Pharmacology Biochemistry and Behavior, Vol. 62, No. 4, pp. 601–605, 1999

0091-3057/99/$–see front matter

PII S0091-3057(98)00179-8

Diet-Induced Enhancement of Naloxone Sensitivity Is Independent of Changes in Body Weight

SAIMA SHABIR AND TIM C. KIRKHAM

Department of Psychology, University of Reading, Whiteknights, P.O. Box 238, Reading RG6 6AL, UK

Received 28 December 1997; Revised 1 September 1998; Accepted 1 September 1998

SHABIR, S. AND T. C. KIRKHAM. *Diet-induced enhancement of naloxone sensitivity is independent of changes in body weight.* PHARMACOL BIOCHEM BEHAV **62**(4) 601–605, 1999.—Intake of palatable solutions can enhance the anorectic potency of opioid antagonists. This experiment examined the relative contributions of orosensory experience and body weight gain to the enhanced anorectic potency of naloxone (0.125, 0.25, 0.5, and 1.0 mg/kg IP). Four groups of male hooded Lister rats (Charles River) were maintained on separate feeding regimes for 3 months. S-ADLIB rats were nondeprived with free access to lab chow and 20% (w/v) sucrose solution. S-RESTRICT rats received limited sucrose (50 ml/day) and chow (15 g/day) access, yoking their body weights to ADLIB rats receiving free access to lab chow only. RESTRICT rats received approx.15 g of chow/day to maintain their body weights at 90% of the ADLIB rats. Fifteen-minute sucrose intake tests revealed marked differences between naloxone sensitivity of chronic sucrose drinkers and sucrose-naive groups. Intakes of S-ADLIB and S-RESTRICT were suppressed at all doses (max suppression >60%). In comparison to animals given sucrose, ADLIB and RESTRICT animals were significantly less sensitive (maximum suppression = 35%). Naloxone potency was independent of body weight differences. The data demonstrate that overconsumption of palatable ingesta, and not diet-induced weight gain, is sufficient to enhance antagonist potency. The study confirms that orosensory stimulation can induce plasticity in opioid systems, supporting an important role for opioids in intake regulation and general reward processes. © 1999 Elsevier Science Inc.

Opioid Appetite Obesity Anorexia Reward Eating Sucrose Palatability

OPIOID peptides may play an important role in the regulation of eating, and particularly those processes mediating the hedonic evaluation of food (orosensory reward) (2,16,19). For example, opioid receptor agonists or antagonists respectively increase or decrease intake (7,13,14); effects that are most marked with palatable foods (18,23). In people, opioid antagonists attenuate the reported pleasantness of normally preferred, palatable foods (3).

More direct support for the opioid–palatability hypothesis comes from the finding that ingestion of palatable foods can stimulate opioid release in brain tissues (4,25). Additionally, in both humans and rats, opioid antagonists exert a substantially greater suppressive effect on the ingestion of palatable foods than on less preferred, lower palatability foods (18,22).

Elevation of opioid activity in response to overconsumption of palatable foods may induce significant neural plasticity, evident experimentally as an increase in the anorectic potency of opioid antagonists. For example, avid sucrose consumption by sham-feeding rats induces naloxone supersensitivity after only 2–3 h: an effect that is clearly linked to the palatability (concentration) of sucrose solutions consumed (13,15). Such behavioral changes may reflect changes in opioid receptor density or affinity in response to elevated brain opioid levels (15,21).

In most studies of these phenomena in laboratory species, the supplement of standard laboratory diets with palatable foods and fluids generally also results in significant body weight gain. Indeed, one of the earliest studies of opioids and ingestive behavior found that rats made obese through overconsumption of palatable, calorically dense foods were also more sensitive to the anorectic effects of opioid receptor antagonists (20). We believe that these changes in sensitivity result

Requests for reprints should be addressed to Dr. T. C. Kirkham, University of Reading, Department of Psychology, P.O. Box 238, Whiteknights, Reading, UK RG6 6AL.

directly from the orosensory reward derived from food stimuli, a proposition supported by a recent study by Kanarek et al. (9). However, genetically obese rats, fed more monotonous diets, also display enhanced antagonist sensitivity (22). It is possible, therefore, that altered antagonist potency may be secondary to the weight gain associated with palatable food intake, and is not solely due to orosensory experience.

