tae sedis: *Sciurotamias*. These represent all tribes of the subfamily recognized by J. C. Moore [*Am. Mus. Nat. Hist.* **118**, 153 (1959)]. Flying squirrels examined were *Glaucomys*, *Eoglaucomys*, *Iomys*, *Petaurista*, *Aeromys*, *Trogopterus*, *Pteromyscus*, and *Petinomys*, representing all generic groups of Petauristinae proposed by M. C. McKenna [*Am. Mus. Novit.* **2014**, 1 (1962)] and P. Mein (*3*).

14. The syndesmosis was not reported by G. H. Perkins, *Proc. AAAS* **27**, 289 (1879), probably because he prepared his specimens by maceration, which would separate the bones. That the radius and ulna were fused was reported by M. D. Bryant (*10*), but in specimens I have examined there is no osseous fusion.
15. Rotation of the ulna by the epitrochlea-anconeus muscle was proposed by J. W. Hultkrantz [*Das Ellbogengelenk und seine Mechanik* (Fischer, Jena, 1897)] and supported by R. W. Haines [*J. Anat.* **84**, 13 (1950)]. A. B. Howell [*J. Morphol.* **60**, 287 (1936)] and W. L. Straus, Jr. [*J. Anat.* **70**, 281 (1942)] have argued that the muscle only fixes the elbow joint. Supination of the forearm can be demonstrated in a freshly killed *Glaucomys*, with rotation of approximately 40°, accompanied by rotation of the ulna on the trochlear surface of the humerus. An illustration of a living *Glaucomys* with forearms supinated, probably more than 40°, is to be found in E. P. Walker (*11*).
16. The pronator quadratus muscle was reported to be present in *Glaucomys* by H. E. Peterka (*9*); it was not found in specimens dissected by M. D. Bryant (*10*) or by me (*Glaucomys*, *Petinomys*, *Pteromyscus*).
17. Photographs of gliding marsupials are to be found in S. Breeden and K. Breeden, *Animals of Eastern Australia* (Australasian, Sydney, 1967), pp. 41 and 42 and plate 3.

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Prostaglandin E_2: A Neuromodulator in the Central Control of Gastrointestinal Motility and Feeding Behavior by Calcitonin

Abstract. *Two micrograms of prostaglandin E_2 injected into the lateral ventricle of the brain in rats had the same anorectic and gastrointestinal motor effect as central administration of 0.02 unit of calcitonin. The effects of calcitonin were blocked by a previous intracerebroventricular administration of 0.25 milligram of indomethacin. These results suggest that both anorectic and gastrointestinal motor effects of calcitonin are centrally mediated by the release of prostaglandins.*

Prostaglandins mediate the responses at neuroendocrine junctions in the preoptic area of the anterior hypothalamus that are involved in thermoregulation and fever. There is now evidence that prostaglandins are also involved in other neuroendocrine responses (*1*). For instance, intracerebroventricular administration of prostaglandin E_2 (PGE_2) causes tachycardia and a rise in blood pressure (*2*), confirming that the pressure response to parenteral PGE_1 and PGE_2 is partially mediated by the central nervous system (*3*). This pressure response has been attributed to a prostaglandin-induced activation of adrenergic or cholinergic neurons rather than being considered a reflex reaction to the prostaglandin-induced hyperthermia (*4*). Prostaglandins also participate in the regulation of hypothalamic (*5*) and adenohypophyseal hormone secretions (*6*), and centrally administered prostaglandins affect gastrointestinal function; prostaglandins inhibit forestomach motility in the goat (*7*) and insulin-stimulated gastric acid secretions in rats (*8*) and cause anorexia in rats (*9*).

