Pharmacology Biochemistry & Behavior, Vol. 20, pp. 29–33, 1984. © Ankho International Inc. Printed in the U.S.A.

Feeding, Drinking, Urine Osmolality in DI Brattleboro Rats: Changes by Morphine, Naloxone, D-Amino Acids, Prolyl-leucyl-glycinamide (PLG)

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Received 13 December 1982

KOYUNCUOĞLU, H., K. BERKMAN AND H. SABUNCU. *Feeding, drinking, urine osmolality in DI Brattleboro rats: Changes by morphine, naloxone, D-amino acids, prolyl-leucyl-glycinamide (PLG).* PHARMACOL BIOCHEM BEHAV 20(1) 29–33, 1984.—Brattleboro rats placed in metabolism cages were injected with morphine (Mor), naloxone (Nal), D- and L-aspartic acid (D- and L-Asp), D-phenylalanine (D-Phe), D-leucine (D-Leu) and prolyl-leucyl-glycinamide (PLG), alone and in suitable combinations. Food and fluid intake, urine outflow, faeces weight, rectal temperature and urinary osmolality were determined at the end of seven hours period of time. Mor, Nal, D-Asp and PLG alone caused a significant decrease in food and fluid intake, urine volume and faeces weight and a significant increase in urinary osmolality being the osmolality of the Mor, D-Asp and PLG injected groups higher than 300 mOsmol/kg. The combination of Nal with Mor, D-Asp and PLG appeared to intensify the changes induced by Mor, D-Asp and PLG whereas L-Asp antagonized the majority of changes caused by Mor or PLG. The results were discussed in the light of the previous experimental findings.

DI Brattleboro rats Food intake Fluid intake Urine volume Faeces weight Rectal temperature
Urine osmolality Morphine Naloxone D- and L-Aspartic acid D-Phenylalanine D-Leucine PLG

IT has been hypothesized that opioid systems may take part in the regulation of behaviour in relation to acquisition of nutrients and in the conservation and expenditure of energy [40]. Several studies have provided some supporting experimental results showing that naloxone (Nal), an opiate antagonist, suppresses food and fluid consumptions under a variety of experimental conditions [3, 4, 10, 15, 41]. In favour of the hypothesis by Margules [40] morphine (Mor) and other opiates have been reported to increase eating and drinking behaviours [1, 17, 39], but several other studies have generally found a decrease [7, 15, 36, 42]. The results obtained from the experiments performed on Brattleboro rats, animals suffering from severe diabetes insipidus, have also shown that Mor agonists and antagonists have a suppressive effect on water intake [5, 6, 22]. On the other hand it has generally been accepted that Mor and its surrogates cause antidiuresis most probably by the release of vasopressin from hypophysis [2, 14, 25, 38, 44] as was first claimed by DeBodo [12] although they have not unanimously substantiated the mechanism of action underlying the release of vasopressin [16, 18, 21–24].

The systemic administration of D-phenylalanine (D-Phe), an inhibitor of carboxypeptidase A and D-leucine (D-Leu), an inhibitor of leucine aminopeptidase have been shown to produce analgesia. The analgesia produced by these D-amino acids has been attributed to inhibitory effects on the degradation of endogenous opioids in other words through endorphinergic system since D-Phe and D-Leu produced analgesia was naloxone reversible. Moreover, a clear cross-tolerance between the analgesic actions of D-Phe and D-Leu, and Mor has been shown [8, 12, 13]. In addition to these D-aspartic acid (D-Asp) has been found to cause a decrease in food and fluid intakes, urine outflow, rectal temperature and an increase in urine osmolality [26, 29, 35]. All the effects caused by D-Asp have been abolished by the concomitant administration of L-Asp [26, 29, 35] which can antagonize some acute effects of Mor [32] including those on the activity of L-asparaginase [33], the development of physical dependence on and tolerance to morphine [30,34], the morphine withdrawal and levallorphan-precipitated abstinence signs [31].