The present study was designed to investigate the relative contributions of body weight gain and orosensory stimulation in the development of diet-related changes in naloxone potency. Rats were given 3 months ad lib access to a highly palatable sucrose solution, in addition to free access to their normal maintenance diet. The potency of naloxone in suppressing intake in these animals was then compared against rats given access to sucrose, but whose body weights were matched by caloric restriction to a control group given only access to chow.

Food deprivation has been shown to attenuate antagonist anorectic potency (12). Therefore, drug effects were also assessed in rats that received a restricted daily allowance of chow alone, to determine the extent of any interaction between food availability and changes in naloxone potency that could confound our interpretation of the sucrose effects. To further aid our analysis, sucrose solution rather than chow was used as the test food in acute intake tests, because sweet solutions are generally more susceptible to antagonist anorexia. Finally, because naloxone effects are typically apparent within a few minutes of the initiation of ingestion (14), and prolonged tests may mask those short-term effects, intake tests were limited to 15 min. Through this combination of dietary regimen and brief sucrose intake tests, we hoped to optimize our ability to detect alterations in opioid antagonist sensitivity.

METHOD

Animals

Twenty-four naive, male, Hooded Lister rats (Charles River), weighing between 420–540 g at the start of drug testing, were individually housed in stainless steel cages. Animals had ad lib access to water. Rats were maintained on a reversed 12–12-h dark–light cycle (lights off at 1030 h), with the room temperature kept at 20–23°C. Rats were habituated to handling and injection procedures before drug testing.

Drugs

Naloxone hydrochloride (Sigma Chemicals, UK), was dissolved in sterile isotonic saline, injected intraperitoneally in a volume of 1 ml/kg body weight.

Procedure

Diet regimes. Rats were randomly allocated to four groups ($n = 6$ per group) and were adapted to one of four feeding regimes. Group 1 (S-ADLIB) consisted of nondeprived rats with free access to lab chow (PC Modified Diet, Special Diet Services, Essex, UK) and 20% (w/v) sucrose solution. In group 2 (ADLIB), rats had free access to lab chow only. Rats in group 3 (S-RESTRICT) received limited access (50 ml/day) to sucrose solution and chow access was chronically restricted to 15 g/day, to match their body weights to ADLIB weights while providing adequate nutrition. Group 4 (RESTRICT) received only sufficient food (approx. 15 g/day) to maintain their body weights at 90% of the ADLIB rats (which pilot tests indicated would produce a similar weight discrepancy as between ADLIB and S-ADLIB rats given the sucrose supplement). Animals in all groups had free access to drinking water.

The maintenance diet (PCD Mod. C; Special Diet Services, Witham, UK) was presented in food hoppers attached to each cage. The water and sucrose solutions were presented in graduated cylinders, with stainless steel drinking spouts. Intake of food pellets, water, and sucrose solution were measured daily. All groups were maintained on these schedules for a period of 3 months, and stable baselines for both daily energy intakes and body weight had been established when drug testing commenced.

Prior to acute drug testing, ADLIB and RESTRICT rats that had not previously received sucrose were given overnight access to 20% sucrose for familiarization. Subsequently, these animals were given daily 15-min sucrose solution intake tests until stable test intakes were obtained (4 days).

Intake tests. All testing was conducted at the beginning of the dark phase (1030–1230 h), under red light. Thirty minutes before dark onset (1000 h) food, and for S-ADLIB and S-RESTRICT rats, sucrose solution was removed from the cages and all rats were weighed. At 1100 h rats were injected with either vehicle or naloxone. Intake tests with 20% sucrose solution began 30 min later. Sucrose intake was measured gravimetrically. Naloxone doses (0.125, 0.25, 0.5, and 1.0 mg/kg) were administered according to a counterbalanced design, with at least 48 h between successive naloxone injections. Each drug day was preceded by one or more control days, when saline was administered to ensure that predrug baseline intake levels were maintained. Control intake values were calculated by averaging individual intakes after saline injection on predrug days. Food, water, and where appropriate, sucrose solutions, were restored 15 min after the end of each intake test.

