conditions which release the amines from adrenal medulla (COMLINE[16]). VAN ORDEN[17] came to the same conclusion after trying to release the catecholamines with pharmacological agents. However, it should be pointed out that even extensive discharge of catecholamines from the tissue might escape detection if only observation by eye is used. Therefore, judging from the present results, discharge of an endocrine nature cannot be ruled out. The paraganglia of human fetus and newborn rabbit, on the other hand, react clearly to strong hypoxia (BRUNDIN[11], HERVONEN and KORKALA[12]). The catecholamine-storing cells induced by glucocorticoids might be different from normal ones with respect to the endocrine function.

Zusammenfassung. In den paraganglionären Zellen der neugeborenen Ratte wird die Catecholaminfluoreszenz durch schwere Hypoxie nicht beeinflusst.

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Quantitation of the Number of Villi and Crypts in the Intestine of Rodent Animals

In the studies of gastro-intestinal damage induced by radiation, many kinetic analyses[1–5] on the cell population in the epithelium of gut in rodent animals have been made on tissue sections, and it was proved that the epithelial cells were produced in the crypt, transfered upwards to the villus tip, and extruded into the lumen. The explanation has been gained through the studies of 2-dimensional relationship of the crypt to the villus. Accurate information is not always available concerning the 3-dimensional relationship of the crypt to the villus[6–10]. By the routine histological technique using paraffin, it might be rather difficult for the crypts and the villi to be fixed in the desired orientation, e.g., so as to run parallel with each other, so that accurate 3-dimensional relationships between them were not proved.

The present study was undertaken to examine the other from the routine histological technique to obtain accurate measurements of the respective number of crypts and villi per unit area of mucosal surface.

Experimental animals used were mice, rat, and golden hamsters, all 3-month-old adults. From each animal small intestines were removed, cut off at a desired length, and fixed in a 3:1 mixture of 70% ethanol and glacial acetic acid for 1 to 2 h, and then hydrolized in N-HCl[10] by means of standing at room temperature for 1 to 2 days.

The segments were opened by cutting along the long axis of intestine. The mucous membrane of the flattened segments was swept gently with a plastic or bamboo-made knife taking care that the lamina propria was not damaged. It is favorable that the edge of knife is dull. Sweeping was done in a pail continuously supplied with tap water. By such sweeping the epithelial cells of villi were almost completely removed leaving lamina propria and crypts. After sweeping the segment was placed in a Petri dish with a small quantity of water and covered gently with a thin leaden perforated plate.

Through the perforated area in the leaden plate, the crypts and the villi were photographed using a microscope equipped with water-immersion lenses of suitable magnification. Enumeration of the number of crypts and villi in unit surface of mucosa was performed in the photographs. Examples of such photographs are shown in Figures a) and

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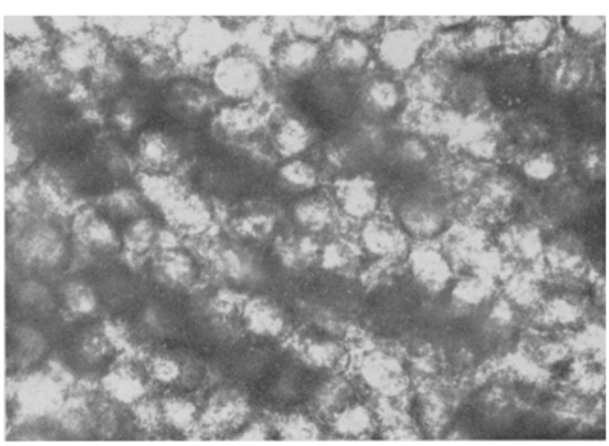
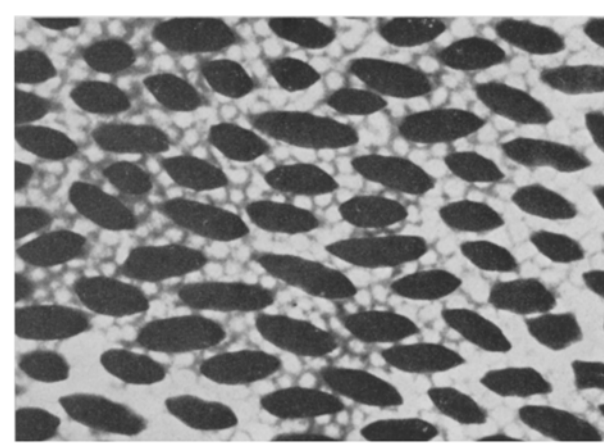


a) Crypts (× 100)



b) Villi (× 40)