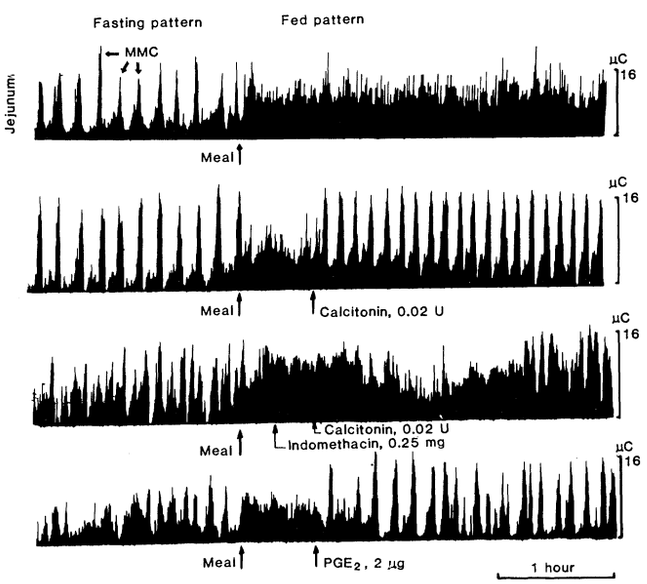

Fig. 1. Influence of intracerebroventricular administration of calcitonin (0.02 unit) with or without previous intracerebroventricular treatment with indomethacin (0.25 mg) and PGE_2 (2 µg) on the pattern of electrical activity (integrated record) of the small intestine (jejunum) in rats (vertical scale in microcoulombs). In food-deprived rats, the pattern of electrical activity is organized in cyclic migrating myoelectric complexes that were disrupted for 6 to 8 hours after feeding (fed pattern). Both calcitonin and PGE_2 restored the fasting pattern in the fed rat, and this effect was blocked by previous treatment with indomethacin.

Gastrointestinal secretory (*10*) and motility (*11*) responses, as well as feeding behavior (*12*), are now known to be mediated by neuropeptides in the brain. Calcitonin injected intracerebroventricularly in picomolar doses restores the fasting pattern of motility in fed rats (*13*), reduces gastric acid secretion (*14*), and produces anorexia in rats (*15*). Our results show that intracerebroventricularly administered PGE_2 and calcitonin have similar effects on intestinal motility and feeding behavior. This suggests that the effects of calcitonin are centrally mediated by the release of prostaglandins within the brain.

These experiments were performed in two series to investigate, respectively, the digestive motor profile and feeding behavior of rats. In the first series of experiments, 12 male Wistar rats weighing 250 to 350 g were prepared for long-term electromyographic recording of intestinal motility with implanted Nichrome wires (diameter, 80 µm) placed on the duodenojejunum at 5, 30, and 60 cm from the pylorus. The electrode wires, 60 cm in length, were exteriorized on the back of the neck. In addition, a small polyethylene catheter was inserted into the right lateral ventricle of the brain (*16*).

Electrical activity of the small intestine was recorded twice per week with an electroencephalograph (Reega VIII Alvar, paper speed 2.4 cm/min) in rats deprived of food for 12 hours. The identification of the motility pattern was facilitated by summing the spiking activity every 20 seconds; this gave an integrated record of the myoelectrical activity, which was recorded on a chart recorder at a slow paper speed (6 cm/hour).

After 2 hours of control recordings and 40 minutes after the beginning of a meal consisting of 6 g of a balanced laboratory ration, 5 µl of sterilized water either alone or containing 0.02 unit (1 unit = 208 ng) of salmon calcitonin (Calsyn) was injected intracerebroventricularly twice in each rat. Similar injections were given 20 minutes after intracerebroventricular administration of 0.25 mg of indomethacin dissolved in 10 µl of 5 percent $NaHCO_3$. The rats also received an intracerebroventricular injection of 2 µg of PGE_2 (Upjohn) dissolved in 5 µl of

sterilized water (*17*) or an intramuscular injection of 100 μg of PGE_2, with or without previous treatment with indomethacin. All of the treatments were given in a random fashion. The results were analyzed with a paired *t*-test and covariance analysis applied to multiple-regression curves.

In food-deprived rats, as in other species, the electrical activity of the small intestine is organized into migrating myoelectric complexes (MMC) (*18*). In the rat the MMC lasted 9 to 10 minutes and recurred at approximately 14-minute intervals (Fig. 1); MMC, which propelled the intestinal contents, were propagated on the duodenojejunum at a rate of 3.4 ± 0.7 cm/min. After feeding 6 g of laboratory ration, this pattern was disrupted for 6 to 8 hours. Similar disruption of the MMC by feeding has been observed in other species (*19*).

Calcitonin (0.02 unit), given intracerebroventricularly 40 minutes after feeding, changed the electrical activity recorded at each electrode site from the fed, disrupted pattern to the fasting MMC pattern for 132 ± 27 minutes (mean ± standard deviation). Similarly, PGE_2, 2 μg given intracerebroventricularly, to fed rats restored the MMC pattern immediately for 83 ± 31 minutes; a 50 times larger dose of PGE_2 given intramuscularly had no effect. The disrupting effects of calcitonin (0.02 unit) were abolished, and those of PGE_2 slightly reduced, to 52 ± 15 minutes ($P < 0.05$), when they were injected 20 minutes after an intracerebroventricular administration of 0.25 mg of indomethacin (Fig. 1).

We investigated the anorectic effects of calcitonin in 20 additional rats equipped with lateral ventricle catheters and housed singly in wire-bottomed cages. The rats were accustomed to receiving the laboratory ration (UAR, France) for 2 hours each day from 9 to 11 a.m. Food intake was measured at the end of the feeding period. Ten minutes before being given access to food, the rats received intracerebroventricular injections of 10 μl of sterilized water; calcitonin (0.02 unit), with or without a previous intracerebroventricular injection of indomethacin (0.25 mg); or PGE_2 (2 μg) or indomethacin (0.25 mg) alone. Each treatment was applied to groups of four rats and the order was randomized in a Latin square design.