On account of the relationships between vasopressin release and endogenous as well as exogenous opioids [2, 6, 11, 14, 21, 22, 23, 25, 44] in general, the enormous amount of water consumption and the lack of urine concentration capacity even despite of water deprivation in particular,

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TABLE 1
THE SCHEDULE OF THE EXPERIMENTS

Groups	10 ml/kg IP injection	1 ml/kg SC injection	1 ml/kg SC injection
Control	5% glucose	0.9% saline	0.9% saline
MOR	5% glucose	0.9% saline	2.5 mg/kg MOR
NAL	5% glucose	0.9% saline	10 mg/kg NAL
MOR+NAL	5% glucose	2.5 mg/kg MOR	10 mg/kg NAL
MOR+L-Asp	200 mg/kg L-Asp	0.9% saline	2.5 mg/kg MOR
D-Asp	200 mg/kg D-Asp	0.9% saline	0.9% saline
D-Asp+NAL	200 mg/kg D-Asp	0.9% saline	10 mg/kg NAL
D-Phe	200 mg/kg D-Phe	0.9% saline	0.9% saline
D-Phe+NAL	200 mg/kg D-Phe	0.9% saline	10 mg/kg NAL
D-Leu	200 mg/kg D-Leu	0.9% saline	0.9% saline
D-Leu+NAL	200 mg/kg D-Leu	0.9% saline	10 mg/kg NAL
PLG	5% glucose	0.9% saline	3 mg/kg PLG
PLG+NAL	5% glucose	10 mg/kg NAL	3 mg/kg PLG
PLG+L-Asp	200 mg/kg L-Asp	0.9% saline	3 mg/kg PLG

MOR—Morphine; D-Asp—D-Aspartic acid; NAL—Naloxone; D-Phe—D-Phenylalanine; L-Asp—L-Aspartic acid; D-Leu—D-Leucine; PLG—Prolyl-leucyl-glycinamide (MIF).

Brattleboro rats suffering from severe hereditary diabetes insipidus were considered as suitable experimental animals for short-lasting experiments like ours where the effects of Mor, Nal, D- and L-Asp, D-Phe, D-Leu and PLG (the C-terminal tripeptide of oxytocin, prolyl-leucyl-glycinamid, MIF), which has been reported to facilitate morphine dependence [43] on food and fluid intakes, urine outflow, amount of faeces, rectal temperature and urine osmolality were aimed to investigate.

METHOD

The experiments were performed on 140 male homozygous Brattleboro rats (Central Proefdierenbedrijf, Zeist, Holland) weighing 180–200 g at the beginning of the experiments. Before and during the experiments the rats were kept in a room at 22–23°C on a 12 hours light/dark cycle (6.00 a.m.–6.00 p.m.) and they had always standard powdered food and drinking water available.

Nal was a gift from Endo Laboratories (NY, USA). Mor and other chemicals were obtained from Sandoz (Basle, Switzerland) and Sigma (St. Louis, USA), respectively. Mor, Nal and PLG solutions containing 250 mg Mor, 1000 mg Nal and 300 mg PLG in 100 ml 0.9% saline were prepared fresh daily. The concentration of the L-Asp, D-Asp, D-Phe and D-Leu solutions were 2 g%. The osmolality and pH of the latter solutions were adjusted to 290 mOsm/k and 7.4 with glucose and 1 N.NaOH, respectively. Rectal temperature was measured by means of a "Tele-thermometer" Model 46 TUC (Yellow Springs Instruments, OH, USA) by placing its lubricated probe (2 cm) into rectum. The osmolality of the solutions and urines was estimated by Knauer osmometer (Berlin, FR Germany).

