Ultrastructural Observations on the Embryogenesis of Islet Cells in the Rat Pancreas *in Vitro*[1] (35283)

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(Introduced by G. W. Gander)

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The embryogenesis of pancreatic islets has been extensively studied at the light microscopic level (1–3). As a result of these studies most investigators have concluded that the islet cells arise from ductual or tubular epithelium. Although the possibility that islet cells might also arise as a result of acinar to islet transformation has been considered, this concept has not received general acceptance (3, 4).

The present paper presents ultrastructural evidence which implicates both acinar and ductal elements in the embryogenesis of rat pancreatic islets *in vitro*.

Materials and Methods. The animals used in this study were rats of the Sprague-Dawley strain. The age of a litter of embryos was reckoned from the moment of a witnessed mating.

Developing pancreatic tissue was explanted at 17.5 days gestation. This gestational age was chosen because it is known to show developing ducts, acini and islets (2). The watch glass method of organ culture was used. The particulars of the culture technique have been previously described (2).

Multiple tissue samples for electron microscopy were taken from explants sacrificed after periods of 1 through 4 days *in vitro*. The tissues were fixed immediately in 6.25% buffered glutaraldehyde, postfixed in 1.0% buffered osmic acid, and then processed for electron microscopy. An RCA emu 3 G electron microscope was used.

Results. The normal acinar cells in the embryonic rat pancreas contained empty vesicles of the approximate size of zymogen granules and also immature and more nearly mature zymogen granules (5) (Fig. 1). The normal islet cells in the embryonic rat pancreas contained mainly beta-type secretory granules which are characterized in the rat (5) by an osmiophilic core and a wide halo between the core and the secretory granule membrane (Fig. 2). Some cells with secretory granules characteristic of islet cells were seen within the wall of tubules or small ducts (Fig. 3). Further, in tissues sampled from the 2 and 3 day explant stages but not before or after, an occasional individual cell or group of cells was seen containing both acinar cell zymogen granules and islet cell beta-granules (Figs. 4–6). In most instances these acinar-islet cells contained a relatively dense cytoplasm with ovoid, pleomorphic mitochondria, which have been thought to be a feature of cells that are less well differentiated (6), and an abundance of ribonucleoprotein particles.

Discussion. The presence of islet cells within the wall of small ducts or tubules suggests that they might be arising there. This is in keeping with the light microscopic findings in the rat and the generally accepted concept that islet cells arise from ductal epithelium (1–3). Furthermore, ultrastructural findings in this investigation of the embryonic rat pancreas agree with the ultrastructural findings of Munger (6) who studied the embryogenesis of mouse islet cells. Unfortunately, this must remain as circumstantial evidence to suggest the formation of islet cells from ductal epithelium until more concrete findings are recorded. Such findings might come from the identification of ductal-islet transition forms (which incidentally were not obvious to us at this time probably because of the similarities in the organelles of

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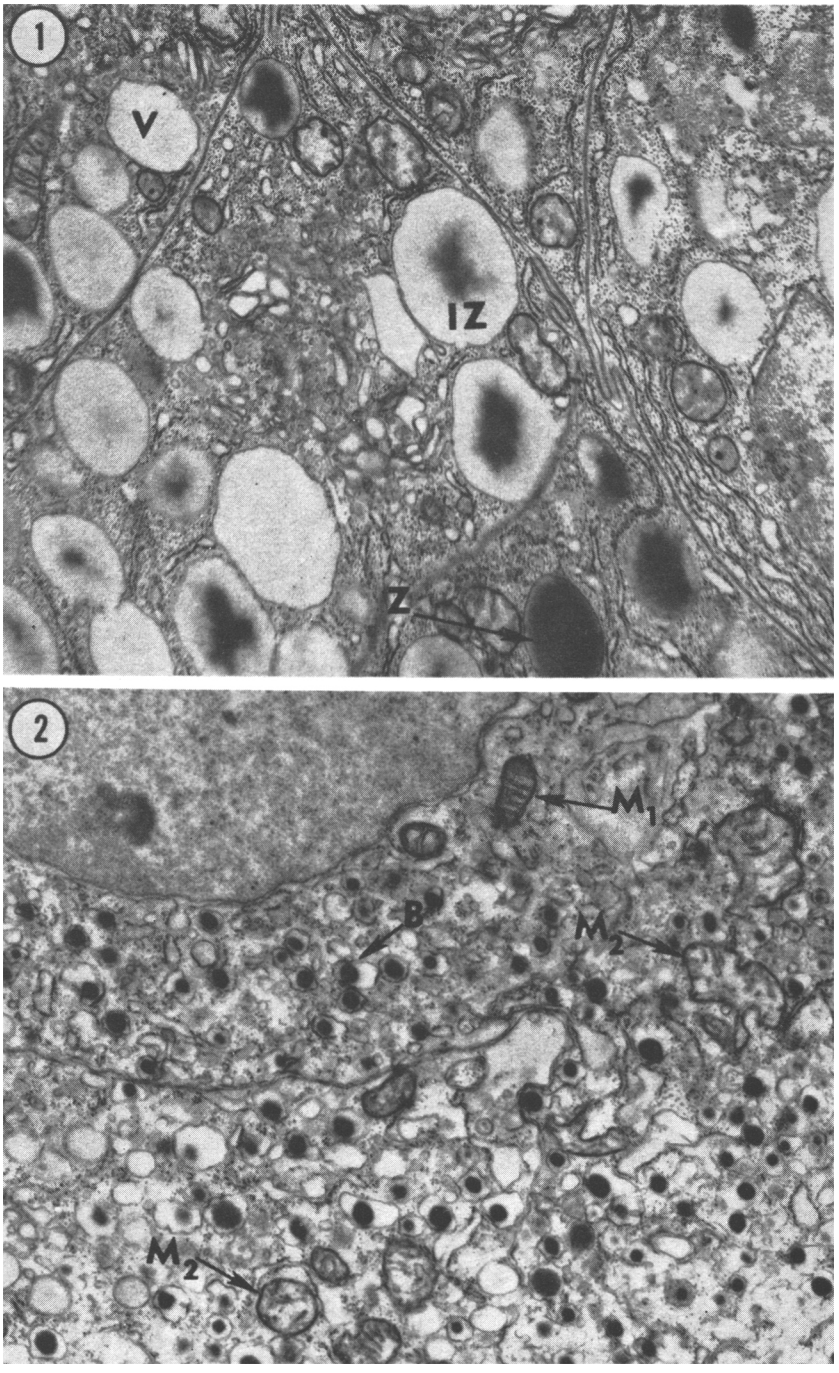

