It has been generally accepted that most of APUD cells are derived from the neural crest. However, there exists evidence contrary to a neural crest origin for the pancreatic islet cells[13,14] although their possible neuroectodermal origin has not been precluded and data supporting the latter hypothesis has been reported[15].
Our findings, while not providing evidence to support either of the above views, indicate that the rat pancreatic islets do not differ from pancreatic islets in other species in terms of their ability to take up, decarboxylate and store amines.

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So-called annular gap junctions in bone cells of normal mice

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Summary. Spherical bodies consisting of a granular matrix and a pentalaminar limiting membrane were found in cells from the proximal tibial metaphysis of normal mice. The structures measured about 280–570 nm in diameter and were located mainly in the cytoplasm of osteoblasts and occasionally in preosteoblasts. The granules within the bodies resembled ribosomes. The multi-layered composition of the limiting body membrane was identical with that of intercellular gap junctions.

Peculiar spherical structures characterized by a pentalaminar limiting membrane have been observed in cells from a variety of tissues of different species[2–8]. The bodies have been termed 'sphaerae occlusae'[3], 'bulb gap junctions'[4], 'gap junction-bounded vesicles'[5,7], 'annular nexuses'[6], or 'annular gap junctions'[6]. Recently, such intracytoplasmic structures were found in cells from the periosteum of normal and lathyritic rats[8]. Electron microscopic study of cells from the metaphyseal trabecular bone tissue of the tibia of normal mice has revealed the presence of similar configurations as described below.
Materials and methods. Six normal female NMRI mice and 3 normal $(C3H \times 101)F_1$ hybrid mice, 2 males and 1 female, were used. The randomly outbred NMRI mice weighing 19–25 g were 3–4 weeks old. The inbred hybrid mice with a weight of 11 g were 3 weeks old. Immediately after sacrifice of the apparently healthy mice, the proximal part of the right tibia of each animal was rapidly removed and split longitudinally. Both halves of each tibial metaphysis were cut into cubes of about 1 mm^3. The specimens from 3 NMRI mice were fixed in phosphate-buffered 1% osmium tetroxide and those from the other 3 NMRI mice in phosphate-buffered 6.25% glutaraldehyde. The specimens from the 3 $(C3H \times 101)F_1$ mice were fixed in cacodylate-buffered 3% glutaraldehyde. The glutaraldehyde-fixed tissue cubes from the NMRI mice were postfixed in phosphate-buffered 1% osmium tetroxide and those from the hybrid mice in chrome-osmium[9]. Following dehydration through a graded series of ethanols to propylene oxide, the specimens were embedded in Epon 812. Thin sections were cut with a diamond knife on a Reichert Om U3 microtome and stained with uranyl acetate and lead citrate. They were examined in an AEI EM6B electron microscope.
Results. Many bone cells which were randomly examined in the undecalcified specimens from the proximal tibial metaphysis of the young normal mice contained peculiar membrane-bound structures (figures 1–4). These intracytoplasmic bodies which were more frequently seen in the NMRI than in the $(C3H \times 101)F_1$ mice were spherical and varied in size. The outer diameters of the inclusion bodies ranged from about 280 to 570 nm. The bodies consisted of a matrix being surrounded by a multi-layered limiting membrane. The matrix, similar in appearance to the cellular cytoplasm, contained diffusely scattered granules which resembled ribosomes (figures 1–4). Occasionally, an electron-lucent halo was interposed between the granular core of a body and the annular limiting membrane (figure 4).
The unusual membrane limiting the bodies had a pentalaminar appearance which was not modified in its preservation by the various fixatives used. The membrane consisted of an innermost and outermost electron-dense layer, both similar to the outer leaflets of a unit membrane. Between these 2 layers, an intermediate punctuated one was to be seen. This leaflet seemed to consist of periodically arranged subunit particles appearing in cross-section as a chain of electron-dense dots. It was separated from the innermost and outermost electron-dense leaflet by an electron-lucent layer on either side. The five-layered membrane was approximately 20 nm thick. Its multi-layered composition was identical with that of intercellular gap junctions[10] ('tight junctions'[11,12]) occasionally observed between cells from the trabecular bone tissue examined (figure 3).
The innermost leaflet of the limiting membrane of a few inclusion bodies was continuous with a saccular structure measuring up to about 150 nm in size (figures 2 and 3). Such a saccule was formed by a protrusion of the innermost layer of the pentalaminar membrane into the matrix of a body. In one body, an intrasaccular dense, knob-like

