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Adrenal Modulation of Opiate Induced Feeding

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LEVINE, A. S. AND J. E. MORLEY. *Adrenal modulation of opiate induced feeding.* PHARMACOL BIOCHEM BEHAV 19(3) 403–406, 1983.—Endogenous opioids have been shown to initiate feeding in sated animals. In the present study adrenalectomy enhanced the feeding response to the kappa opiate agonist, ethylketocyclazocine and the kappa/sigma opiate agonist, butorphanol tartrate. Adrenalectomy abolished the anorectic effect of naloxone at doses as high as 10 mg/kg. Corticosterone replacement did not alter the opiate induced feeding and adrenal demedullated rats continued to show enhancement to opiate induced feeding. These data suggest that in addition to the central nervous system, the adrenal medulla is involved in opiate related induction of feeding.

Adrenal Opiates Feeding Opiate-induced feeding Ethylketocyclazocine Naloxone Butorphanol tartrate

THERE is a substantial body of evidence that the endogenous opiates play a role in the central regulation of food intake [27, 29, 34, 40]. Administration of various opiates induce feeding in sated rats [27, 29, 34, 40]. Opiate receptors have been classified into mu, sigma, kappa, delta, epsilon and iota receptors using bioassays and/or binding assays to isolated brain membranes [7, 38, 50]. We and others have demonstrated that the endogenous kappa ligand, dynorphin [16, 31, 32, 33, 46], and the exogenous kappa agonist, ethylketocyclazocine [23, 35, 41] potently stimulate feeding in sated rats, suggesting a role for the kappa opiate receptor in feeding. Opiate blockade with naloxone suppresses many forms of feeding including that stimulated by starvation, tail-pinch, diazepam and catecholamines [4, 5, 15, 22, 30, 36, 42, 44]. Opiate induced feeding is integrated with monoaminergic, neuropeptidergic and purinergic mechanisms which modulate feeding behavior [27, 29, 34, 40].

Recently there has been considerable interest in the role of the pituitary-adrenal axis in the antinociceptive properties of opiates [8, 9, 25]. Opiate mediated stress induced analgesia is diminished in hypophysectomized animals suggesting a role for pituitary hormones [1, 3, 6]. The synthetic glucocorticoid, dexamethasone, which suppresses β-endorphin as well as ACTH release from the anterior pituitary, diminishes stress-induced analgesia [17,18]. However, intravenous β-endorphin administration has negligible effects on nociception [17,18]. ACTH, which regulates corticosterone production, might be indirectly involved in regulating opioid mediated stress induced analgesia [26]. Adrenalectomy diminishes this form of analgesia and corticosterone administration reinstates it [24]. Adrenal demedullation and degeneration also has been reported to diminish stress-induced analgesia [19]. As it appears that the adrenal gland has a role in the antinociceptive properties of opiates, we decided to investigate whether the adrenal gland also plays a role in the appetitive properties of opiates.

METHOD

Animals

Male Sprague-Dawley rats (125–200 g) given free access to Purina Lab Chow and tapwater and housed under conditions of controlled temperature and illumination (0600–1800 hours) were used in all studies. For adrenalectomy, animals were anesthetized with Nembutal, bilateral flank incisions were made, the adrenals were visualized and surgically excised and the incision was sutured closed. Sham animals were prepared using the same procedure except that the adrenals were not excised. All animals which had surgery were given at least a seven day recovery period prior to experimental use. Adrenalectomized rats received normal saline 10 days post-operatively and thereafter were given tap water. One group of adrenalectomized rats were given replacement corticosterone therapy (0.75 mg/kg b.i.d.) for two days and used for experimentation on the third day 2 hours following corticosterone injection. A further group of rats had the adrenal medulla surgically removed and a sham demedullation group was also included (Zivic-Miller Laboratories, Allison Park, PA).

Experimental Design

Feeding was stimulated in sated adrenalectomized and sham control rats by subcutaneous administration of the kappa opiate agonist, ethylketocyclazocine (EKC) (Winthrop Laboratories, Renssalaer, NY) between 0830–0930

hours. Feeding was quantified for the ensuing 6 hour period by providing a pre-weighed portion of Purina Lab Chow to the rats in their home cage and weighing the remaining food plus spillage at the end of the study. The effect of the kappa/sigma opiate agonist, butorphanol tartrate (BT) (Bristol Laboratories, Syracuse, NY) on feeding was also evaluated over a four hour time period. We have previously demonstrated that maximum stimulation of feeding by kappa opiates occurs between 0800–1200 hours [35].

In a second study we evaluated the effect of adrenalectomy on naloxone induced suppression of nocturnal feeding. Naloxone (Endo Laboratories, Garden City, NY) was administered subcutaneously to spontaneously feeding rats in their home cages at 1900 hours and food intake was measured over the ensuing 60 minute period.