One day before the treatments given in Table 1 the rats which would have been subject to the experiments were placed into metabolism cages for the acclimatization. On the following day after weighing the rats received three injections according to the schedule shown in Table 1 at 8.30, 11.00 and 13.30 o'clock respectively. The estimation of food and fluid intakes, the collection of faeces and urine were started at 9.00 o'clock. One hundred and fifty min after the third injections (at 16.00 o'clock) the experiments were stopped, and rectal temperature, body weight, food and fluid intakes, faeces weight, urine output and osmolality were determined. Food consumed, weight of faeces collected, water drunk, and volume of urine excreted during seven hours period of time were expressed as gram (g) and mililitre (ml) pre 100 gram body weight (g/100 g BW, ml/100 g BW), respectively.

The results on food and fluid intakes, urine volume, faeces weight, rectal temperature and urine osmolality were analysed by the Kruskal-Wallis' analysis of variance and subsequent between group comparisons were made with the Mann-Whitney U-test provided the analysis of variance indicated a significant effect ($p<0.05$).

RESULTS

The results of the experiments are shown in Table 2. Mor significantly reduced fluid intake, urine volume and faeces weight whereas it significantly increased rectal temperature and urine osmolality. The increase in urine osmolality was over the physiological plasma osmolality (280–290 mOsmol/kg). Nal caused a significant decrease in food and fluid intakes, urine volume and faeces weight and a significant increase in core temperature and urine osmolality. Also in the group injected with Mor and Nal together the food and fluid intakes, urine volume, faeces weight were found to be significantly lower than control. On the other hand the rectal temperature increased in the Mor group appeared to be normalized. When Mor was administered together with Nal, antagonizing and intensifying effects by Nal on rectal temperature and urine osmolality, and on food and fluid intakes respectively were found. The administration of L-Asp with Mor antagonized the decreasing effect of Mor on fluid intake and urine volume, the increasing effect of Mor on urine osmolality. No antagonistic effect was observed in the increased rectal temperature and decreased faeces weight which is considered rather as a peripheral effect of Mor in the Nor + L-Asp group.

D-Asp caused a significant decrease in food and fluid in-

TABLE 2

THE MEAN VALUES (±STANDARD ERRORS) OF THE FOOD AND FLUID INTAKES, URINE VOLUME, FAECES WEIGHT (EXPRESSED PRO 100 g BW), RECTAL TEMPERATURE AND URINE OSMOLALITY IN BRATTLEBORO RATS AFTER THE ADMINISTRATIONS OF MORPHINE (2.5 mg/kg), NALOXONE (10 mg/kg), L-ASPARTIC ACID (200 mg/kg), D-ASPARTIC ACID (200 mg/kg), D-PHENYLALANINE (200 mg/kg), D-LEUCINE (200 mg/kg) AND PLG (3 mg/kg) ACCORDING TO THE SCHEDULE SHOWN IN TABLE 1