All procedures were performed in compliance with the requirements of the UK Animals Scientific Procedures Act 1989.

Statistical Analyses

Sucrose solution intake and percent suppression data were analyzed by two-way analyses of variance, with diet/food availability and dose as main factors. The significance of differences between specific treatment means were assessed using the Newman–Keuls test for multiple comparisons. All tests were run using Statistica™.

RESULTS

The separate dietary regimes succeeded in producing the planned separation of body weights between groups. Thus, S-ADLIB rats with free access to chow and sucrose solution were significantly heavier than all other groups over the experimental period, $F(3, 20) = 8.242$, $p < 0.001$. Restricting access to both chow and sucrose in S-RESTRICT rats restrained their body weights to approximately 90% of S-ADLIB weights ($p < 0.05$); matching (and not significantly different from) ADLIB rats that received chow only. Finally, the body weights of RESTRICT animals were stabilized at approximately 90% of ADLIB rats. Average weights over the period of drug testing were: 544.2 ± 15.5 g (S-ADLIB), 465.8 ± 13.9 g (ADLIB), 490.8 ± 18.9 g (S-RESTRICT), 431.6 ± 21.6 g (RESTRICT).

During the predrug sucrose habituation phase, average daily caloric intakes (including 15-min sucrose test intakes) were: 119.6 ± 2.8 kcal (S-ADLIB), 107.8 ± 3.5 kcal (ADLIB), 103.5 ± 3.7 kcal (S-RESTRICT), 100.6 ± 1.8 kcal (RESTRICT); $F(3, 20) = 15.674$, $p < 0.001$. Caloric intake for ADLIB was significantly greater than for RESTRICT ($p < 0.001$), and caloric intake of S-ADLIB rats was greater than for S-RESTRICT rats ($p < 0.001$). Similar energy intakes

were maintained over the rest of the study, although differences between ADLIB and RESTRICT groups and between S-ADLIB and S-RESTRICT groups were no longer significant, $F(3, 20) = 2.628$, $p > 0.05$.

The different dietary experience and body weight levels had no marked effect on the general acceptance of sucrose during the intake tests under control conditions. Baseline intakes (Fig. 1) were closely matched, although ADLIB rats tended to consume slightly (but not significantly) less than the other groups.

Analysis of variance revealed a highly significant, dose-dependent reduction of intake after naloxone, $F(4, 80) = 32.211$, $p < 0.0001$. As may be seen in Fig. 1, the higher doses of naloxone produced significant intake suppression in all groups. However, diet regime exerted a substantial influence on sensitivity to the drug, $F(3, 20) = 3.8$, $p < 0.05$, with the chronic sucrose-drinking groups (S-ADLIB and S-RESTRICT) being most susceptible. In particular, the test intake of S-ADLIB rats was reliably reduced by all doses of naloxone, and intake of S-RESTRICT rats were reliably suppressed by all but the lowest dose. Intakes of ADLIB and RESTRICT animals were significantly reduced only at the highest doses.

The influence of pretest dietary experience on naloxone potency is more clearly seen when data are considered in terms of the percentage suppression (Fig. 2). The difference in sensitivity between chronic sucrose drinkers and sucrose-naive groups is very striking, $F(3, 20) = 19.925$, $p < 0.0001$. Intake by S-ADLIB and S-RESTRICT rats was suppressed to a similar extent. The ADLIB and RESTRICT animals displayed almost identical sensitivity to naloxone, but were substantially less affected than were the chronic sucrose drinkers. Additionally, S-ADLIB and S-RESTRICT groups were sensitive to a far wider range of naloxone doses, $F(3, 60) = 16.095$, $p < 0.0001$. In S-ADLIB animals, intake was suppressed by approximately 50% by even the lowest, 0.25 mg/kg, dose of naloxone ($p < 0.01$); while maximum suppression after 1 mg/kg was >60%. By contrast, in ADLIB and RESTRICT groups, maximum suppression was only of the order of 35%. With the single exception of 0.125 mg/kg in S-RESTRICT rats, naloxone-induced suppression in chronic sucrose drinkers was significantly greater than in the reference ADLIB group across the dose range.

