kainic acid (Fig. 2) (*11*); we looked at the GABA receptors in the pars reticulata of the substantia nigra 12 days later. [^{3}H]Muscimol binding was 20 to 60 percent above that on the control side (Fig. 1). The change was secondary to an increase in receptor number, with no change in K_D. The magnitude of the change was correlated with the size of the striatal lesion and its location. The supersensitivity was confined to the rostral, medial, and intermediate parts of the pars reticulata and did not involve the pars compacta.

This method can be applied to other ligands, such as deoxyglucose, flunitrazepam, strychnine, and phencyclidine (*12*). Furthermore, we have been able to study tissues after 2-[^{14}C]deoxyglucose administration. The labeled glucose analog is eluted in the successive buffer washes, and radioactivity in the blanks is not detectable. The method is 1000 times more sensitive than the homogenate techniques. Areas that when evaluated by homogenate studies require pooled tissue from microdissections can be evaluated easily in a single animal with this method. For receptor studies in lesioned animals, this method can supply kinetic, saturation, and competition data, as well as the detailed regional localization of receptor changes.

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Stimulation of Intestinal Calcium Transport and Bone Calcium Mobilization by Prolactin in Vitamin D–Deficient Rats

Abstract. *In vitamin D–deficient rats intestinal calcium transport increased significantly 4 hours after an injection of prolactin, reached a maximum after 8 hours, and declined to preinjection levels after 24 hours. Similarly, in vitamin D–deficient rats fed a diet low in calcium or phosphorus prolactin stimulated an increase in serum calcium in both groups and an increase in serum phosphorus in the rats fed the diet low in phosphorus. Thus it appears that prolactin affects organs involved in calcium regulation in a manner that is independent of the vitamin D endocrine system.*

For some time it has been accepted that intestinal calcium transport responds solely to the active form of vitamin D—1,25-dihydroxyvitamin D_3 [1,25-$(OH)_2D_3$]—or to one of its analogs (*1–3*). Liberation of bone calcium is believed to result from a combined action of the vitamin D endocrine system and parathyroid hormone (*1–4*). However, it is also known that prolactin can stimulate absorption of calcium by the intestine (*5, 6*). This has been believed to result from prolactin stimulating the conversion of 25-hydroxyvitamin D_3 (25-OH-D_3) to 1,25-$(OH)_2D_3$ (*7*). Recently it was shown that female rats severely deficient in vitamin D can reproduce and lactate, demonstrating that calcium can be obtained by the pregnant female from intestinal and bone sources (*8–10*). The loss of mineral from bone during reproduction and lactation in the vitamin D–deficient mothers was similar to that in vitamin D–replete control mothers (*9*). Also, intestinal calcium transport increased in the vitamin D–deficient mothers (*10*). It was suggested that some humoral factor might be involved in the liberation of calcium from intestine and bone (*9, 10*). The most likely hormone that could satisfy this role is prolactin, which is secreted at precisely the appropriate time and is maintained at elevated concentrations in the circulation during pregnancy and lactation (*11–13*).

To test the possibility that prolactin affects intestinal calcium transport and the liberation of calcium from bone in a manner that is independent of vitamin D, weanling male albino rats (Holtzman) were placed for 10 to 16 weeks on a diet low in vitamin D (*14*). That this regimen led to vitamin D deficiency was revealed not only by hypocalcemia and lack of growth but also by the lack of detectable levels of 25-OH-D_3 and 1,25-$(OH)_2D_3$ in plasma (*15, 16*). These rats were then divided into groups of six to nine ani-

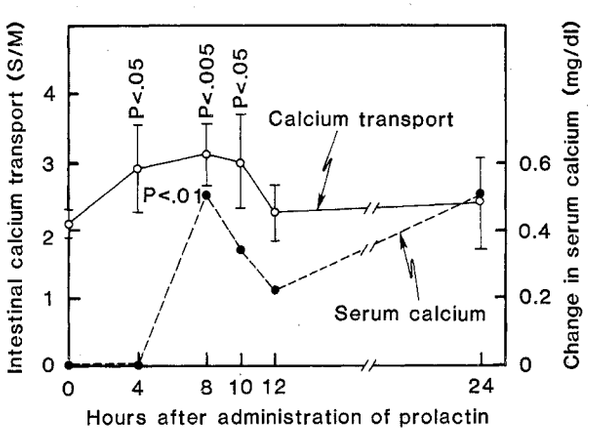

Fig. 1. Changes in intestinal calcium transport and serum calcium concentration in vitamin D–deficient male rats following an injection of prolactin (250 μg per 250 g of body weight). The data (means ± standard deviations) are expressed as either the rise in serum calcium or as the ratio of ^{45}Ca in the serosal medium (*S*) to ^{45}Ca in the mucosal medium (*M*). Before incubation the ^{45}Ca concentration was equal in both the serosal and mucosal media.