Groups	Food Intake (g)	Fluid Intake (ml)	Urine Volume (ml)	Faeces Weight (g)	Rectal Temperature (°C)	Urine Osmolality (mOsmol/kg)
Control (10)	2.30 ± 0.26	17.17 ± 1.33	15.62 ± 1.32	1.34 ± 0.17	37.39 ± 0.07	175.50 ± 7.28
Mor (10)	2.82 ± 0.29	9.30 ± 1.28*	7.58 ± 1.17*	0.52 ± 0.11*	38.05 ± 0.21*	340.50 ± 24.27*
Nal (10)	0.10 ± 0.10*	5.92 ± 0.78*	11.80 ± 1.02*	0.80 ± 0.06*	38.03 ± 0.09*	218.00 ± 5.73*
Mor + Nal (10)	0.25 ± 0.11*†	4.06 ± 0.79*†	5.44 ± 0.75*	0.60 ± 0.13*	37.62 ± 0.19†	271.50 ± 15.67*†
Mor + L-Asp (10)	2.13 ± 0.29	18.66 ± 1.23‡	17.47 ± 1.26‡	0.44 ± 0.1*	38.01 ± 0.19*	198.00 ± 9.52*‡
D-Asp (10)	0.90 ± 0.22*	7.32 ± 1.33*	7.51 ± 0.96*	0.80 ± 0.18*	36.57 ± 0.11*	343.50 ± 19.49*
D-Asp + Nal (10)	0.12 ± 0.06*	2.46 ± 0.98*†	6.89 ± 1.14*	0.76 ± 0.14*	36.68 ± 0.14*	377.50 ± 26.32*†
D-Phe (10)	1.28 ± 0.06*	19.50 ± 1.04	17.23 ± 1.15	0.91 ± 0.09*	37.26 ± 0.11	213.00 ± 7.34*
D-Phe + Nal (10)	0.33 ± 0.06*†	7.18 ± 0.98*†	10.21 ± 0.29*†	0.60 ± 0.08*†	37.46 ± 0.14	215.00 ± 6.83*
D-Leu (10)	0.80 ± 0.09*	19.76 ± 2.26*	20.08 ± 1.45*	1.10 ± 0.08	37.81 ± 0.45	220.00 ± 11.92*
D-Leu + Nal (10)	0.36 ± 0.04*†	10.36 ± 0.98*†	13.10 ± 0.83*†	1.02 ± 0.17*	37.89 ± 0.19	224.00 ± 7.68*
PLG (10)	0.72 ± 0.09*	4.38 ± 0.52*	5.20 ± 0.72*	0.69 ± 0.08*	37.36 ± 0.12	316.00 ± 7.14*
PLG + Nal (10)	0.03 ± 0.005*†	4.26 ± 0.17*	5.76 ± 0.55*	1.42 ± 0.08†	37.99 ± 0.13*†	308.00 ± 14.10*
PLG + L-Asp (10)	0.99 ± 0.08*‡	18.82 ± 1.79‡	19.94 ± 1.91*‡	0.61 ± 0.08*‡	37.40 ± 0.65	193.50 ± 7.19*‡

The figures in brackets indicate the number of animals in each group.
*Significantly different from the control values, p at least <0.05 Mann-Whitney U-test.
†Significant difference between drug + naloxone and drug, namely Mor + Nal to Mor, D-Asp + Nal to D-Asp, D-Phe + Nal to D-Phe, D-Leu + Nal to D-Leu and PLG + Nal to PLG, p at least <0.05 Mann-Whitney U-test.
‡Significant difference between drug + L-Asp and drug, namely Mor + L-Asp to Mor and PLG + L-Asp to PLG, p at least <0.05 Mann-Whitney U-test.

takes, urine volume, faeces weight, and rectal temperature, and a significant increase in urine osmolality being higher than the physiological plasma osmolality. The administration of Nal together with D-Asp significantly intensified the decrease in fluid intake and the increase in urine osmolality found in the D-Asp group.

In the group received D-Phe the food intake and faeces weight significantly decreased, the osmolality of urine significantly increased. The administration of Nal together with D-Phe appeared to cause further decreases in food intake and faeces weight and it showed a decreasing effect on fluid intake and urine volume which were close to the control values in the D-Phe group.

D-Leu exerted a decreasing effect on food intake whereas it significantly increased fluid intake, urine volume and urine osmolality. The combination of Nal with D-Leu abolished the increase and caused further significant decrease in fluid intake and urine volume on one hand, and it significantly intensified the decrease in food intake on the other hand. Additionally this combination showed a decreasing effect on faeces weight.

PLG exhibited a significant decreasing effect on food and fluid intakes, urine volume and faeces weight, and a significant increasing effect on urine osmolality. When PLG was administered together with Nal, the food intake had a further decrease, the decrease in faeces weight significantly diminished and the rectal temperature appeared to be significantly higher than control. The concomitant administration of L-Asp with PLG in turn abolished the decreasing and increasing effects of PLG on food and fluid intakes, urine volume, and urine osmolality, respectively.

No significant change in body weight occurred during the experiments in all the groups.