The 2-hour food intake was 13.5 ± 3.3 g in rats receiving sterilized water. Food intake was reduced by 34.4 percent in rats treated with calcitonin and by 44.7 percent in animals receiving PGE_2 (Table 1).

Table 1. Comparative influence of intracerebroventricular administration of calcitonin and PGE_2 and antagonistic effects of intracerebroventricular treatment with indomethacin on 2-hour food consumption in the rat. Data are means ± standard error for 20 rats. Treatments were applied in randomized order at 3-day intervals to groups of four rats.

Treatment	Food intake (g)
Control distilled water (10 μl)	13.5 ± 3.3
Calcitonin (0.02 unit)	8.6 ± 3.1*
PGE_2	6.7 ± 1.7*
Indomethacin (0.25 mg)	17.7 ± 2.4**
Indomethacin (0.25 mg) + calcitonin (0.02 unit)	17.3 ± 5.5*†

*Significantly different from control value at $P \leq 0.01$; **at $P \leq 0.05$. †Not significantly different ($P \geq 0.05$) from indomethacin alone.

Rats treated with indomethacin consumed more food (17.7 ± 2.4 g) than did control rats. The anorectic effects of the intracerebroventricularly administered calcitonin were also abolished by previous treatment with indomethacin, the food intake (17.3 ± 5.5 g) being not significantly different ($P > 0.05$) from that observed after indomethacin alone.

Calcitonin present in the pituitary and hypothalamus of several species of mammals may modify or regulate physiological functions such as gastric acid secretion (*14*), intestinal motility (*13*), prolactin release, and the perception of pain (*20*). The central origin of the anorectic effect of calcitonin has been confirmed (*15*), and injection of Ca^{2+} into the hypothalamus or cerebroventricles elicits feeding (*21*). Calcitonin, like some other neuropeptides, depresses Ca^{2+} uptake by the hypothalamic cells (*22*) and may exert its effects by altering neuronal Ca^{2+} fluxes.

We suggest that prostaglandins may be intermediates in at least some of the responses to calcitonin. The mode of action of prostaglandins varies from tissue to tissue and can result from the occupancy of prostaglandin receptors or from the arachidonic acid metabolites acting within the cells or as local hormones modulating the activity of surrounding cells. In the cat spleen, Ca^{2+} can antagonize the inhibition of sympathetic transmission by prostaglandins (*23*). This suggests that some effects of prostaglandins, like those of calcitonin, may involve changes in Ca^{2+} fluxes or Ca^{2+} binding.

In our experiments, PGE_2 had the same effects as calcitonin on appetite and intestinal motility; intracerebroventricularly administered indomethacin blocked both the anorectic and motility effects of calcitonin. Prostaglandins have anorectic properties in rats, the sites of action being the medial and lateral hypothalamus (*24*). The simplest explanation for these results is that prostaglandins are intermediates in the responses to calcitonin.

The peripheral pathways in these central responses to calcitonin and prostaglandins are not established; cholinergic or adrenergic neurons may participate, but none of the antagonists that we have tested (α- and β-blockers, anticholinergic and antidopaminergic drugs) have blocked the effect of calcitonin or prostaglandins on intestinal motility. Whatever the pathway, these results suggest that there may be a link between the central control of food intake and the changes in gastrointestinal motility occurring after feeding.

M. J. Fargeas
J. Fioramonti
L. Buéno

Station de Pharmacologie-Toxicologie, Institut National de la Recherche Agronomique, 180 chemin de Tournefeuille, 31300 Toulouse, France

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Prolonged Survival and Remyelination After Hematopoietic Cell Transplantation in the Twitcher Mouse

Abstract. *The twitcher mouse is an animal model of galactosylceramidase deficiency (Krabbe's disease), a human sphingolipidosis. The effects of hematopoietic cell transplantation as potential enzyme replacement therapy were examined in the twitcher mouse. Survival in twitcher mice with transplants was significantly prolonged and was associated with gradual repair of demyelination in peripheral nerves. In contrast, there was no improvement in the neurodegenerative process in the central nervous system after transplantation. These observations indicate that cellular transplantation may effectively provide in vivo enzyme replacement for the peripheral manifestations of genetic storage diseases. Strategies to perturb the blood-brain barrier may be necessary for enzyme replacement to be therapeutic in diseases with central nervous system manifestations.*

The treatment of lysosomal hydrolase deficiency states such as the mucopolysaccharidoses and sphingolipidoses is limited to symptomatic and supportive care, since no definitive therapy is available. Several in vitro studies have indicated that these heritable storage diseases might be treated by specific replacement of the deficient enzyme (*1, 2*). However, isolation and purification of adequate quantities of specific lysosomal hydrolases are impractical, and in vivo administration of limited amounts of enzyme has not been associated with consistent clinical improvement in human recipients (*3*).