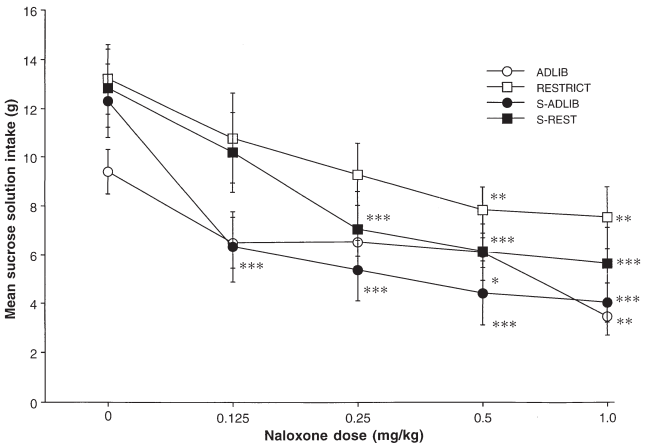

FIG. 1. Fifteen-minute intake of 20% sucrose solution for the four diet groups, following IP vehicle or naloxone injection. All values are the mean (±SEM) of six rats. Asterisks indicate significant difference from vehicle intake for each group: *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$. ADLIB: nonrestricted, chow-fed animals; S-ADLIB: nonrestricted, chow-fed animals with free access to sucrose solution during the pretest phase; RESTRICT: chow-fed rats maintained at 90% of ADLIB body weights; S-RESTRICT: rats given limited access to sucrose and chow and maintained at 90% of S-ADLIB weights.

DISCUSSION

In this experiment chronic consumption of sucrose has clearly been demonstrated to enhance animals' susceptibility to the intake suppressing actions of naloxone. This demonstration was considerably assisted by assessing naloxone's effects on sweet solutions, which are consistently reported to be more sensitive to the anorectic actions of opioid antagonists than are bland laboratory diets. The high palatability of sucrose in this study also allowed us to compare naloxone's effects against similar baseline intakes across the different treatment groups.

The most striking result of this experiment, however, is the differentiation between the sensitivity of the separate groups to naloxone on the basis of their orosensory experience, rather than their respective body weights. Previous studies have shown that the anorectic potency of opioid antagonists can be enhanced by the consumption of many different foods, sharing the common property of high palatability (2). However, in the majority of those reports dietary manipulation was accompanied by elevated body weight relative to controls. By directly addressing the issue of body weight in relation to diet-induced alteration of opioid antagonist sensitivity, we hoped to substantiate the general hypothesis that consumption of palatable foods and fluids may induce significant modification of central opioid systems. In particular, the use of measured access to sucrose and chow (as in the S-RESTRICT group) allowed us to yoke weight gain to that of rats consuming chow alone and so separate experiential factors from the influence of body weight differences.

We believe that the present findings reliably demonstrate that it is overconsumption of palatable ingesta, per se, and not their postabsorptive consequences for adiposity, which represent the key factor in inducing these modifications. Thus, it is