DISCUSSION

Because of the relatively low hydrosolubility and high necessary quantity of amino acids used in the experiments L-Asp which has been found to antagonize the effects of

D-Asp [26, 29, 35] was not given intraperitoneally together with D-Asp to avoid any possible chemical interference with each other and unusual amount of intraperitoneal solution injection. The parenteral administration of the drugs was considered to be more suitable and advantageous than oral administration before and during the experiments in order to make sure of the quantity of the drugs which could have been different from animal to animal when administered orally even though the animals had to receive 36 ml/kg of water (containing easily metabolized substances) within seven hours which should negatively have effected some results such as fluid intake, urine volume and urine osmolality.

In addition to the fact that DI Brattleboro rats consume much more water than normal ones and the osmolality of their urine cannot reach the physiological plasma osmolality in spite of water deprivation the main reason of using DI Brattleboro rats as experimental animal in this study was to provide some more supporting findings on the mechanism(s) of the increase in the urine osmolality observed in a previous study of ours [26]. The increase in urine osmolality in the normal rats might have depended on the decrease in fluid intake. Instead if an increase in urine osmolality had been found in DI Brattleboro rats after some of the treatments used in the experiments it would have been much more meaningful and promising.

The decreasing effects of Mor and Nal on food and fluid intakes seen in the present study are quite consistent with some previous ones [5, 6, 7, 15, 24, 36, 42]. Rather small dose of Mor (2.5 mg/kg) did not inhibit food intake as its higher doses do it. However it caused a decrease in faeces weight presumably due to its peripheral constipating effect. The increase in urinary osmolality by Mor might be, to some extent, independent from the decrease in the fluid intake since there was no correlation between the decrease in fluid intake and urine volume, and no dehydration was found. Furthermore the increase in urinary osmolality found in the Nal group was not as high as that observed in the Mor group in spite of the less water consumption in the Nal group.

It has recently been shown that Mor, Nal and D-Asp suppress the in vivo and in vitro activity of angiotensin converting enzyme (ACE) which catalyzes the conversion of angiotensin I into a very strong dipsogenic angiotensin II [28]. Mor, like D-Asp [27,37] possesses an inhibitory effect on the brain L-asparaginase activity which is antagonized by L-Asp [33]. Additionally PLG which exerts very similar effects to those of Mor on food and fluid intakes, urine volume, faeces weight and urine osmolality is an inhibitor of the brain L-asparaginase [27] but not of the ACE (our unpublished observation). The effects of PLG are, to a great extent, antagonized by L-Asp just as those of Mor are (Table 2). But Nal does not show any antagonistic effect on them. In contrast Nal appears to intensify them (Table 2). Now, there appear to be two different groups of drugs inhibiting fluid intake and urine outflow, increasing urinary osmolality. Nal represents the first group which has an inhibitory effect on ACE in addition to its suppressing effects on drinking, eating, urine volume and faeces weight, and its increasing effect on urinary osmolality. But the latter does not seem to be as effective enough to elevate the osmolality over the osmolality of the normal plasma as that of Mor, D-Asp and PLG (Table 2) in spite of its much higher doses (10 mg/kg) than that of Mor (2.5 mg/kg). The second group contains Mor, D-Asp and PLG the first two of which inhibit both ACE and the brain L-asparaginase whereas the latter inhibits only the brain L-asparaginase. The durgs of the second group also cause a decrease in drinking, eating, urine volume and faeces weight, and an increase in urinary osmolality over the physiological plasma osmolality which is not usually seen in the absence of vasopressin despite water deprivation (The uneffectiveness of the deep decrease by Nal in fluid intake seen in the present experiments may also take account for it). As a result it can be said that the ACE activity through the conversion of angiotensin I into angiotensin II and the brain L-asparaginase activity via some mechanism(s) unclarified yet plays an important role in drinking, eating, urine formation, amount of faeces, thermoregulation and concentration of urine. The mechanism of action of Nal, which can, at least partly, be attributed to the inhibition of ACE, may differ from those of Mor, D-Asp and PLG which are possibly related to the inhibition of both ACE and brain L-asparaginase activities and the inhibition of the brain L-asparaginase activity, respectively. The intensifying effect of Nal on the effects of Mor, D-Asp and PLG can be considered as further supporting evidence for our point of view and it can also be interpreted as a finding showing that opioid receptors have less importance in the regulation of eating and drinking than they are believed to.

