cose on central dopaminergic transmission are physiologically relevant. The lower dose of glucose (15 mg/kg, intravenous) elevated blood glucose concentrations by only 30 to 40 mg per 100 ml of blood. Food consumption or the mobilization of liver glycogen during stress can produce similar elevations (*16*). Moreover, the length of time dopaminergic activity is suppressed after a 15-mg/kg dose of glucose appears to coincide with the period of elevated blood glucose concentrations. Thus, daily physiological fluctuations in glucose availability may significantly influence dopamine-mediated activity in the brain.

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11. A brief increase in neuronal discharge was observed in a subgroup of dopaminergic cells immediately after the subcutaneous administration of glucose or saline. These cells were type A neurons (*4*), which increase their firing in response to activating sensory stimuli. This effect lasted for 30 to 60 seconds and was presumably due to local irritation at the site of injection.
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17. We thank I. J. Kopin, E. M. Stricker, A. R. Caggiula for their comments on the manuscript and D. Shirk for manuscript preparation. Supported by The National Institute of General Medical Sciences Pharmacology Research Associate Program (C.F.S.), and National Institute of Mental Health grants 5T 32-MH14634 and MH 16581 (L.A.C.).

1 February 1980; revised 17 June 1980

Fasting Associated with Decrease in Hypothalamic β-Endorphin

Abstract. *In rats that were fasted for 2 to 3 days there was a decline in hypothalamic, but not pituitary, β-endorphin. There was no change in pituitary or hypothalamic adrenocorticotropin content as a result of fasting. Endogenous opiates may be involved in physiological adaptation to fasting.*

Organisms conserve energy during fasting in part by lowering the serum concentration of 3,5,3′-triiodothyronine (T_3) and concomitantly increasing the concentration of 3,3′,5′-triiodothyronine (reverse T_3) (*1, 2*), the latter having little or no calorigenic activity. In anticipation of famine, hibernating animals accumulate extra calories either by increasing food intake and thus adipose tissue stores or by hoarding food in their nest (*3*). Hibernation and fasting both result in a state of anorexia (*3*), which may be an adaptation to prevent food-seeking energy expenditure until a time when food is plentiful.

β-Endorphin, an endogenous opiate found primarily in the central nervous system and anterior pituitary (*4*), stimulates food intake when administered intraventricularly (*5*). Concentrations of β-endorphin are increased in pituitaries from genetically obese mice and rats (*6, 7*). Naloxone, an opiate antagonist, suppresses spontaneous food intake and weight gain when administered subcutaneously to normal rats (*8*), normalizes food intake in genetically obese rodents (*9*), and awakens hibernating animals (*10*). In the present study our purpose was to determine whether fasting affects the concentration of β-endorphin in the anterior pituitary or hypothalamus, areas that are rich in this opiate and that have been linked with the regulation of feeding behavior.

Male Sprague-Dawley rats (225 to 250 g) were housed at 22°C with a 12-hour dark-light cycle in individual cages for 1 week, during which time they received daily handling and free access to food and water. They were then divided into four groups, and food was withheld from three of the four groups for 1, 2, or 3 days.

On the morning of study, the animals were decapitated, and the hypothalamus and pituitary were rapidly removed. The posterior pituitary was gently removed from the remaining pituitary with an ophthalmic forceps and discarded. The tissues were then homogenized in buffer (0.05*M* PO_4, 0.15*M* NaCl, *p*H 7.4, with 1 m*M* *N*-ethylmaleimide, a potent peptidase inhibitor) at 4°C (*4, 5, 11*) with a Brinkmann polytron at setting 6 for 10 seconds. β-Endorphin was measured by radioimmunoassay based on the method of Guillemin *et al.* (*12*) with an antibody that has an approximate 10 percent cross-reactivity with human β-lipoprotein but no cross-reactivity with adreno-

Table 1. Effect of fasting on central nervous system β-endorphin. Values represent means ± standard deviation. Nonsignificant = $P > .05$; P values represent comparisons of fasted animals to nonfasted controls (unpaired t-test). N.S., not significant.

