Gillin, *Substance Alcohol Actions/Misuse* **2** (No. 4), 233 (1981); H. L. Altshuler, P. E. Phillips, D. A. Feinhandler, *Life Sci.* **26**, 679 (1980); D. H. Ross, I. Geller, R. J. Hartmann, *Proc. West. Pharmacol. Soc.* **19**, 326 (1976).
22. K. Blum, A. H. Briggs, S. F. A. Elston, L. DeLallo, *Substance Alcohol Actions/Misuse* **1** (No. 2), 255 (1980). In experiments in which radioimmunoassay was used to investigate the interrelatedness between peptidyl opiates and ethanol-seeking behavior, we found a significant negative correlation between the amount of ethanol consumed in four different inbred strains of mice and methionine-enkephalin content in whole brains [K. Blum, A. H. Briggs, S. F. A. Elston, L. DeLallo, *Toxicol. Eur. Res.* **3** (No. 5), 261 (1981)].
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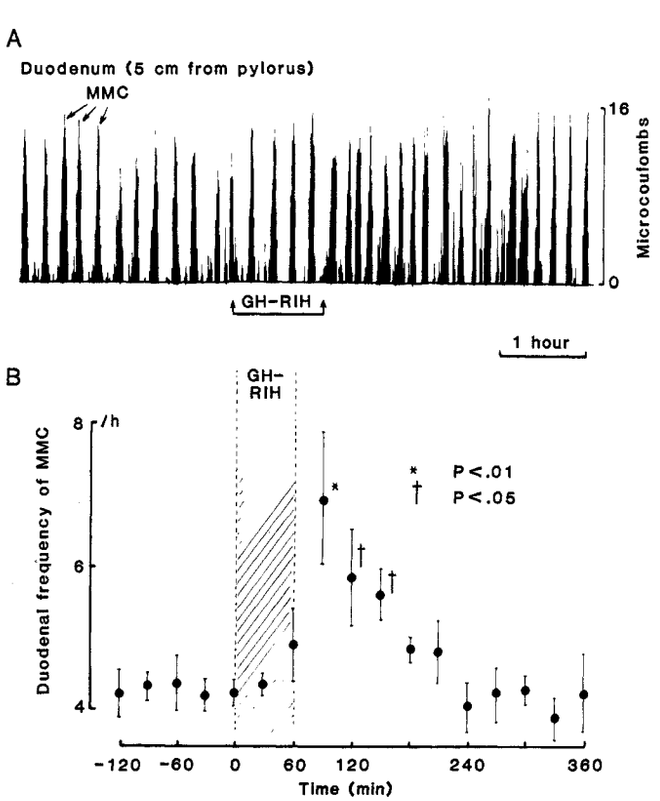
Central Regulation of Intestinal Motility by Somatostatin and Cholecystokinin Octapeptide

Abstract. *When injected continuously into the lateral ventricles of the rat, somatostatin increased the frequency of the migrating myoelectric complexes of the small intestine in a dose-related manner. A significant increase was obtained at a dose as low as 0.066 picomole per minute. In contrast, cholecystokinin octapeptide decreased the frequency of the migrating myoelectric complex of the small intestine or disrupted this pattern when injected into the lateral ventricle at rates of 0.073 to 0.23 picomole per minute. These findings support the hypothesis that somatostatin and cholecystokinin octapeptide act on central nervous system structures that are involved in the control of intestinal motility.*

Many peptides in the brain also occur in the gut. Biologically active peptides such as somatostatin and cholecystokinin octapeptide (CCK-OP), which are secreted from endocrine cells of the gastrointestinal tract, have also been identified in afferent fibers of the vagus nerves (*1*). There is evidence that CCK-OP is a satiety factor in the rat (*2*) and sheep (*3*); CCK-OP has been shown to both originate and act in the brain (*3, 4*). Somatostatin has been found in the cerebrospinal fluid (CSF) and in several areas of the brain in larger amounts than in peripheral blood (*5*), but no experiments have been performed on its role in feeding behavior.