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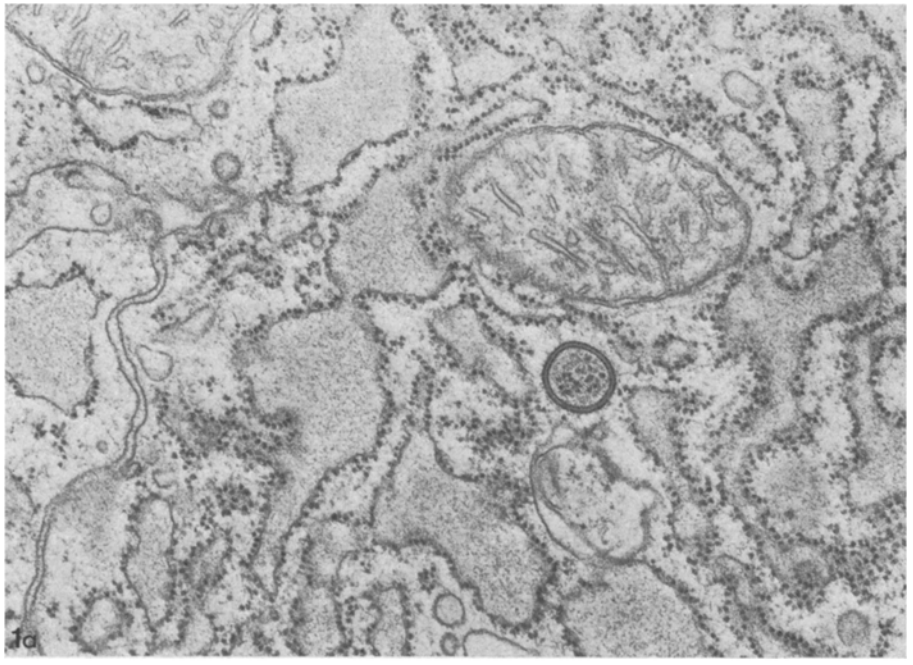
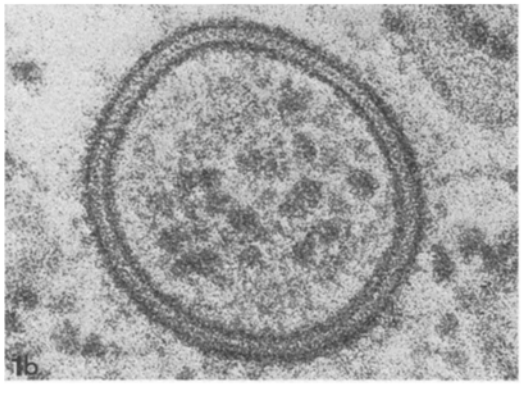


Fig. 1. *a* A spherical membrane-bound body is located in the cytoplasm of an osteoblast. A mitochondrion and cisternae of the rough-surfaced endoplasmic reticulum are seen in the vicinity of the inclusion body (× 40,000). *b* The intracytoplasmic body shown in *a* at a higher magnification. The granular matrix and the pentalaminar limiting membrane of the body are clearly visible (×200,000).



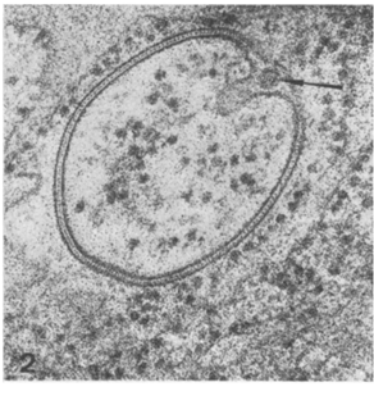


Fig. 2. The innermost leaflet of the pentalaminar limiting membrane of this oval-shaped inclusion body is continuous with a saccule containing a knob-like structure (arrow; ×80,000).