Animal	N	β-Endorphin (nanograms per milligram of protein)			
		Hypothalamic		Pituitary	
		Concentration	P	Concentration	P
Control	20	3.45 ± 1.7		1,510 ± 998	
Fasted 1 day	12	3.40 ± 0.4	N.S.	1,151 ± 518	N.S.
Fasted 2 days	7	1.69 ± 0.8	< .02	1,231 ± 592	N.S.
Fasted 3 days	12	1.22 ± 0.3	< .01	1,171 ± 542	N.S.

0036-8075/80/1212-1271$00.50/0

Table 2. Effect of fasting on central nervous system ACTH. Values represent mean ± standard deviation. Nonsignificant = $P > .05$; P values represent comparisons of fasted animals to nonfasted controls (unpaired t-test). N.S., not significant.

Animals	N	ACTH			
		Hypothalamic		Pituitary	
		Concentration (picograms per milligram of protein)	P	Concentration (nanograms per milligram of protein)	P
Control	20	1,769 ± 1,154		1,494 ± 934	
Fasted 1 day	12	1,874 ± 1,314	N.S.	1,042 ± 299	N.S.
Fasted 2 days	7	2,142 ± 353	N.S.	1,318 ± 351	N.S.
Fasted 3 days	12	875 ± 474	N.S.	930 ± 264	N.S.

corticotropin (ACTH), melanotropin (MSH), or enkephalins. ^{125}I-Labeled human β-endorphin, antibody, and standards or unknowns were incubated for 24 hours, then separated with 2 percent charcoal coated with 1 percent bovine serum albumin in assay buffer. All samples from each experiment were assayed in triplicate in a single assay. The intraassay coefficient of variation was 3 percent, and the minimum sensitivity was 9.2 fmole per tube. All data are expressed as milligrams of protein per homogenate as determined by the method of Lowry (*13*). We performed all experiments two or more times and obtained close agreement in the results. We therefore pooled the data and analyzed them using one-way analysis of variance and Student's t-test for unpaired data. Values from fasting animals were compared to nonfasted controls.

The fasted rats remained active and were indistinguishable from control animals by external appearance and behavior. Rats fasted for 1 day had a mean weight loss of 6 percent; for 2 days, 24 percent; and for 3 days, 41 percent. Mean values for blood glucose decreased to 66 percent of control ($P < .001$) after 1 day of fasting and was 74 percent of control ($P < .01$) after 3 days of fasting. There was no change in pituitary β-endorphin throughout the fast; however, β-endorphin in the hypothalamus became significantly reduced after 2 and 3 days ($P < .02$ and $P < .01$, respectively) of fasting (Table 1).

Immunoreactive ACTH was measured in the brain tissues studied by methods previously reported (*14*) (Table 2), with ACTH (α1-24) being used as standard. Calculations were based, however, on ACTH (1-39), which has a molecular weight of 4500. Processing of the 31,000 dalton precursor to β-endorphin and ACTH into smaller peptides may or may not occur concomitantly (*15*). A concomitant processing of these peptides would result in both β-endorphin and ACTH being changed in a similar direction and magnitude. There was no change in immunoreactive ACTH in the pituitary in response to fasting, and, unlike the results with β-endorphin, there was no significant reduction in hypothalamic ACTH as a result of fasting up to 3 days. Thus, fasting may alter the synthesis, intracellular processing, or secretion of the common precursor to ACTH and β-endorphin.

Our data indicate that hypothalamic β-endorphin is modified by acute starvation. The hypothalamus appears to be involved in feeding behavior since electrolytic lesioning in this area results in either obesity or anorexia, depending on the location of the lesion (*16*, *17*). The mechanism whereby acute starvation accomplishes a reduction in hypothalamic β-endorphin is unknown. This reduction is probably due to a change in the hormonal or metabolic milieu imposed by fasting. Anorexia due to starvation has been thought by many to be in part secondary to ketosis, but the effect of ketone bodies on central nervous system β-endorphin is still unknown. The decrease in β-endorphin in the hypothalamus during a short fast may serve as a mechanism for the down-regulation of feeding behavior, enhancing energy conservation during periods of food shortage.

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18. Supported by research funds from the Department of Medicine, the Medical College of Wisconsin and the Veterans Administration.

15 April 1980; revised 4 August 1980