These two neuropeptides modify the pattern of intestinal motility when they are injected intravenously. The motility of the small intestine is organized in many species in cyclical sequences of contractions occurring regularly in the duodenum and propagated through the entire small intestine. Each contraction is associated with a burst of potentials, and the recording of the electrical activity shows the presence of migrating myoelectric complexes (MMC) consisting of irregular spiking activity followed by a short period of intense activity sometimes called regular spiking activity (*6, 7*). This motor sequence progressively migrates to the ileum propelling the digestive contents at a mean rate of 15 cm/min. Successive MMC are separated by periods of quiescence. In fasted rats these MMC last 12 to 15 minutes and occur in the duodenum at about 20-minute intervals (*7*).

Systemic infusion of somatostatin increases the frequency of the MMC in the dog and reduces gastric activity (*8, 9*); in contrast, CCK-OP stimulates antroduodenal motility and disrupts the MMC pattern in the dog (*10*). Intracerebroventricular administration of somatostatin can stimulate gastric secretion (volume, titrable acidity, and pepsin), and it has been proposed that this peptide acts initially within the brain, stimulating afferent vagal fibers to cause the increase in gastric secretion via a cholinergic mech-



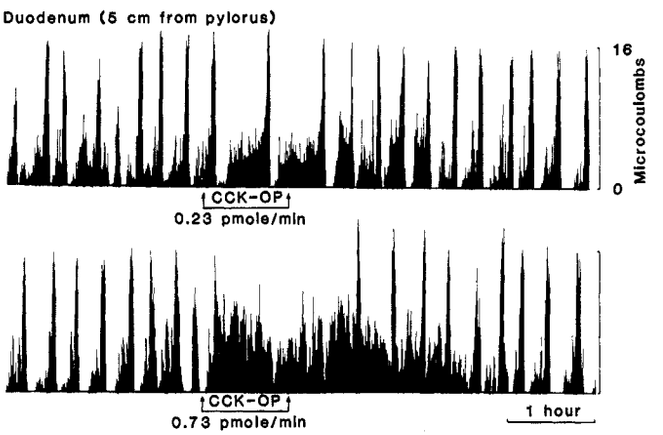


Fig. 1 (left). Influence of intracerebroventricular administration of somatostatin (*GH-RIH*) on the frequency of the migrating myoelectric complex (*MMC*) of the small intestine in a fasted rat. (A) Integrated record of electrical activity of the duodenum obtained by continuous 20-second summation of spikes collected from intramuscular electrodes. The period of somatostatin infusion (0.2 pmole/min) is indicated by the vertical arrows. The interval between two consecutive MMC shortened after somatostatin infusion. (B) Frequency of duodenal MMC (mean ± standard error, $N = 12$) measured during consecutive 30-minute periods. Somatostatin infusion (0.2 pmole/min) significantly increased the frequency of MMC. Fig. 2 (right). Comparative effects of two doses of CCK-OP administered intracerebroventricularly on the pattern of MMC of the small intestine in a fasted rat. The duration of the MMC cycle increased at the lower rate of infusion while at the higher rate (0.73 pmole/min) the MMC was disrupted in a manner similar to that seen after a meal.

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Table 1. Effects of intracerebroventricular infusion of somatostatin and CCK-OP (0 to 60 minutes) on the frequency of intestinal migrating myoelectric complexes (MMC) during and after infusion (60 to 180 minutes) in fasted rats.

Time (min)	Frequency of MMC per hour*					
	Somatostatin (pmole/min)			CCK-OP (pmole/min)		
	0.066	0.20	0.53	0.073	0.23	0.73
Control	4.2 ± 0.6	4.4 ± 0.3	3.9 ± 0.3	4.6 ± 0.5	4.6 ± 0.4	4.1 ± 0.4
0 to 60	3.9 ± 0.3	5.1 ± 0.6†	6.2 ± 0.‡	2.7 ± 0.8†	2.4 ± 0.6‡	Disruption of
60 to 120	5.2 ± 0.5	6.2 ± 0.9‡	8.2 ± 1.1‡	3.9 ± 0.4	3.5 ± 0.5†	MMC pattern
120 to 180	4.4 ± 0.6	5.2 ± 0.7	7.3 ± 1.0‡	4.2 ± 0.6	4.7 ± 0.3	3.3 ± 1.2

*Measured in the duodenum at 5 cm from the pylorus. †$P \leqq .05$. ‡$P \leqq .01$.

anism (*11*). When injected intracerebrally in sheep, CCK-OP affected the motility of the reticulo-rumen which is centrally controlled by the vagus (*3*). In this report we demonstrate that somatostatin and CCK-OP are able to regulate the intestinal motor pattern when administered centrally at doses which are inactive systemically.

