INTERMEDIATE CELLS OF THE PANCREAS

I. ULTRASTRUCTURAL CHARACTERIZATION

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SUMMARY

The normal existence of cells in the pancreas with a structure intermediate between those of exocrine and endocrine cell types has long been a matter of dispute. The present study shows that, based upon morphological criteria, such intermediate cells are present in both the endocrine and exocrine tissues of the normal pancreas of the rat, guinea-pig, rhesus monkey, goat, chicken and frog. There is a tendency for intermediate cells to occur most frequently in the frog, where exocrine and endocrine cells are intermingled, and least frequently in higher species such as the rat, guinea-pig, monkey and goat where the endocrine cells are localized in discrete islets. Their occurrence in the chick appears to lie between these 2 extremes. The existence of intermediate cell types has been attributed to a 'transformation' of one form of specialized cell in the pancreas into another in response to a metabolic demand. However, the widespread occurrence of intermediate cells in the normal pancreas suggests that they represent, ab initio, a distinct category of cell, the existence of which poses interesting questions concerning the genetic control of their specialized functions and of developmental processes in the pancreas. Moreover, intermediate cells, such as acinar cells containing endocrine $\beta$-granules might serve as a source of insulin additional or alternative to that provided by cells that are wholly endocrine in character.

INTRODUCTION

The occurrence of cells intermediate in morphology between that of exocrine and endocrine cells has frequently been described in the pancreas of animals subjected to metabolic stress of several kinds (see p. 456), but their existence in the normal pancreas has remained a controversial question. In the past, with accurate identification being hampered by the limited resolution of the light microscope and by the apparent rarity of the phenomenon in higher vertebrates there have been arguments both for (Mankowski, 1902; Laguèse, 1905; Saguchi, 1920; Sergeyeva, 1940; Hughes, 1947) and against (Lane, 1907; Bensley, 1914; Gomori, 1941) their normal occurrence. In recent years, new support for their existence has come from the description of cells with both exocrine and endocrine ultrastructural characteristics which have been observed in a variety of species (Table 1). However, scepticism has remained in some quarters, with the reported electron-microscopic demonstration of intermediate cells being attributed to structural artifacts occurring during tissue preparation (Marx, Schmidt, Herrmann & Goberna, 1970).

Resolution of this controversy is important for a number of reasons. First, if intermediate cells occur in the normal pancreas, then their greater prominence in abnormal states is more likely to reflect the existence of a population of cells capable of more than
Table 1. Intermediate cells previously described in the pancreas of some normal vertebrates

Only those authors who have documented their observations with electron micrographs are quoted.

<table>
<thead>
<tr>
<th>Species</th>
<th>Intermediate cell types</th>
<th>Authors</th>
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<tbody>
<tr>
<td>Toad (Bufo vulgaris formosus)</td>
<td>Mixed</td>
<td>Kobayashi (1966)</td>
</tr>
<tr>
<td>Frog (Rana esculenta)</td>
<td>Mixed</td>
<td>Geuze (1970)</td>
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<tr>
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<td>Rhoten (1970)</td>
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<tr>
<td>Duck</td>
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<td>Björkman &amp; Hellman (1964)</td>
</tr>
<tr>
<td>Chick</td>
<td>α-Acinar; β-acinar</td>
<td>Mikami &amp; Mutoh (1971)</td>
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<tr>
<td>Rat</td>
<td>Acinar-β</td>
<td>Herman et al. (1964)</td>
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<tr>
<td>Cat</td>
<td>Acinar-α</td>
<td>Brown &amp; Still (1970)</td>
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<td>Saimiri monkey</td>
<td>α-β</td>
<td>Winborn (1963)</td>
</tr>
<tr>
<td>Man</td>
<td>Acinar-α</td>
<td>Zagury, de Brux, Ancla &amp; Leger (1961)</td>
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one specialized function, rather than the 'transformation' of one type of specialized cell into another. Secondly, intermediate cell types as a species of cell sui generis would be of considerable biological interest, since they pose the problem of how multiple specialized functions in one cell are genetically regulated, and how different proteins synthesized in the endoplasmic reticulum of one cell are packaged into separate and characteristic granule types. In addition, intermediate cells such as acinar cells containing β-granules may possibly provide an alternative or additional store of insulin, perhaps for use in time of need.