We also evaluated the effect of EKC (35 μmol/kg) on food intake in sated adrenalectomized rats which had received corticosterone replacement and in adrenal demedullated rats and sham controls. Food intake was evaluated as described above.

Statistics

All data are expressed as mean±standard error of the mean. Data were analyzed by one way ANOVA followed by a two-tailed Student's *t*-test.

RESULTS

Adrenalectomized rats ingested significantly more food at all doses of EKC compared with sham controls (Fig. 1). Butorphanol tartrate (BT) induced greater food intake at the 3.5 μmol/kg dose in the adrenalectomized rats at all time points (Fig. 2). Opiate blockade with naloxone resulted in a significant suppression of food intake at doses ranging from 0.5–5 mg/kg in the sham rats, whereas the adrenalectomized rats were insensitive to all doses of naloxone used (Fig. 3).

As was found for adrenalectomized rats, corticosterone repleted adrenalectomized rats still ingested more food (5.2±0.4 g/6 hr; n=10; $p<0.05$) compared with the sham animals (3.3±0.8 g/6 hr; n=10) after administration of EKC. Adrenal demedullated rats also demonstrated enhanced feeding when compared to sham controls following the 3.5 and 35 μmol/kg dose of EKC (Table 1).

DISCUSSION

In the current study we found that adrenalectomized rats are more sensitive to ethylketocyclazocine induced feeding. Adrenalectomy also resulted in enhanced sensitivity to butorphanol tartrate induced feeding as indicated by increased food intake at the 3.5 μmol/kg dose in adrenalectomized rats and a shift in the dose response curve in these animals. In contrast, adrenalectomy resulted in total insensitivity to naloxone's suppressive effect on food intake.

Since dexamethasone has been reported to suppress feeding, one might hypothesize that adrenalectomized rats ingested more food following opiate stimulation due to the absence of the anorectic effect of corticosteroid. However, in our study corticosterone replacement had no effect on feeding behavior following EKC injection. This is in contrast to the report that corticosterone reinstated opioid "long term analgesia" which was blocked by adrenalectomy [19]. As adrenal demedullation and total adrenalectomy both resulted in an enhanced feeding response and corticosterone replacement had no effect on feeding behavior it appears that

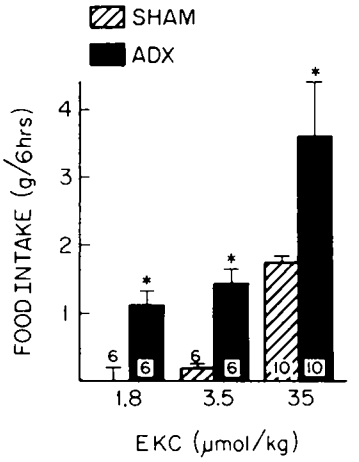

FIG. 1. Effect of adrenalectomy on ethylketocyclazocine (EKC) induced feeding. The mean quantity of food eaten by those rats given saline was subtracted from the amount eaten following administration of EKC. $F=3.74$, $p<0.01$, $^*p<0.01$. The number of animals in each group are given at the base of the figure.

TABLE 1

FOOD INTAKE FOLLOWING ADMINISTRATION OF ETHYLKETOCYCLAZOCINE IN DEMEDULLATED RATS AND SHAMS‡

	n	Food Intake (g/6 hr) Sham	DEM
EKC (35 μmol/kg)	7	3.3 ± 0.6	5.7 ± 0.6*
(3.5 μmol/kg)	8	2.5 ± 0.5	4.5 ± 0.7†
(0.35 μmol/kg)	7	3.6 ± 0.5	3.5 ± 0.7

*$p<0.025$; †$p<0.05$.
‡$F=3.66$, $p<0.01$.

the adrenal medulla plays a key role in opiate regulation of feeding. It is not clear why adrenal demedullation would enhance opiate induced feeding, although it may involve changes in catecholamine and enkephalin levels. Plasma norepinephrine is generally unchanged after adrenalectomy, but the epinephrine concentration falls markedly to a negligible concentration [11]. Aside from catecholamines, enkephalins and larger enkephalin-containing peptides are synthesized and stored in chromaffin granules of the adrenal medulla [20, 21, 43, 45, 49]. Adrenalectomy has been reported to increase binding of etorphine to opiate receptors [2,39]. This possibility could be related to an "up-regulation" of the opiate receptor due to the loss of the medullary enkephalins following adrenalectomy. Demedullation or adrenalectomy may therefore result in enhanced binding of opiates to their receptor with a concomitant enhancement of opiate induced feeding.