FIG. 1. Acinar cells in embryonic rat pancreas with empty vesicles (V) and also immature (IZ) and more nearly mature zymogen granules (arrow Z). Two-day explant; ×11,100.

FIG. 2. Islet cells with beta-type secretory granules (arrow B). Rod shaped (arrow M_1) and ovoid, plenomorphic (arrow M_2) mitochondria are present. Two-day explant; ×16,800.

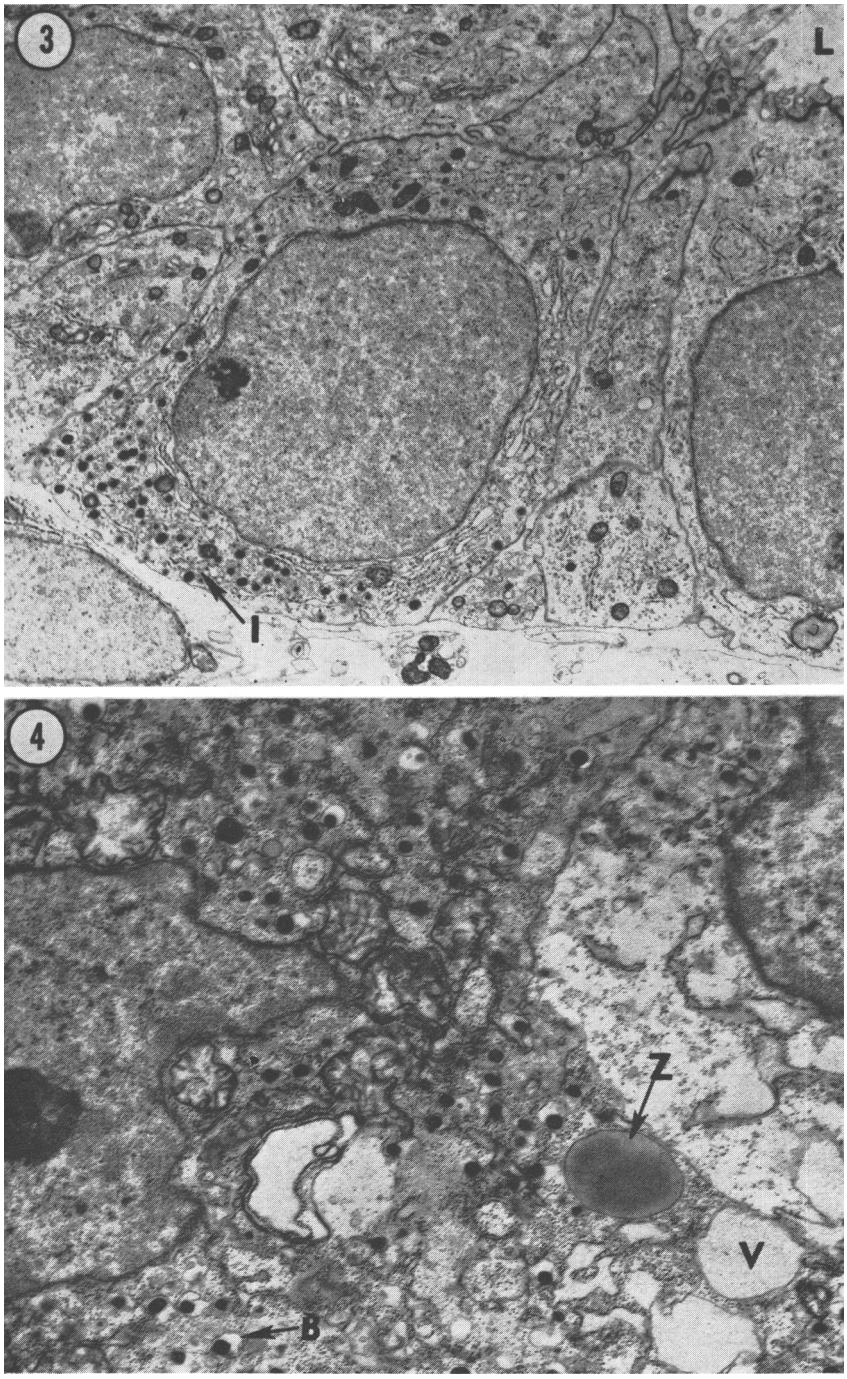

FIG. 3. Islet cell (arrow I) with secretory granules arising within wall of tubule. Lumen (L) is in upper right. One-day explant; ×5,600.

FIG. 4. Acinar-islet cell with dense cytoplasm, zymogen granule (arrow Z), large vesicles (V), beta-type islet cell granules (arrow B) and ovoid, pleomorphic mitochondria. Two-day explant; ×10,800.

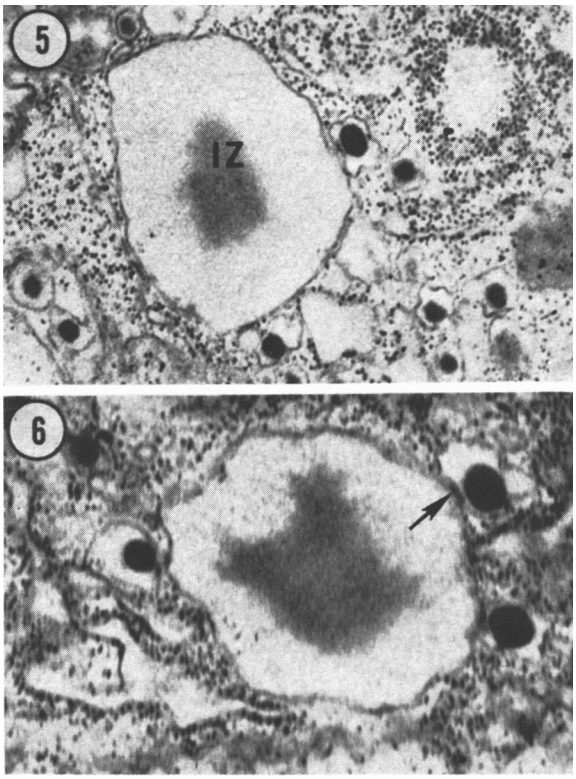

FIG. 5. Portion of acinar-islet cell showing presence of immature zymogen granule (IZ) and beta-type secretory granules. Two-day explant; ×25,500.

FIG. 6. Portion of another acinar-islet cell intending to show immediate juxtaposition of immature zymogen granule's membrane and that of the beta-type secretory granule (arrow) indicating that the two different secretory products are within the same cell. Two-day explant; ×28,000.

these two cell types) or from the identification of transformation by applying a "tag" to undifferentiated ductal epithelium and finding it in the islet cells.

Unlike Munger who did not see any acinar-islet type cells in the embryonic mouse pancreas, we were able to find such cells in our *in vitro* model. However, their presence in the embryonic rat pancreas is not surprising since their formation can be induced in the postembryonic rat pancreas (5) which is theoretically less competent.

The acinar-islet cell is of biological significance because it presents an intermediate or transition form of cell that suggests acinar-islet transformations. Its existence also raises some question about the hypothesis held by some that islet cells cannot arise from acinar epithelium (3, 7).

Current studies are underway to ascertain whether such acinar-islet cells are present in the embryonic rat pancreas *in vivo* at a gestational age relatively comparable (8) to the *in vitro* stages used in this study.

Summary. Ultrastructural studies on the embryonic rat pancreas grown in organ culture show the presence of islet cells within the wall of small ducts or tubules and acinar-islet cell types. These findings provide morphologic evidence which implicates both acinar and ductal elements in the embryogenesis of rat islet cells.

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