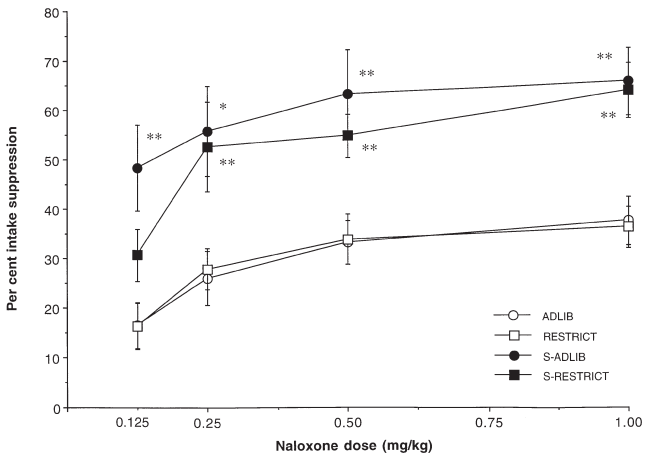

FIG. 2. Percent suppression of sucrose solution intake for each diet group, following vehicle or naloxone injection (see Fig. 1 for explanation of symbols). All values are the mean (±SEM) of six rats. Asterisks indicate significant difference from suppression obtained in ADLIB rats (nonrestricted, chow-fed animals) at each dose: *$p < 0.05$; **$p < 0.01$.

crucial that in this experiment animals with similar body weights but different orosensory experience (S-RESTRICT and ADLIB) showed markedly disparate responses to the anorectic actions of naloxone. In strong contrast, animals with significantly different body weights (S-ADLIB and S-RESTRICT), but similar ingestive experience of sucrose displayed almost identical sensitivity to the drug.

The mechanisms underlying the altered sensitivity to naloxone following overconsumption of sucrose remain to be determined. However, a number of experiments indicate that they are a consequence of elevated opioid release induced by consumption of palatable foods. In an early study with rats given palatable food supplements, Dum et al. reported changes in brain opioid receptor binding that were consistent with increased hypothalamic β-endorphin release (4). More recently, Welch et al. (25) found that rats given ad lib access to a high fat, sucrose-containing diet displayed elevated hypothalamic prodynorphin mRNA and dynorphin A_{1-17} levels. However, the authors could not clearly distinguish the consequences of orosensory stimulation from those of increased caloric intake and body weight. Thus, animals fed the sweet-fat diet also consumed 20% more calories and gained almost 50% more weight than control rats. When caloric intake was yoked to that of controls, both the increase in body weight and the elevation of prodynorphin mRNA and dynorphin A_{1-17} were prevented. In a related study, Kim et al. (10) observed that body weight reduction induced by chronic food deprivation or limited daily food access resulted in reduced levels of arcuate nucleus opioid mRNA. These studies, therefore, indicate a close relationship between ingestion and opioid activity, but do not definitively separate the relative influence of eating per se from that of changes in body weight.

A greater influence of orosensory stimulation, rather than energy intake or level of adiposity, in the activation of opioid systems is supported by experiments using animals sham feeding sucrose solutions. In the sham-feeding paradigm, sucrose solutions are recovered from the stomach within seconds of ingestion by means of an open gastric fistula. This procedure minimizes the influence of normal satiating mechanisms and allows intake rate to be precisely controlled by the palatability (concentration) of the test solution. We have previously reported that sham feeding of very palatable 30% sucrose solutions can render animals supersensitive to the suppressive effects of naloxone. As this increased sensitivity occurs in the absence of intestinal absorption of the sucrose, it is presumed that the sensory stimulation or experience of sucrose reward are sufficient to induce changes in brain opioid systems.

The sham feeding-induced naloxone supersensitivity may be related to opioid receptor adaptation in response to the release of endogenous opioids. Possibly, downregulation of receptors may occur as a homeostatic, regulatory response to elevated opioid levels. Indirect support for this hypothesis comes from the finding that chronic opioid antagonist administration, which has been demonstrated to cause opioid receptor upregulation, can prevent the development of supersensitivity to the acute anorectic effects of naloxone. Analysis of brain opioid peptide and receptor changes in relation to sucrose sham feeding are necessary to confirm these hypotheses. However, the ability of kappa receptor antagonists (in addition to the nonspecific antagonist, naloxone) to attenuate sucrose sham feeding (17) clearly complements the changes in dynorphin synthesis and release discussed above, and support a key role for this family of opioids in food reward.

The issue, referred to earlier, of greater antagonist sensitivity of genetically obese animals remains unresolved (20,22). Genetically obese animals have been reported to have elevated brain opioid levels (11). It is notable, however, that obesity in these animals is also associated with hyperphagia, so that the critical factor in their response to opioid antagonists may be related to hyperphagia and increased orosensory stimulation rather than their increased body weight. In lesion- and diet-induced obesity models, elevated opioid levels are detected only when hyperphagia is expressed, but not if overconsumption is prevented by limiting food access (6).