Morphological evidence is now presented for the normal occurrence of intermediate cell types in both the exocrine and endocrine pancreas of the rat, guinea-pig, monkey, goat, chicken and frog. These observations confirm the electron-microscopic findings of others, provide additional data in some species (rat, monkey, chicken and frog) and extend them to the guinea-pig and the goat. They also form the basis of confirmatory studies of intermediate cell structure and function to be presented later.

MATERIALS AND METHODS

All materials used for electron microscopy were obtained from TAAB Laboratories, 52 Kidmore End Road, Emmer Green, Reading, England.

The tissue surveyed was obtained from 17 albino Wistar rats (Courtauld Institute inbred strain) of which 12 were adults (200–300 g), 4 were suckling rats (20–30 g) and one was a pregnant female after 18 days gestation, and from 4 adult guinea-pigs (500–1000 g), 4 chickens, 4 hibernating frogs (Rana pipiens), 1 goat and 1 rhesus monkey. The rats and chickens had
been fed on a mixture of equal weights of powdered Oxoid 41B diet (W. Lillico & Son, Wonham Mill, Betchworth, Surrey) and heat-processed soya flour (Soya Foods Ltd., 30 Mincing Lane, London, E.C. 3). The small animals were killed by cervical dislocation and the goat and monkey by an overdose of an anaesthetic agent. In each case, a piece of tissue was removed from the tail of the pancreas immediately after death and diced into small cubes and immersed in fixative at room temperature. Because the nature of the fixative may influence granule morphology (Munger, Caramia & Lacy, 1965), various fixation schedules were tried. The best preservation was obtained with a mixture of 2 % glutaraldehyde and 3 % formaldehyde buffered at pH 7.2 with 67 mM cacodylate buffer (after Karnovsky, 1965). After primary fixation in this aldehyde mixture, the tissue was postfixed in 1 % osmium tetroxide buffered at pH 7.2 with 0.1 M phosphate buffer. The duration of primary and secondary fixation is not critical. However, primary fixation of less than 2 h gave inferior preservation of fine structure, while osmication for more than 8 h caused extraction artifacts which included the destruction of zymogen granules (see Palade, 1956a), although membrane contrast was enhanced. After fixation, the tissue was dehydrated in graded ethanols and passed via propylene oxide into Epon 812 (Luft, 1961).

Exocrine intermediate cells are usually found at junctions of acinar tissue with islets, or at those of ducts and islets. Intermediate cells in these areas can usually be recognized by staining 0.25-4μm sections with toluidine blue and examining them by light microscopy. Intermediate cells in islets are located in the same way. Silver-grey sections were then cut with glass or diamond knives from appropriate regions of the block, collected on uncoated 50-μm TAAB grids, stained with uranyl acetate (1 min) and lead citrate (3 min) and examined in a Philips EM 200 electron microscope at 60 kV.