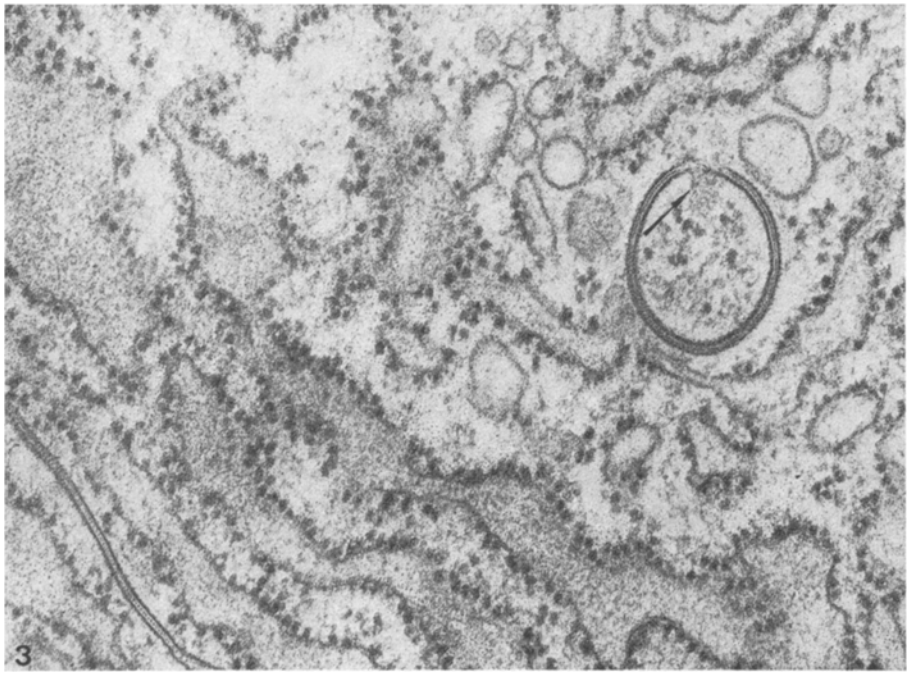


Fig. 3. Portion of an osteoblast containing a membrane-bound body. The innermost layer of the pentalaminar body membrane appears to be continuous with a saccular structure (arrow) obscured by the tangential plane of the section. An intercellular gap junction is to be seen in the lower left corner of the picture (×80,000).