D-Phe and D-Leu, whose mechanism of action is analgesia is via the inhibition of endogenous opioid degredation, did not show similar effects to those of Mor, D-Asp and PLG. Furthermore, their decreasing and increasing effects on food and fluid intakes, urine volume, faeces weight and urinary osmolality were not antagonized by Nal. Since D-Phe and D-Leu have been postulated to act through endorphinergic system their failure in showing the parallel effects to those of Mor may support the idea that the seen effects of Mor are not related to the stimulation of opioid receptors. As it has recently been shown that both D-Phe and D-Leu cannot inhibit the activities of the ACE (our unpublished observation) and the brain L-asparaginase [27] the uneffectiveness of these two D-amino acids may be taken into consideration as other supporting evidence that the brain L-asparaginase and ACE activities have an important role in eating and drinking behaviours, and in the regulation of urinary osmolality.

The increasing effect on urine osmolality by Mor, D-Asp and PLG which inhibit the brain L-asparaginase activity suggests that these drugs may have something to do with the biosynthesis and/or release of vasopressin even in DI Brattleboro rats. The results of our previous experiments [26] performed on the normal rats may, to some extent, support this idea. However, our experiments on DI Brattleboro rats in connection with the determination of plasma vasopressin level (the manuscript in preparation) after the long-term administration of Mor, D-Asp and PLG may be lighting this point remained obscure at the moment.

REFERENCES

1. Belluzzi, J. D. and L. Stein. Do enkephalin systems mediate drive reduction? *Soc Neurosci Abstr* **8:** 405, 1978.
2. Bisset, G. W., H. S. Chowdrey and W. Feldberg. Release of vasopressin by enkephalin. *Br J Pharmacol* **62:** 370–371, 1978.
3. Brown, D. R. and S. G. Holtsman. Suppression of deprivation-induced food and water intake in rats and mice by naloxone. *Pharmacol Biochem Behav* **11:** 567–573, 1979.