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mals. One group was given vehicle and immediately decapitated for the measurement of intestinal calcium transport by the everted sac method (*17*). The remaining groups were injected intraperitoneally with prolactin (250 μg) (*18*) and killed 4, 8, 10, 12, or 24 hours later. Serum was collected for the measurement of serum calcium concentration by atomic absorption spectrometry on a sample diluted with 0.1 percent $LaCl_3$ (Fig. 1). Within 4 hours after prolactin injection intestinal calcium transport increased significantly, reaching a maximum after 8 hours and then falling to preinjection levels after 12 to 24 hours. Since no 25-OH-D_3 or 1,25-$(OH)_2D_3$ could be detected in the plasma of these rats [the detection limit of 1,25-$(OH)_2D_3$ is 2 pg/ml], the increase in intestinal calcium transport could not have been caused by increased levels of residual metabolite. A 10-pmole dose of 1,25-$(OH)_2D_3$ would be required for the intestinal response to prolactin observed in this experiment (*19*). Such a dose would increase plasma 1,25-$(OH)_2D_3$ to 30 to 40 pg/ml (*20*).

Other groups of rats were then tested to determine the optimal dose of prolactin required for maximum intestinal calcium transport within 8 hours. This dose was found to be 100 to 250 μg per 250 g of body weight. Serum calcium concentration also increased significantly after 8 hours (Fig. 1). The question then arose as to whether the elevation in serum calcium was the result of the improved intestinal calcium absorption or due to the liberation of calcium from bone. An experiment was therefore carried out in which vitamin D–deficient rats, 10 weeks after weaning, were placed for an additional 2 weeks on a diet low in either calcium or phosphorus. The rats were then given a single intraperitoneal injection of prolactin (300 μg). Eight hours later blood calcium and phosphorus were measured (Table 1). Serum calcium was increased in both groups, and serum phosphorus was significantly increased in rats on the diet low in phosphorus.

These results suggest that prolactin can act independently of vitamin D and parathyroid hormone in stimulating intestinal calcium transport and the liberation of calcium and phosphorus from bone (*21*). Thus, in pregnant mammals prolactin may have a secondary role in liberating calcium to support fetal calcification and the lactation process. If the effect of prolactin can be confirmed in the pregnant or lactating vitamin D–deficient rat, prolactin would have to be considered as an important regulator of calcium metabolism under some circumstances. These results may aid in the elucidation of certain disease states [it has, however, been reported that hyperprolactinemia in man does not appear to be associated with abnormal calcium metabolism (*22*)]. The results are sufficient to warrant additional detailed investigation into the possible role of this hormone in the regulation of calcium, especially during pregnancy and lactation.

Table 1. Effect of prolactin on bone mineral liberation in vitamin D–deficient rats fed diets low in calcium or phosphorus. Diet A contained 0.02 percent calcium and 0.3 percent phosphorus and diet B contained 1.2 percent calcium and 0.1 percent phosphorus. Values are means ± standard deviations.

Diet	Prolactin treatment	Serum calcium (milligrams per 100 ml)	Serum phosphorus (milligrams per 100 ml)
A	No	3.4 ± 0.1	8.4 ± 0.8
	Yes	3.8 ± 0.3*	9.1 ± 1.0
B	No	7.9 ± 0.2	2.4 ± 0.4
	Yes	8.2 ± 0.2†	3.4 ± 0.5*

*Significantly different from corresponding control value at $P < .01$ (Student's t-test). †$P < .025$.

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Active Ion Transport in Dog Tongue: A Possible Role in Taste

Abstract. *An in vitro preparation of the dorsal epithelium of the dog tongue actively transports ions, producing a transepithelial potential difference characteristic of the ions and their concentration. Hypertonic sodium chloride solutions generally cause increased potentials and short-circuit currents and reduced resistances when placed on the mucosal surface. This hypertonic flux is eliminated by ouabain and is not found in ventral lingual epithelia. When either sodium acetate or tetramethylammonium chloride is substituted for sodium chloride in the mucosal medium, the currents are diminished but their sum at a given concentration approximates that for sodium chloride at the same concentration. This result suggests a current composed of inward sodium ion movement and outward chloride ion movement. Actively regulated potentials and currents, whether generated in the taste buds or in supporting cells, may be important in both normal chemotransduction and in taste responses evoked by currents passing through the tongue.*

The dorsal lingual epithelium has been generally regarded as a tissue of low permeability (*1*). Although this conclusion seems justified for a variety of nonelectrolytes diffusing across nonmetabolizing tissue, we show here that it is invalid for ionic species in metabolizing epithelia. This result is particularly important in the area of taste reception because in current theories of salt taste reception it has been assumed that the lingual epithelium is virtually impermeable to ion transport (*2*). This has resulted in models of taste reception in which ion transport across the lingual epithelium has been largely ignored as a factor in receptor activation. There has been rather an emphasis on events at the interface between the oral cavity fluid and the receptor cell membrane. Ion transport

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