Photographs of crypts and villi in intestinal mucosa of golden hamster

C/V-values of 8 fragments in the intestines of mouse, rat and golden hamster

Segment	Duodenum				Jejunum		Ileum		
Numbers	1	2	3	4	5	6	7	8	Means
Mouse	7.6[a]	7.3	5.8	4.9	4.5	4.2	4.3	4.6	5.4
	1.18[b]	1.33	1.04	0.57	0.59	0.68	0.53	0.70	
Rat	18.2	16.7	14.3	13.2	12.2	10.4	10.5	10.2	13.2
	3.15	3.22	1.82	1.67	1.29	0.88	0.123	0.83	
Golden	5.3	5.3	4.1	4.5	4.0	4.2	4.1	3.3	4.4
hamster	1.44	0.68	0.76	0.71	0.68	0.79	0.57	0.49	

[a] The average value of 9 measurements on 3 animals [b] S.D.

b). In Figure a), the edge of crypt was brought into focus, so that villi were visualized as dim spindle-like images.

The small intestine was cut up evenly into 8 segments and the number of crypt and villus was mesaured, respectively, in the phrotographs taken at 3 different regions in each segment. In the Table are shown the ratios of crypts to villus of 3 animals tested; hereafter the ratio will be referred to as 'C/V-value'. The Table reveals that there are apparent gradual decreases of the C/V-values along the intestine from upper duodenum to lower ileum, and that the averages of all 8 segments coincide with the value at the 4th segment in all animals tested. At the 4th segment, therefore, the mean C/V-values are comparable with each other. It is noticeable that the mean C/V-value is different in each species and that of rat is markedly higher than those of the others. These facts may imply the probable variance in static or dynamic aspects of epithelial cell population of various animals, and furthermore the difference of mean survival times in the gastrointesinal death induced by radiation.

In fact, the author has gained a few noticeable features on dynamic aspects of epithelial population through reconsidering the problem by introducing the C/V-values into the 2-dimensional pattern known from the studies of conventional histological sections. They will be reported elsewhere.

Zusammenfassung. Quantitative Bestimmung der Krypten- und Zottenzahl im Darm der Nagetiere. Die Zahl der Zotten ist bei verschiedenen Tieren eine unterschiedliche und nimmt zudem vom Duodenum gegen das Coecum zu ab.

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Myoneural Junctions in Larval Ascidian Tail

Myoneural junctions are typically formed at the surfaces of muscle cells by peripheral nerve fibers which extend from a centrally located spinal cord or nerve cord. Two instances are known, however, in which this pattern is reversed. In the nematode worm *Ascaris*[1] and in the primitive chordate *Amphioxus*[2] it is the muscle cells that send processes toward the nerve cord, and myoneural junctions are established at the surface of the nerve cord rather than within the muscle.

The tail of the larval ascidian *Amaroucium constellatum* is known to contain acetylcholinesterase[3]; however, no myoneural junctions have been found previously in the tail muscle[4]. This report presents evidence for a unique type of muscle innervation in this animal which bears some resemblance to that reported previously in *Ascaris* and *Amphioxus*: muscle fibers run right alongside the dorsal nerve cord in close apposition to it throughout the entire length of the tail and it is at this interface that the structural specializations characteristic of myoneural junctions occur.

Methods. Free-swimming larvae were collected from parent colonies of *Amaroucium constellatum*. Colonies which have been kept in running water in the dark for several hours will readily produce hundreds of larvae within 15 min after their exposure to light. The rapidly-swimming larvae were collected with a Pasteur pipette and immediately placed in a freshly-prepared solution of 5% glutaraldehyde in 0.1 *M* phosphate buffer (pH 7.4). Specimens were fixed for approximately 3 h at room temperature, followed by a rinse in buffer, and were post-fixed with 1% OsO_4 in phosphate buffer. The tissue was then dehydrated in a graded series of methanol solutions followed by propylene oxide, and embedded in Araldite. Thick sections for light microscopy were stained with toluidine blue. Thin sections were stained with uranyl acetate und lead hydroxide, and then examined by electron microscopy. Numerous animals were surveyed and in two cases serial sections of the tail were made as follows: a survey series of 5 to 10 sections 1–2 μm thick was made, followed by a series of thin sections. This pattern was repeated for the length of the tail. In 4 other animals, random thick and thin sections at various levels representing the entire tail were studied. In one experiment, tails were amputated from free-swimming larvae at various distances from the trunk and at various time intervals after emergence from the parent organism until the beginning of tail resorption. The amputated tails were then observed for signs of motility.

Results. In transverse sections of the proximal part of the tail, 8 muscle cells can be seen surrounding the notochord. The dorsal nerve cord, or neural tube, is situated at the dorsal edge of the notochord, wedged be-

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