4. Brown, D. R. and S. G. Holtzman. Evidence that opiate receptors mediate suppression of hypertonic saline-induced drinking in the mouse by narcotic and antagonists. *Life Sci* **26:** 1543–1550, 1980.
5. Brown, D. R. and S. G. Holtzman. Suppression of drinking by naloxone in rats homo- and heterozygous for diabetes insipidus. *Pharmacol Biochem Behav* **15:** 109–114, 1981.
6. Cantalamessa, F., G. de Caro and M. Perfumi. Water intakes inhibition and vasopressin release induced by eledoisin and Leu-enkephalin in rats of the Brattleboro and Wistar strain. *Pharmacol Res Commun* **12:** 375–378, 1980.
7. Chance, W. T. and J. A. Rosecrans. Inhibition of drinking by intrahypothalamic administration of morphine. *Nature* **270:** 167–168, 1977.
8. Cheng, R. and B. Pomeranz. The antipeptidases increase electroacupuncture analgesia and show predictable hypalgesic effects on mice with congenitally abnormal endorphin systems. In: *Endogenous and Exogenous Opiate Agonists and Antagonists*, edited by E. L. Way. New York: Pergamon Press, 1980, pp. 383–386.
9. Chevillard, C. and J. M. Saavedra. High angiotensin-converting enzyme activity in the neurohypophysis of Brattleboro rats. *Science* **216:** 646–647, 1982.
10. Cooper, S. J. Naloxone: Effects on food and water consumption in the non-deprived and deprived rat. *Psychopharmacology (Berlin)* **71:** 1–6, 1980.
11. DeBodo, R. C. The antidiuretic action of morphine and its mechanisms. *J Pharmacol Exp Ther* **82:** 74–85, 1944.
12. Ehrenpreis, S., R. C. Balagot, J. E. Comaty and S. B. Myles. Naloxone reversible analgesia in mice produced by D-phenylalanine and hydrocinnamic acid, inhibitors of carboxypeptidase A. In: *Advances in Pain Research and Therapy*, vol 3, edited by J. J. Bonica *et al.* New York: Raven Press, 1979, pp. 479–488.
13. Filibeck, U., C. Castellano and A. Oliverio. Cross-tolerance between D-amino acids and morphin in mice. *Brain Res* **212:** 227–229, 1981.
14. Firemark, H. M. and R. E. Weitzman. Effects of β-endorphin, morphine and naloxone on arginine vasopressin secretion and the electroencephalogram. *Neuroscience* **4:** 1895–1902, 1979.
15. Frenk, H. and G. H. Rogers. The suppressant effects of naloxone on food and water intake in the rat. *Behav Neural Biol* **26:** 23–40, 1979.
16. Fujimoto, J. The Kidney. In: *Narcotic Drugs Biochemical Pharmacology*, edited by D. H. Clouet. New York: Plenum Press, 1971, pp. 366–393.
17. Grandison, S. and A. Guidotti. Stimulation of food intake by muscimol and beta-endorphin. *Neuropharmacology* **16:** 533–536, 1977.
18. Handley, C. A. and A. Keller. Continous changes of renal function produced by morphine in normal dogs and in dogs with diabetes insipidus. *J Pharmacol Exp Ther* **99:** 33–37, 1950.
19. Henderson, I. W., A. McKeever and C. J. Kenyon. Captopril (SQ 14,225) depresses drinking and aldosterone in rats lacking vasopressin. *Nature* **281:** 569–570, 1979.
20. Holtzman, S. G. Suppression of appetitive behavior in the rat by naloxone: lack of effect of prior morphine dependence. *Life Sci* **24:** 219–226, 1979.
21. Huidobro, F. Antidiuretic effect of morphine in rats; tolerance and physical dependence. *Br J Pharmacol* **64:** 167–171, 1978.
22. Huidobro-Toro, J. P. Antidiuretic effect of β-endorphin and morphine in Brattleboro rats: Development of tolerance and physical dependence after chronic morphine treatment. *Br J Pharmacol* **71:** 51–56, 1980.
23. Huidobro, F. and J. P. Hidobro-Toro. Antidiuretic effect of morphine and vasopressin in morphine tolerant and non-tolerant rats, differential effect on urine composition. *Eur J Pharmacol* **59:** 55–64, 1979.
24. Huidobro-Toro, J. P., F. Huidobro and R. Croxatto. Effect of β-endorphin and D-alanine$_2$-enkephalinamide on urine production and urinary electrolytes in the rat. *Life Sci* **24:** 697–704, 1979.