Intermediate cells of pancreas

Nomenclature and classification of intermediate cells

The exocrine and islet cells of the pancreas have been extensively studied in a variety of species and the identity of the various principal storage granules has been firmly established. In the exocrine pancreas, the zymogen granule has been characterized as the storage and secretory organelle of the digestive enzymes synthesized by the acinar cell (Keller & Cohen, 1961). In the endocrine pancreas, the nature of the 2 predominant granule types (α and β) has been established in a wide variety of species through correlated light- and electron-microscopic techniques (Lacy, 1957; Bencosme & Pease, 1958; Winborn, 1963; Björkman & Hellman, 1964; Herman, Sato & Fitzgerald, 1964; Sato, Herman & Fitzgerald, 1966; Munger et al. 1965; Kobayashi, 1966; Lange, 1967; Like, 1967), by immunochemical studies using antibodies to insulin or glucagon (Lacy, 1959; Baum, Simons, Unger & Madison, 1962; Lange, 1970; Bussolati, Capella, Vassallo & Solcia, 1971), by analysis of isolated islet secretory granules (Lindall, Bauer, Dixit & Lazarow, 1963; Greider, Howell & Lacy, 1969; Sorensen, Lindall & Lazarow, 1969) and by selective destruction of β-granules by alloxan (Williamson & Lacy, 1959) or Streptozotocin (Arison et al. 1967). While the content of the α-granules of the endocrine pancreas has not been firmly established, it appears from recent immunohistochemical studies that they contain gastrin (Lomský, Lange & Vortel, 1969; Greider & McGuigan, 1971).

The nomenclature used to describe the ultrastructure of intermediate cells in the pancreas has varied a great deal, e.g. acinar-islet cells (Gusek & Kracht, 1959; Herman et al. 1964), intergrade cells (Winborn, 1963), mixed or intermediate cells (Pictet et al. 1967), extrainsular cells (Kobayashi & Fujita, 1969), exocrine-endocrine cell types (Geuze, 1970) and neisidiolecta (Brown & Still, 1970). It is probable, too, that 'transitional' (Sergeyeva, 1940) and 'amphiphil' (Epple, 1966) cells identified with the light microscope represent intermediate cells, and that 'X' cells (Maniowski, 1902) may also have done so on occasions.

To establish a consistent and unambiguous nomenclature, the following system is suggested and is used here. Each cell clearly showing the ultrastructural characteristics of more than one pancreatic cell type is termed an intermediate cell. The name of the intermediate cell is derived first from its location in exocrine (acinar-; duct-) or endocrine (α-; β-; δ-) tissue, and then from the nature of the participating granule types. Thus, an acinar cell containing β-granules is termed an acinar-β intermediate cell and an α-cell containing zymogen granules is an α-acinar intermediate cell. This system permits a precise morphological classification of intermediate cell types.

The criteria for classification of an acinar cell used here are the presence of zymogen granules
and/or the very extensive rough-surfaced endoplasmic reticulum characteristic of this cell type. The characterization of endocrine cells depended upon the recognition of endocrine granules morphologically typical of the species being studied. This presented no difficulty except in the case of the frog, where the granules cannot easily be recognized as α-, β-, or δ-(cf. Kobayashi 1966; Lange, 1967, 1970). With the goat, in which the endocrine granules of the pancreas have not been described before, they were identified by their similarity to the endocrine granules of other species (see p. 454).

The cytoplasm of duct cells is more difficult to characterize as it has few distinctive ultrastructural features. For this reason, only those intermediate cells directly continuous with a lumen lined by normal duct cells are considered here. Endocrine type cells forming part of a duct but not contributing to the luminal margin are also excluded from consideration.

RESULTS

The intermediate cell types encountered in the exocrine and endocrine pancreas of the 6 species studied are summarized in Table 2. They are described in detail below.

Rat

The predominant intermediate cell type is the acinar-β cell (Fig. 1). In general, the zymogen and β-granules are mixed to varying degrees, although the β-granules are sometimes segregated into purely β-granule areas. The variable occurrence of other cytoplasmic organelles, such as mitochondria, from the contributing cell types (Herman et al. 1964) has been confirmed. Although usually seen close to the plasma membrane in other areas, β-granules in these intermediate cells are sometimes mixed with the zymogen granules in the cell apex (Fig. 2). The zymogen granules rarely approach other parts of the plasma membrane of the acinar-β cell.

Evidence that crinophagy (Smith & Farquhar, 1966) occurs in acinar-β intermediate cells is provided by the occasional presence of membrane-bound vacuoles containing several endocrine granules (Fig. 1).