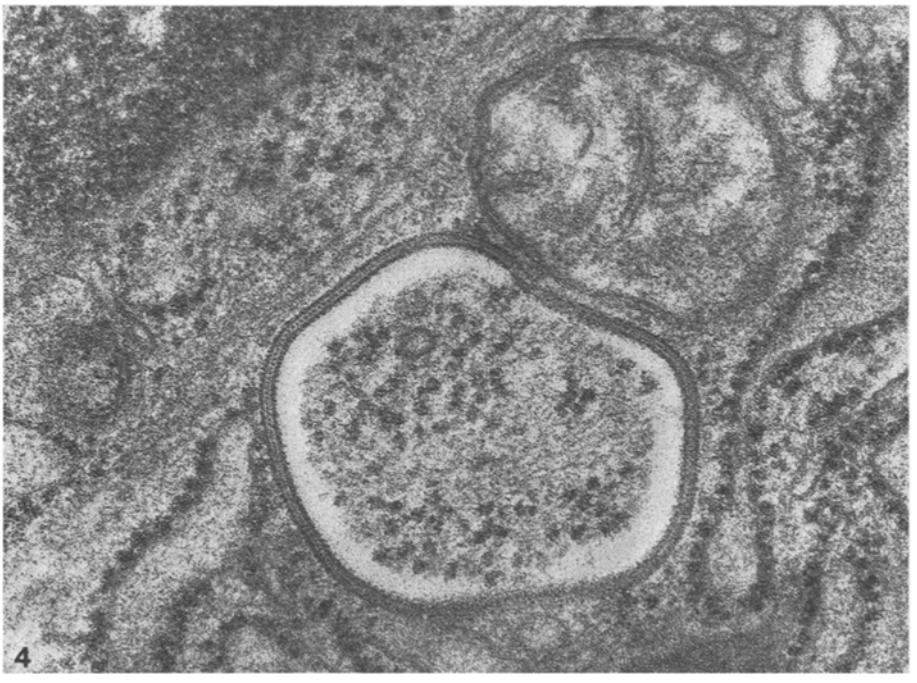


Fig. 4. An intracytoplasmic inclusion body is found in close contact with a mitochondrion and in the vicinity of the nucleus, a part of which is seen in the upper left corner of the picture. The granular core of the body is separated from the pentalaminar limiting body membrane by an electron-lucent halo (×120,000).

structure was found located near the outermost layer of the body membrane (figure 2). The intermediate dotted layer of the body membrane appeared to be discontinuous at sites of saccule location (figures 2 and 3).

The spherical membrane-bound bodies found in the metaphyseal trabecular bone tissue of the tibia of mice occurred mainly in osteoblasts and occasionally in preosteoblasts. They were not observed in capillary endothelial cells, osteoclasts and osteocytes. All bodies were located freely in the cytoplasmic matrix of cells. Continuities of the limiting body membrane with the limiting membrane of the cell within which a body was located were never detected.

Discussion. The bodies described in the present study are similar in their ultrastructure to those which several authors have found in cells from various normal or neoplastic tissues of hamsters[6], rats[8], rabbits[3,7], sheep[5] and men[2,4]. The structures have been observed in luteal cells of the ovary[2], cells of the membrana granulosa of the ovarian follicle[3], cultured cells from a small-cell carcinoma of the adrenal cortex[4], keratinizing cells of the wool follicle[5], cells of estrogen-induced renal adenocarcinomas[6], cells of vitamin A acid-treated keratoacanthomas[7] and periosteal cells of normal and lathyritic femurs[8]. Such inclusion bodies have not, to our knowledge, yet been reported as occurring in bone cells from the proximal tibial metaphysis of normal mice.

It has been presumed that the spherical structures might originate from intercellular gap junctions by bulging of such junctions into the cytoplasm of cells and by pinching off of the invaginations from the cell membranes[2,3,5,6]. According to this postulated development, a so-called annular gap junction must contain a portion of the cytoplasm from an adjacent cell[2,3,5]. The functional significance of such a cell-to-cell transfer of cytoplasmic components remains obscure at present.

Since the greatest number of intercellular gap junctions are found between osteoblasts in active bone formation[11], it is speculated that the so-called annular gap junctions, which we found predominantly in apparently protein-synthesizing osteoblasts, may also be somewhat associated with osteogenesis. In this respect, it should be emphasized that the so-called annular gap junctions present in metaphyseal bone cells contained mainly granules resembling ribosomes on which protein synthesis normally takes place.

Turnover of 5-hydroxydopamine in adrenergic nerves

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Summary. In adrenergic nerve endings of the guinea-pig vas deferens the population of small granular vesicles increases from 19% in control animals to 80–90% 1–3 h after the administration of 5-hydroxydopamine, and gradually declines to control values in 10 days. Large granular vesicles were also loaded but the loss of enhanced granulation was more rapid than in the small granular vesicles.

The osmiophilic false-transmitter 5-hydroxydopamine (5-OHDA) has been used as a specific electronmicroscopical marker for adrenergic nerves[2–6], and to investigate the storage of transmitter within the adrenergic neurone[7–8]. In the present experiments the time-course of the uptake and loss of 5-OHDA into synaptic vesicles of adrenergic nerves innervating the vas deferens has been studied.

Adult male guinea-pigs weighing 400–460 g were given 5-OHDA HCl (Sigma, 170 mg/kg in 1 ml isotonic saline