Six male Wistar rats weighing 250 to 350 g were prepared for long-term electromyographic recording of intestinal motility with implanted Nichrome wires (80 μm in diameter) placed on the duodeno-jejunum at 5, 30, and 60 cm from the pylorus. The connecting wires, 60 cm in length, were exteriorized on the back of the neck (*7*). In addition, a small polyethylene catheter was inserted into the right lateral ventricle of the brain.

Electrical activity of the small intestine was recorded twice per week in 12-hour-fasted rats by means of an electroencephalograph (Reega VIII Alvar) at 13 cm/min. The identification of the MMC was facilitated by the summation of spiking activity every 20 seconds, giving an integrated record of myoelectrical activity which was recorded on a chart recorder at a slow paper speed (6 cm/hour). After 2 hours of control recordings, sterilized water alone or containing somatostatin or sulfated CCK-OP was infused intracerebroventricularly for 1 hour at a rate of 5 μl/hour; the order of treatment with somatostatin (0.066, 0.20, and 0.53 pmole/min) or CCK-OP (0.073, 0.23, and 0.73 pmole/min) was randomized according to Latin-square designs. Motility recording continued from 2 hours before to 6 hours after the beginning of infusion. The frequency of MMC was measured over 30-minute periods and compared to control infusion by variance analysis. The MMC were seen to propagate through the small intestine, each MMC being recorded successively by each set of electrodes.

Intracerebral infusion of somatostatin increased the frequency of MMC in rats in a dose-related manner. Statistical analysis showed that at 0.20 pmole/min this increase was significant and maximal (2.6 ± 0.7 MMC per hour) ($P \leqq .01$) from 60 to 90 minutes after the beginning of infusion (Fig. 1). At the highest dose (0.53 pmole/min), the increase in frequency lasted more than 2.5 hours; at the lowest level (0.066 pmole/min), the frequency was increased only during half an hour at the end of the infusion period. No effect was observed when the highest dose (0.53 pmole/min) was infused intravenously, and a similar increase in MMC frequency occurred when the rate of systemic infusion was increased 20 times. This very high rate of infusion probably caused the concentration of somatostatin to rise above the physiological level.

The intracerebroventricular infusion of CCK-OP at 0.23 pmole/min lengthened the MMC cycle for at least 2 hours; during the infusion (1 hour) only 2.4 ± 0.6 MMC were present in the small intestine as opposed to 4.6 during the control infusion (Table 1), whereas both the intensity and number of spikes recorded increased leading to an increase in the integrated activity during infusion (Fig. 2). At the lowest dose tested (0.073 pmole/min) the frequency of MMC on the duodenum was reduced only during the infusion. It was not possible to establish a dose-effect correlation in the post-infusion period because the highest rate of infusion (0.73 pmole/min) disrupted the MMC and replaced it by irregular spiking activity for 67 ± 21 minutes. When infused intravenously, CCK-OP abolished the MMC pattern only at rates higher than 10 nmole/min.

Although large doses (5 μg/kg) of somatostatin injected intravenously inhibit gastrointestinal motility in the rat, our work shows that very slow intracerebroventricular infusions of somatostatin or higher rates of intravenous infusion cause a delayed increase in MMC frequency. This effect is similar to that caused by intravenous infusion of somatostatin in the dog (*8, 9*). It seems, therefore, that somatostatin may centrally modulate the intestinal motor pattern as well as the rate of gastric secretion (*11*).

These results do not indicate how somatostatin acts in the brain to modulate gastrointestinal motor activity. However, the long delay after the start of infusion before the increased frequency of MMC was seen suggests that somatostatin has an indirect action. Changes in duodenal acid disposal have been proposed to explain the occurrence of cyclic duodenal MMC (*12*), suggesting that the central stimulation of gastric secretions by somatostatin may explain the higher frequency of MMC. A central mechanism may also account for the reduced plasma insulin concentration brought about by somatostatin infusion (*13*). Such a decrease in insulin secretion may explain the shorter duration of the MMC cycles that occurs during somatostatin infusion; a similar decrease is observed in alloxan-induced diabetes in the dog (*13*).