Acinar-α (Fig. 3) and acinar-δ (Fig. 4) cells occur much less frequently in the rat than the acinar-β type. The acinar-α, β cell seems to be of 2 types. One type was found between islet and acinar cells that occurred together in small lobules. This type was difficult to classify as it was not always possible to identify the primary cell type. However, the second type of acinar-α, β cell was quite clearly part of the exocrine system (Fig. 5).

A fairly common feature of the acinar-intermediate cells, not usually seen in the islet- or duct-intermediates is the presence of intracellular membranous partitions (Fig. 1). These are frequently in continuity with the plasma membrane, and extend into the cell to a variable degree. They have previously been described in the rat by Herman et al. (1964) and by Marx et al. (1970). Another common feature of acinar-intermediate cells is the occurrence of an unusual papillary type of rough-surfaced endoplasmic reticulum (Fig. 5).

In the islets, zymogen granules were occasionally observed in β-cells (Fig. 6) where usually, but not invariably, they are associated with a more extensive rough-surfaced endoplasmic reticulum than is found in normal β-cells.

In duct-β cells (Fig. 7), the endocrine granules are always separated from the
Intermediate cells observed in this study of the pancreas of some normal vertebrates

<table>
<thead>
<tr>
<th>Species</th>
<th>Intermediate cell types</th>
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<tr>
<td>Rat</td>
<td>Acinar-α; acinar-β; acinar-δ; acinar-α, β; acinar-δ, δ</td>
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<tr>
<td></td>
<td>β-Acinar</td>
</tr>
<tr>
<td></td>
<td>Duct-β</td>
</tr>
<tr>
<td>Guinea-pig</td>
<td>Acinar-α</td>
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<tr>
<td></td>
<td>β-Acinar</td>
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<tr>
<td></td>
<td>β-α, Acinar</td>
</tr>
<tr>
<td>Rhesus monkey</td>
<td>Acinar-α; α-Acinar; β-acinar; α, β-acinar</td>
</tr>
<tr>
<td></td>
<td>α-β</td>
</tr>
<tr>
<td>Goat</td>
<td>Acinar-α; acinar-β</td>
</tr>
<tr>
<td></td>
<td>α, β-Acinar</td>
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<tr>
<td></td>
<td>α-β</td>
</tr>
<tr>
<td>Chick</td>
<td>Acinar-α; acinar-β</td>
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<td></td>
<td>α-acinar</td>
</tr>
<tr>
<td>Frog</td>
<td>Acinar-I; acinar-III; acinar-I, II; acinar-I, II, III, IV</td>
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<td>I-Acinar</td>
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Luminal border of the cell by a narrow zone of cytoplasm. However, the granules are usually close to the plasma membrane in other regions, suggesting that they are discharged into the interstitial space rather than into the duct lumen.

**Guinea-pig**

The predominant intermediate cell type in this species is the acinar-α cell (Fig. 8). The α-granules are easily distinguished from the intracisternal granules of the endoplasmic reticulum by the close-fitting ribosome-free limiting membrane of the former, and by their usual proximity to the plasma membrane. Acinar-α cells occur less frequently than in the rat, although the general features of this cell type are similar in both species.

In the islets there was a striking gradation in the morphology of some cells from that of an obvious β-cell (Lacy, 1957; Caramia, Munger & Lacy, 1965) to others also presenting a well-developed rough-surfaced endoplasmic reticulum complete with intracisternal granules characteristic of the acinar cell (Palade, 19566) (Fig. 9).