Bone marrow transplantation repopulates the reticuloendothelial and lymphohematopoietic systems of the recipient with normal donor cells (*4–6*). Marrow transplants from normal mice into catalase-deficient (*7*) or glucuronidase-deficient (*8*) animals restore normal enzyme activity in the peripheral blood and other tissues of the recipients. Theoretically, bone marrow transplantation may be therapeutic in human storage diseases by providing a proliferating, self-renewing, enzymatically normal cell mass (*9*). The lack of suitable animal models of lysosomal hydrolase deficiency states has precluded the preclinical evaluation of cellular transplant strategies in these disorders.

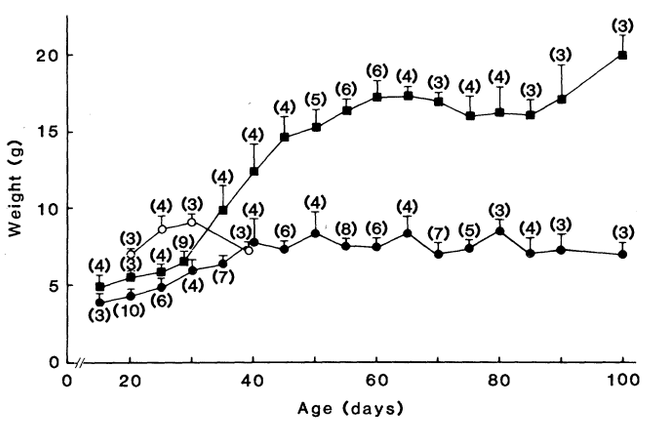

Fig. 1. Weight (mean ± standard error) of untreated twitcher mice, twitcher mice that received hematopoietic cells at 10 days of age, and enzymatically normal control littermates that also received transplants. Numbers in parentheses are the number of animals studied. After the immediate post-transplant period, control littermates gained weight normally. Over the two- to threefold prolongation of life-span the weight of treated twitcher mice did not differ significantly from that of untreated twitchers.

The twitcher mouse, a recently described neurological mutant, appears normal at birth, develops signs of central nervous system (CNS) dysfunction and peripheral demyelination by 3 to 4 weeks of age, and dies from progressive neurodegeneration by 5 to 7 weeks (*10*). Enzymatic studies have shown that affected twitcher mice lack galactosylceramidase, a lysosomal hydrolase that catalyzes the degradation of sphingolipid (*11*). Neuropathologic observations in twitcher mice have demonstrated characteristic periodic acid Schiff (PAS)–positive globoid cells in the central and peripheral nervous systems and extensive demyelination, with mononuclear cell infiltration, of peripheral nerves (*12*). From clinical, enzymologic, and neuropathologic standpoints, the twitcher mouse appears to be an authentic animal model of human galactosylceramidase deficiency (globoid cell leukodystrophy; Krabbe's disease), a progressive neurodegenerative sphingolipidosis (*13*).

We examined the effects of transplanting hematopoietic cells from normal congenic mice into presymptomatic galactosylceramidase-deficient homozygous twitcher mice. For these studies we used offspring of breeding pairs of C57BL/6J mice heterozygous for the twitcher mutation (+/*twi*). Since affected twitcher mice are asymptomatic for the first 3 to 4 weeks of life, we ascertained their genetic status by determining galactosylceramidase activity in aqueous homogenates of clipped tail tips from 7-day-old offspring (*14*). In our laboratory, galactosylceramidase activity in affected twitcher mice was ≤0.10 nmol/hour per milligram of protein, compared with >0.70 nmol/hour in unaffected control littermates. Twenty-four hours before hematopoietic cell transplantation, affected mice and enzymatically normal (+/+) control littermates received 900-rads of total body irradiation from a ^{137}Cs source at a dose rate of 120 rads per minute. At 10 days of age each mouse received an intraperitoneal injection of bone marrow cells (1.0×10^7 to 1.5×10^7) and spleen cells (3.5×10^7 to 5.0×10^7) freshly obtained from 6- to 8-week-old female congenic C57BL/6J mice that had normal galactosylceramidase activity. In addition, these donor mice were homozygous for the A isozyme of erythrocyte glucose-phosphate isomerase 1 (GPI-1A). Since C57BL/6J mice, including those with the twitcher mutation, are homozygous for the B isozyme of GPI-1, we were able to assess hematopoietic engraftment after transplantation by electrophoretic analysis of GPI-1 isozyme patterns in 5-μl samples of blood from recipient mice