The involvement of the adrenal in opiate related behaviors is a complex issue. Opioid stress analgesia has been suggested to be mediated by adrenal medullary enkephalin-like peptides in one study [19], whereas corticosterone was

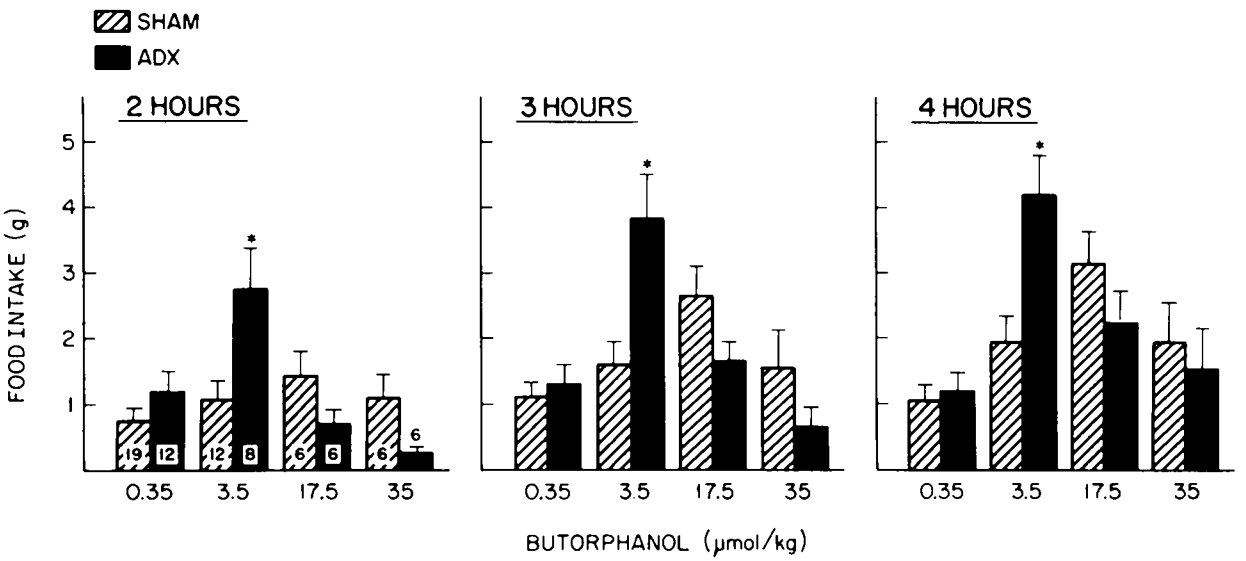

FIG. 2. Butorphanol tartrate induced feeding in adrenalectomized rats and their sham controls. The mean quantity of food eaten by those rats given saline was subtracted from the amount eaten following administration of butorphanol tartrate. 2 hr: $F(7,67)=3.81$, $p<0.005$, 3 hr: $F=8.28$, $p<0.005$, 4 hr: $F=5.68$, $p<0.005$. *$p<0.01$.

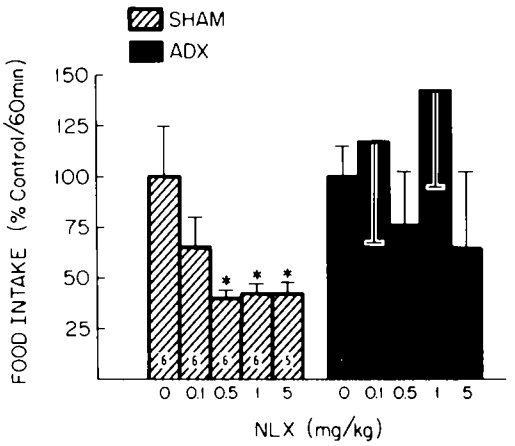

FIG. 3. Effect of naloxone on spontaneous nocturnal feeding. $F_{sham}=5.02$, $p<0.01$, $F_{ADX}=0.85$, $p<0.01$, *$p<0.01$.

reported in another study to be a critical factor in an opioid form of stress-induced analgesia [24]. It should be noted that different means of stress were used in these studies although analgesia was quantified using the tail flick test in both studies. Opiate blockade with naloxone in unstressed animals results in an increase in corticosterone concentrations [10], whereas naloxone caused a decrease in corticosterone levels in stressed animals [12,13]. Inhibition of corticosteroid synthesis results in potentiation of cold swim induced analgesia [37]. To further complicate the picture adrenalectomy blocks opioid related stress analgesia, whereas it enhances the analgesic response to morphine [17,18]. The adrenal medulla also has been reported to play a role in morphine-induced hyperthermia. Following adrenalectomy, adrenal demedullation or adrenal denervation (splanchnicotomy), 5 mg/kg morphine did not produce hyperthermia, whereas the expected hyperthermia occurred in the sham animals [47,48].

The present study suggests that opiate induced feeding involves inputs from the adrenal medulla which may involve the release of catecholamines and/or opioid-like peptides. McLean and Hoebel [28] have shown that adrenalectomized rats have an enhanced response to D-Ala-leucine-enkephalin induced feeding when the differences in basal food intake between adrenalectomized and sham animals are taken into account. These findings contrast with the decreased responsiveness of adrenalectomized animals to norepinephrine induced feeding [14]. These findings stress the importance of communication networks between the central regions and peripheral sites involved in feeding behaviors.

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