In discussing models of obesity, it may be relevant to consider recent developments regarding the ob gene-derived hormone leptin. Leptin is produced in adipocytes and, as its levels are directly related to the degree of adiposity, has been proposed as a crucial signal for the regulation of appetite in response to changing body weight (1,8). Thus, obese ob/ob mice that cannot synthesize leptin display hyperphagia and have decreased energy expenditure. Administration of leptin to these animals reduces food intake, increases energy expenditure and restores more normal body weight (5).

Very little information exists relating opioid activity to variation in circulating levels of leptin. It has been reported that plasma leptin concentrations are unaffected by acute naloxone administration, although Wand and Schumann have proposed that plasma leptin levels may influence hypothalamic opioidergic tone (24). It is, therefore, possible that increased adiposity and the associated elevation of leptin output may contribute to the enhanced antagonist sensitivity displayed by dietary obese animals. However, leptin-induced changes in opioid function cannot account for the increased anorectic potency of opioid antagonists in ob/ob mice, given their inability to produce the hormone. Again, autoradiographic, immunochemical, and in situ hybridization assays are required to measure changes in opioid receptor density or opioid peptide synthesis and release, to determine whether genetically obese animals possess opioid systems that are modified in similar ways to those produced here by orosensory experience alone, and whether such modifications precede or are dependent on changes in body weight.

Overall, the present findings extend a growing body of evidence indicating that consumption of palatable foods activates central opioid systems and strengthen support for an important role for those systems in food reward. Further, chronic overconsumption of palatable ingesta renders animals more sensitive to the intake suppressing action of opioid receptor antagonists, suggesting induction of significant changes in brain opioid systems. The pattern of results reported here strongly suggest that these changes occur independently of alterations in body weight. The concept of neural plasticity induced by simple orosensory stimulation is exciting, and deserving of greater efforts to unravel the precise mechanisms involved and their significance to intake regulation and general reward processes.

ACKNOWLEDGEMENTS

This study was supported by the University of Reading Research Endowment Trust Fund.