25. Ishikawa, S. E. and R. W. Schrier. Evidence for a role of opioid peptides in the release of arginine vasopressin in the conscious rat. *J Clin Invest* **69:** 666–672, 1982.
26. Koyuncuoğlu, H. and K. Berkman. Effect of D- and/or L-aspartic acids on feeding, drinking, urine outflow and core temperature. *Pharmacol Biochem Behav* **17:** 1265–1269, 1982.
27. Koyuncuoğlu, H. and K. Berkman. Effects of morphine, D-aspartic acid, D-phenylalanine, D-leucine and PLG on L-asparaginase activity in rats. *Med Bull Ist Med Fac*, in press.
28. Koyuncuoğlu, H., I. Hatipoğlu and N. Enginar. The effect of morphine, naloxone, D- and L-aspartic acid on the brain and lung ACE in mice. *Pharmac Res Commun*, submitted.
29. Koyuncuoğlu, H., K. Berkman, J. Wildmann and H. Matthaei. Antagonistic effect of L-aspartic acid on decrease in body weight, food and fluid intake, and naloxone reversible rectal temperature caused by D-aspartic acid. *Pol J Pharmacol Pharm*, in press.
30. Koyuncuoğlu, H., E. Genç, M. Güngör, L. Eroğlu and H. Sağduyu. The antagonising effect of aspartic acid on the brain levels of monoamines and free amino acids during the development of tolerance to and physical dependence on morphine. *Psychopharmacology (Berlin)* **54:** 187–191, 1977.
31. Koyuncuoğlu, H., M. Güngör, L. Eroğlu and H. Sağduyu. The antagonizing effect of aspartic acid on morphine withdrawal and levallorphan-precipitated abstinence syndrome signs and on associated changes in brain levels of free amino acids in the rat. *Psychopharmacology (Berlin)* **62:** 89–95, 1979.
32. Koyuncuoğlu, H., M. Güngör, H. Sağduyu and L. Eroğlu. The antagonistic effects of aspartic acid on some effects of morphine on rats. *Eur J Pharmacol* **27:** 148–150, 1974.
33. Koyuncuoğlu, H., M. Kayer-Uysal, K. Berkman, M. Güngör and E. Genç. The relationship between morphine, aspartic acid and L-asparaginase in rats. *Eur J Pharmacol* **60:** 369–372, 1979.
34. Koyuncuoğlu, H., H. Sağduyu, M. Güngör, L. Eroğlu and E. Genç. Antagonising effect of aspartic acid on the development of physical dependence on and tolerance to morphine in the rat. *Drug Res* **27:** 1676–1679, 1977.
35. Koyuncuoğlu, H., J. Wildmann, K. Berkman and H. Matthaei. The effects of D- and/or L-aspartic acids on the total weight of body, the weights of certain organs, and their protein, triglyceride and glycogen contents. *Drug Res* **32:** 738–741, 1982.
36. Kumar, R., E. Mitchell and I. P. Stolerman. Distrubed patterns of behavior in morphine tolerant and abstinent rats. *Br J Pharmacol* **42:** 473–484, 1971.
37. Lerman, M. I. and I. V. Verevkina. The inhibition of asparaginase from the blood serum of guinea pigs. *Biokhimiya* **27:** 526–531, 1962.
38. Lightman, S., N. Langdon, K. Todd and M. Forsling. Naloxone increases the nicotine-stimulated rise of vasopressin secretion in man. *Clin Endocrinol (Oxf)* **16:** 353–358, 1982.
39. Maickel, R. P., M. C. Braude and J. E. Zabik. The effects of various narcotic agonists and antagonists on deprivation-induced fluid consumption. *Neuropharmacology* **16:** 863–866, 1977.
40. Margules, D. L. Beta-endorphin and endoloxone. Hormones of the autonomic nervous system for the conservation or expenditure of bodily resources and energy in anticipation of famine or feast. *Neurosci Biobehav Rev* **3:** 155–162, 1979.
41. Margules, D. L., B. Moisset, M. J. Lewis, H. Shibuya and C. B. Pert. Beta-endorphin is associated with overeating in genetically obese mice (ob/ob) and rats (fa/fa). *Science* **202:** 988–991, 1978.
42. Mark-Kaufman, R. and R. B. Kanarek. Morphine selectively influences macronutrient intake in the rat. *Pharmacol Biochem Behav* **12:** 427–430, 1980.
43. van Ree, J. M. and D. de Wied. Prolyl-leucyl-glycinamide (PLG) facilitates morphine dependence. *Life Sci* **19:** 1331–1340, 1976.
44. Weitzman, R. E., D. A. Fisher, S. Minick, N. Ling and R. Guillemin. β-endorphin stimulates secretion of arginine vasopressin in vivo. *Endokrinologie* **101:** 1643–1646, 1977.