**Rhesus monkey**

Intermediate cells are much more conspicuous in the islets than in the exocrine tissue. Most frequently observed were α-acinar cells (Fig. 10), although α, β-acinar (Fig. 11), α-β and β-acinar cells were also seen. Acinar-α intermediate cells were also occasionally observed (Fig. 11).
Goat

Because the endocrine granules of the goat pancreas have not previously been described they have been identified here by their similarity to the endocrine granules of other species: \(\alpha\)-granule – a compact round granule with a close-fitting membrane and a thin less-dense halo surrounding a very dense core (Fig. 12); \(\beta\)-granule – the most common type, this granule is of comparable density to that of the zymogen granule in the acinar cell. However, it is slightly larger than the \(\alpha\)-granule and its content often completely fills the surrounding membrane (Fig. 13). The mature granules are interspersed with larger, less-dense and presumably immature \(\beta\)-granules.

Acinar-\(\alpha\) (Fig. 14) and acinar-\(\beta\) (Fig. 15) intermediate cells were found in the exocrine tissue of the goat pancreas. In the islets, \(\alpha\)-\(\beta\) (Fig. 16) and \(\alpha\), \(\beta\)-acinar cells were occasionally seen.

Chick

Acinar-\(\alpha\) (Fig. 17), acinar-\(\beta\) (Fig. 18), acinar-\(\delta\) (Fig. 19) and \(\alpha\)-acinar (Fig. 20) intermediate cells were observed in the chick pancreas. The admixture of exocrine and endocrine elements in cells at the periphery of islets was sometimes quite extensive and, as in the rat, the accurate identification of the primary component of the intermediate cell was not always possible.

Frog

In addition to zymogen granules, 4 other distinct granule types are present in the hibernating frog pancreas. They will be designated here by Roman numerals (cf. Kobayashi, 1966; Lange, 1967, 1970). Type I (Fig. 21) is of variable size, round, compact and dense. It sometimes has a narrow, less-dense halo. These granules closely resemble \(\alpha\)-granules in other species. Type II (Fig. 22) is a much less-dense granule which is sometimes very irregular in appearance. It generally fits loosely into its membranous sac. This granule best corresponds to the \(\beta\)-granule in the rat. Type III (Fig. 23) granules are also compact and of variable size and shape. They are similar in density to the Type II granule but always fit snugly into their surrounding membrane sac. These granules are most like the \(\delta\)-granules of the islets of other species, especially the chick. Type IV granules (Fig. 24) are usually smaller than all the above granules. They are intermediate in density between Types I and III, but vary considerably in shape. They seem to correspond most closely to catecholamine storage granules found in other species (Yates, 1964).

The occurrence of intermediate cells in the frog is common. Varying mixtures of all the granule types were observed in acinar cells (Figs. 25, 26). The occasional occurrence of zymogen granules in predominantly endocrine cells was also noticed.

The membranous partitions described above in intermediate cells of the rat were also observed in the frog and the guinea-pig. Their occurrence seems to be common, for they have previously been described in the frog by Kobayashi (1966), in the mouse (de Hoyos-Guevara, 1970) and in the chicken (Mikami & Mutoh, 1971).
DISCUSSION

The identity of storage granules in intermediate cells

The morphological constancy of storage granule types amongst the pancreatic parenchymal cells, each representing the specific end-product of a specialized cell function, has been utilized for the identification of intermediate cells.

The zymogen granule has a distinctive and constant appearance in all species. When it occurs in islet intermediate cells the only known organelle of similar size from which it needs to be distinguished is the autophagic vacuole. However, autophagic vacuoles in islet cells are characterized by a heterogeneity of content and shape not seen in zymogen granules (see Lazarus, Volk & Barden, 1966; Orci et al. 1968; Creutzfeld et al. 1969; Boquist, 1970; Like & Chick, 1970). In general, there is also a striking ultrastructural similarity between the same hormone granule from one species to the next, with the notable exception of the $\beta$-granule. The major difference between $\alpha$- and $\delta$-granules from a variety of species is in their size rather than form. The morphology of the $\beta$-granule varies considerably amongst different species, but the $\beta$-granules in intermediate cells are always characteristic of the species under study.