Sulfated CCK-OP injected intracerebrally disrupts the MMC pattern. We used the sulfated form of CCK-OP because it is this form that is found in large amounts in brain extracts of rats, the highest concentration being present in the caudate nucleus and cerebral cortex (*14*). Cholecystokinin octapeptide and its analogs increase the transport of contrast material through the human jejunum when injected intravenously and stimulate the antroduodenal motility. In contrast, CCK-33 disrupts the fasting pattern of myoelectric activity in canine intestine but does not increase the number and intensity of spike potentials as occurs after a meal or after CCK-OP infusion (*11*). This lack of a stimulating effect of CCK-33 on motility is in agreement with the finding that only the octapeptide of CCK can pass the blood-brain barrier (*15*). Experiments in vitro have shown that CCK-OP has a local effect on gastrointestinal motility by acting on the ganglion cells of Auerbach's plexus to release acetylcholine at neuroeffector

junctions (*16*). The well-known stimulatory effect of CCK-OP injected systemically has been attributed to this direct local action. The present work clearly establishes the presence of a central component.

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References and Notes

1. C. J. Dockray, R. A. Grefory, H. J. Tracy, W. Y. Zhu, *J. Physiol. (London)* **314**, 501 (1981); R. F. T. Gilbert *et al.*, *J. Neurochem.* **34**, 108 (1981).
2. S. M. Anika, T. R. Houpt, K. A. Houpt, *Physiologist* **21**, 3 (1978).
3. M. A. Della-Fera and C. A. Baile, *Science* **206**, 471 (1979); *Physiol. Behav.* **24**, 943 (1980).
4. J. F. Rehfeld and C. Kruse-Larsen, *Brain Res.* **155**, 19 (1978).
5. M. Browstein, A. Arimura, A. V. Schally, J. S. Kizer, H. Sato, *Endocrinology* **96**, 1456 (1975); S. Kronheim, M. Berelowitz, B. L. Pimstone, *Clin. Endocrinol.* **5**, 619 (1976).
6. M. L. Grivel and Y. Ruckebusch, *J. Physiol. (London)* **227**, 611 (1972).
7. M. Ruckebusch and J. Fioramonti, *Gastroenterology* **68**, 1500 (1975).
8. P. Thor, R. Krol, S. J. Konturek, D. H. Coy, A. V. Schally, *Am. J. Physiol.* **235**, E249 (1978).
9. P. Poitras *et al.*, *ibid.* **239**, G215 (1980).
10. J. J. Stewart and P. Bass, *Proc. Soc. Exp. Biol. Med.* **152**, 213 (1976).
11. Y. Tache, R. Collu, M. Brown, in *Abstracts of the Meeting on Brain-Gut Axis* (Florence, Italy), J. H. Walsh, Ed. (1981), p. 176.
12. T. D. Lewis, S. M. Collins, J. A. Fox, E. E. Daniel, *Gastroenterology* **77**, 1217 (1979).
13. L. Bueno and M. Ruckebusch, *Am. J. Physiol.* **230**, 1538 (1976).
14. R. B. Innis, F. M. A. Correa, G. R. Uhl, B. Schneider, S. H. Snyder, *Proc. Natl. Acad. Sci. U.S.A.* **76**, 521 (1979).
15. A. J. Kastin, C. Nissen, A. V. Schally, D. H. Coy, *Pharmacol. Biochem. Behav.* **11**, 717 (1969).
16. E. S. Vizi, G. Bertaccini, M. Impicciatore, J. Knoll, *Gastroenterology* **64**, 268 (1973).
17. We thank R. N. B. Kay and V. Rayner (Rowett Research Institute, Aberdeen, Scotland) for helpful criticism and revision of the manuscript. Supported in part by a grant from Institut National de la Recherche Agronomique.

