psychoneuroendocrinology* 2: 35–41, 1977.
51. van Ree, J. M. and D. de Wied. Heroin self-administration is under control of vasopressin. *Life Sci.* 21: 315–320, 1977.
52. van Ree, J. M. and D. de Wied. Modulation of heroin self-administration by neurohypophyseal principles. *Eur. J. Pharmac.* 43: 199–202, 1977.
53. van Ree, J. M. and D. de Wied. Brain peptides and psychoactive drug effects. In: *Research Advances in Alcohol and Drug Problems, Vol. VI*, edited by Y. Israel, F.B. Glaser, H. Kalant, R. E. Popham, W. Schmidt and R. G. Smart. New York: Plenum, 1980, in press.
54. van Ree, J. M. and R. J. M. Niesink. Pituitary-adrenal axis and oral morphine consumption in rats. *Pharmac. Biochem. Behav.* 9: 493–498, 1978.
55. van Ree, J. M., D. de Wied, A. F. Bradbury, E. C. Hulme, D. G. Smyth and C. R. Snell. Induction of tolerance to the analgesic action of lipotropin C-fragment. *Nature* 264: 792–794, 1976.
56. van Ree, J. M., J. L. Slangen and D. de Wied. Intravenous self-administration of drugs in rats. *J. Pharmac. exp. Ther.* 204: 547–557, 1978.
57. van Ree, J. M., B. Bohus, D. H. G. Versteeg and D. de Wied. Neurohypophyseal principles and memory processes. *Biochem. Pharmac.* 27: 1793–1800, 1978.

58. van Wimersma Greidanus, Tj. B. and D. de Wied. Modulation of passive-avoidance behavior of rats by intracerebroventricular administration of antivasopressin serum. *Behav. Biol.* **18**: 325–333, 1976.
59. van Wimersma Greidanus, Tj. B., H. Tjon Kon Fat-Bronstein and J. M. van Ree. Antisera to pituitary hormones modulate development of tolerance to morphine. In: *Characteristics and Function of Opioids*, edited by J. M. van Ree and L. Terenius. Amsterdam: Elsevier/North-Holland Biomedical Press, 1978, pp. 73–74.
60. van Wimersma Greidanus, Tj. B., J. M. van Ree and D. H. G. Versteeg. Neurohypophyseal peptides and avoidance behavior: the involvement of vasopressin and oxytocin in memory processes. In: Proceedings IBRO-Symposium *Neuropeptides and Neural Transmission*, edited by C. Ajmone Marsan and W. Z. Traczyk. New York: Raven Press, 1980, pp. 293–300.
61. Versteeg, D. H. G., M. Tanaka, E. R. de Kloet, J. M. van Ree and D. de Wied. Prolyl-leucyl-glycinamide (PLG): regional effects on α-MPT-induced catecholamine disappearance in rat brain. *Brain Res.* **143**: 561–566, 1978.
62. Vranckx, C., Ph. Minne, A. Benghezal, J. M. Moeglen and A. Audibert. Vasopressine et schizophrenie. Abstract: *11th Wld Congress Biol. Psychiat.*, Barcelona, 1978.
63. Walter, R., P. L. Hoffman, J. B. Flexner and L. B. Flexner. Neurohypophyseal hormones, analogs and fragments: Their effect on puromycin-induced amnesia. *Proc. natn. Acad. Sci. U.S.A.* **72**: 4180–4184, 1975.
64. Walter, R., R. F. Ritzmann, H. N. Bhargava, Th. C. Rainbow, L. B. Flexner and W. A. Krivoy. Inhibition by Z-Pro-D-Leu of development of tolerance to and physical dependence on morphine in mice. *Proc. natn. Acad. Sci. U.S.A.* **75**: 4573–4576, 1978.
65. Walter, R., R. F. Ritzmann, H. N. Bhargava and L. B. Flexner. Prolyl-leucyl-glycinamide, cyclo(leucylglycine), and derivatives block development of physical dependence on morphine in mice. *Proc. natn. Acad. Sci. U.S.A.* **76**: 518–520, 1979.
66. Wauquier, A. and E. T. Rolls (Eds.). *Brain-Stimulation Reward*. A collection of papers for the First International Conference on Brain-Stimulation Reward at Janssen Pharmaceutics, Beerse, Belgium on April 21-24, 1975. Amsterdam: North-Holland Publishing Company, 1976.
67. Wise, R. A. Catecholamine theories of reward: a critical review. *Brain Res.* **152**: 215–247, 1978.