REFERENCES

1. Campfield, L. A.: Metabolic and hormonal controls of food intake: Highlights of the last 25 years—1972–1997. Appetite 29:135–152; 1997.
2. Cooper, S. J.; Kirkham, T. C.: Opioid mechanisms in the control of food consumption and taste preferences. In: Herz, A.; Akil, H.; Simon, E. J., eds. Handbook of experimental pharmacology, vol. 104, part II. Berlin: Springer Verlag; 1993:239–263.
3. Drewnowski, A.; Krahn, D.; Demitrack, M. A.; Nairn, K.: Naloxone, an opiate blocker, reduces the consumption of sweet high-fat foods in obese and lean females binge eaters. Am. J. Clin. Nutr. 61:1206–1212; 1995.
4. Dum, J.; Gramsch, C.; Herz, A.: Activation of hypothalamic β-endorphin pools by reward induced by highly palatable food. Pharmacol. Biochem. Behav. 18:443–447; 1984.
5. Frederich, R. B.; Lollmann, B.; Hamann, A.; Napolitano-Rosen, A.; Kahn, B. B.; Lowell, B.; Flier, J. S.: Expression of ob mRNA and its encoded protein in rodents. Impact of nutrition and obesity. J. Clin. Invest. 96:1658–1663; 1995.
6. Gunion, M. W.; Peters, R. H.: Pituitary beta-endorphin, naloxone and feeding in several experimental obesities. Am. J. Physiol. 241:R173–R184; 1981.
7. Jackson, A.; Cooper, S. J.: Observational analysis of the effect of the selective kappa opioid agonist, U50 488H, on feeding and related behaviors in the rat. Psychopharmacology (Berlin) 90:217–222; 1986.
8. Joannic, J.-L.; Oppert, J.-M.; Lahlou, N.; Basdevant, A.; Aubiron, S.; Raison, J.; Bornet, F.; Guy-Grand, B.: Plasma leptin and hunger ratings in healthy humans. Appetite 30:129–138; 1998.
9. Kanarek, R. B.; Foulds Mathes, W.; Hiesler, L. K.; Lima, R. P.; Monfared, L. S.: Prior exposure to palatable solutions enhances the effects of naltrexone on food intake in rats. Pharmacol. Biochem. Behav. 57:377–381; 1997.
10. Kim, E.-M.; Welch, C. C.; Grace, M. K.; Billington, C. J.; Levine, A. S.: Chronic food restriction and acute food deprivation decrease mRNA levels of opioid peptides in arcuate nucleus. Am. J. Physiol. 270:R1019–R1024; 1996.
11. Khawaja, X. Z.; Chattopadhyay, A. K.; Green, I. C.: Increased β-endorphin and dynorphin concentrations in discrete hypothalamic regions of genetically obese (ob/ob) mice. Brain Res. 555:164–168; 1991.
12. Kirkham, T. C.; Blundell, J. E.: Effect of naloxone and naltrexone on the development of satiation measured in the runway: Comparisons with d-amphetamine and d-fenfluramine. Pharmacol. Biochem. Behav. 25:12–128; 1986.
13. Kirkham, T. C.; Cooper, S. J.: Attenuation of sham feeding by naloxone is stereospecific: Evidence for opioid mediation of orosensory reward. Physiol. Behav. 43:845–847; 1988.
14. Kirkham, T. C.; Cooper, S. J.: Naloxone attenuation of sham feeding is modified by manipulation of sucrose concentration. Physiol. Behav. 44:491–494; 1988.
15. Kirkham, T. C.: Enhanced anorectic potency of naloxone in rats sham feeding 30% sucrose: Reversal by repeated naloxone administration. Physiol. Behav. 47:419–426; 1990.
16. Kirkham, T. C.; Cooper, S. J.: Opioid peptides in relation to the treatment of obesity and bulimia. In: Bloom, S. R.; Burnstock, G., eds. Peptides: A target for new drug development. London: IBC; 1991:28–44.
17. Leventhal, L.; Kirkham, T. C.; Cole, J. L.; Bodnar, R. J.: Selective actions of central mu and kappa opioid antagonists upon sucrose intake in sham-feeding rats. Brain Res. 685:205–210; 1995.
18. Levine, A. S.; Weldon, D. T.; Grace, M.; Cleary, J. P.; Billington, J. P.: Naloxone blocks that portion of feeding driven by sweet taste in food-restricted rats. Am. J. Physiol. 268:R248–R252; 1995.
19. Lynch, C. L.; Burns, G.: Opioid effects on intake of sweet solutions depend both on prior drug experience and on prior ingestive experience. Appetite 15:23–32; 1990.
20. Mandenoff, A.; Fumeron, F.; Apfelbaum, M.; Margules, D. C.: Endogenous opiates and energy balance. Science 215:1536–1538; 1982.
21. Marks-Kaufman, R.; Hamm, M. W.; Barbato, G. F.: The effects of dietary sucrose on opiate binding in genetically obese (ob/ob) and lean mice. J. Am. Coll. Nutr. 8:9–14; 1989.
22. Thornhill, J. A.; Taylor, B.; Marshall, W.; Parenti, K.: Central as well as peripheral, naloxone administration suppresses feeding in food-deprived Sprague–Dawley and genetically obese (Zucker) rats. Physiol. Behav. 29:841–846; 1982.
23. Yeomans, M. R.: Prior exposure to low or high fat milk enhances naloxone anorexia in rats. Appetite 20:125–143; 1993.
24. Wand, G. S.; Schumann, H. R.: Relationship between plasma adrenocorticotropin, hypothalamic opioid tone, and plasma leptin. J. Clin. Endocrinol. Metabol. 83:2138–2142; 1998.
25. Welch, C. C.; Kim, E.-M.; Grace, M. K.; Billington, C. J.; Levine, A. S.: Palatability-induced hyperphagia increases hypothalamic Dynorphin peptide and mRNA levels. Brain Res. 21:126–131; 1996.