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Temporal Trends in the Lead Concentrations of Umbilical Cord Blood

Abstract. *Umbilical cord blood specimens from 11,837 births between April 1979 and April 1981 have been analyzed for lead by anodic stripping voltammetry. The mean was 6.56 ± 3.19 (standard deviation) micrograms per deciliter of blood, and the range was 0.0 to 37.0 micrograms per deciliter. The mean decreased annually by 0.77 ± 0.03 microgram per deciliter, about 11 percent. Lead concentrations were higher in infants born in summer than in infants born in winter (7.17 versus 5.99, probability < .001). A Fourier model of the data is presented, and possible reasons for the decline are discussed.*

As part of a study of the effects on infant development arising from the exposure of a community population to lead, we have examined the blood lead concentrations of a large number of children at birth. Blood lead concentrations have been widely used to assess body burden, and we have used these values to select 250 children for ongoing follow-up study involving environmental and psychological measurements. Secular trends in the blood lead concentrations of pediatric populations may be useful in assessing the impact of certain recently enacted steps designed to decrease community-wide exposure to lead (*1*).

Lead was measured in 11,837 umbilical cord blood samples from the Boston Hospital for Women Lying-In Division from April 1979 through April 1981. This sample represents 97 percent of the live births. This facility serves an ethnically and economically diverse population living in the urban and suburban Boston area. Samples were not included if the mother had hepatitis or if less than 1 ml of blood was obtained. The specimens were collected in a blue-top, heparinized Vacutainer tube (B-D, Rutherford, New Jersey) after the third stage of labor when other blood samples were taken for typing. Typically, 5 ml of blood was recovered. The samples were refrigerated and kept upright during storage, which averaged 3.7 days, before the start of analysis. Samples were processed in batches of 15 in duplicate. Two sonicated aliquots were taken for digestion with a mixture of acids in a vacuum chamber in a modified microwave oven (*2*). Along with each batch of 30 samples, there were five tubes without blood (blanks) to monitor the contamination during the laboratory procedures and two tubes each of standardized pooled blood containing 10 and 20 μg of lead per deciliter of blood. Lead was analyzed by anodic stripping voltammetry (model 2014, Environmental Sciences Associates, Bedford, Massachusetts). The values reported are the averages of two or three determinations.

Contamination during blood collection and handling was found to be negligible. We measured the "fallout" of lead in the delivery suite by leaving two blue-top Vacutainer tubes open for 66 hours; 4 ng of lead was introduced. Also, a syringe assembly was flushed with 0.35 percent perchloric acid and was found to release 2.5 ng of lead in 5 ml. The Vacutainer yielded an amount of lead equal to 0.13 μg dl^{-1} after 1 day of storage with the solution in contact with the stopper and 0.34 μg dl^{-1} after 36 days, most of which came from the stopper. Sonicating and pipetting together introduced less than 0.04 μg dl^{-1}. Taken together, these contaminations total less than 0.5 μg dl^{-1} or 10 percent of the average lead concentration. No correction for these effects was applied.

In contrast, contamination by reagents and laboratory ware were not negligible and these values were subtracted from each sample. Acid digestion blanks contributed 3.8 ± 0.2 ng of lead per sample (mean ± standard error of the mean), which would represent 1.9 μg dl^{-1} in the blood, about 26 percent of the detected lead. Since the blanks contributed a sizable and variable amount of lead, we evaluated the coefficient of determination of blanks to daily mean blood level concentrations (linear regression model). We found $r^2 = .0025$ (r^2 is a measure of the percentage of variance in the blood lead concentration explained by an independent variable). Thus, 0.25 percent of the variance in the blood values could be attributed to variations in the blank.

Interbatch analytical stability was also assessed for each batch by measurements of the two biological standards. The observed values were 19.79 ± 1.01 and 10.18 ± 0.94 μg dl^{-1} (mean ± standard deviation) for 100 batches. Over time, the standards did not appear to change. Regressing time against the observed concentration yielded an r^2 of .0001 and a slope indistinguishable from zero. By both the Cochran and the Hartley tests for homogeneity of variance, samples were significantly more variable than the standards at less than the .05 level. Furthermore, the Kurtosis indices for the standards were only −0.8 and −0.6, whereas the Kurtosis index for the samples was +0.7. Thus, the variability in umbilical cord blood lead concentrations markedly exceeded the analytical variability.

Independent of the internal checks of reliability, we analyzed a series of samples that were supplied by other laboratories. In the quarterly blind comparisons sponsored by the Centers for Disease Control (Blood Lead Reference Program), the average of the absolute value of the difference between our values and theirs was 1.4 μg dl^{-1} for five

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