Frequency of occurrence and distribution of intermediate cells in the different species

The common embryological origin of exocrine pancreas from a diverticulum of the midgut endoderm (Diamare, 1905; Bensley, 1911) results in the concomitant adjacent development of 2 highly specialized tissues, each of which contains 2 or more distinct cell types. In most species the exocrine and endocrine components are intermixed to varying degrees and one of the striking conclusions to emerge from the present investigation is that the frequency of occurrence of intermediate cells is generally inversely proportional to the degree of separation of the exocrine and endocrine elements. For example, intermediate cells were most frequently found in the frog in which exocrine and endocrine elements are normally intermingled and least often in the rat, guinea-pig, goat and monkey in which discrete islets are the rule. In the chick, where the $\alpha$- and $\beta$-cells are segregated into separate groups which are sometimes contiguous, the frequency of intermediate cell occurrence is between that of the frog and the higher vertebrates studied. This trend finds support in the light-microscope study by Epple (1966) using differential cytochemical techniques. He noted that intermediate ‘amphiphil’ cells occurred in the shark (Scylliorhinus canicula) and in the toad (Bufo bufo) but he was unable to detect their occurrence in normal birds and mammals. The occurrence of intermediate cells in the frog (Rana esculenta) and toad (Bufo vulgaris formosus) has been reported at an ultrastructural level by Kobayashi (1966) and Geuze (1970). Our observations on the chick pancreas parallel quite closely those of Björkman & Hellman (1964) in the duck. These authors reported the fairly frequent occurrence of intermediate cells at the periphery of the segregated islets (i.e. separate $\alpha$-cell and $\beta$-cell regions) and the frequent occurrence of zymogen-like granules in the $\alpha$-cells. It now appears that intermediate cells are situated between cells of the types contributing to the hybrid. Thus, in those species with distinct islets, intermediate
cells are found in the exocrine tissue immediately adjacent to the islets. The location of islet intermediate cells may appear more variable as the precise relationships to other cell types of any cell within an islet will be more varied than for cells in the exocrine pancreas. This is because the majority of islets are between 50 and 150 μm in diameter regardless of the species (Henderson, 1969) and are of variable 3-dimensional conformation (Goldstein & Davis, 1968). This means that many islet cells are adjacent to the acinar cell system or to endocrine cells of a different character.

The origin of the intermediate cell

The observations reported here establish the existence of intermediate cells in the normal pancreas of a variety of species. The currently accepted theory for their origin is that they arise by ‘transformation’ of differentiated pancreatic cells in response to a metabolic disturbance such as occurs in alloxan diabetes (Hughes, 1947; Johnson, 1950; House, 1958; Patent & Alfert, 1967), hereditary diabetes syndrome in the mouse (Pictet & Gonet, 1966; Pictet et al. 1967; Shino & Iwatsuka, 1970), after chronic glucose infusion (Woerner, 1938), ethionine administration (Herman, Sato & Fitzgerald, 1963) or after partial pancreatectomy (Marx et al. 1970). However, under conditions of normal homeostasis, one would expect the available hormone reserves of the endocrine cells in the pancreas to cope adequately with the physiological demands, thus making unnecessary the requirement for such a transformation. A more plausible explanation for the widespread though variable occurrence of intermediate cells in the normal pancreas is that they are present in the organ ab initio. Moreover, it seems highly improbable that islet-intermediate cells (i.e. islet cells containing zymogen granules), or acinar cells containing simultaneously the storage granules of the metabolically antagonistic hormones, insulin and glucagon, could arise by cell transformation in response to any conceivable metabolic demand. The response of pre-existing intermediate cells to altered homeostasis may account for the frequency with which they have been reported in abnormal states (see above).

An alternative hypothesis that intermediate cells arise from cell fusion at some stage of development needs to be considered. The occurrence of membranous partitions in some intermediate cells (Herman et al. 1964) has been suggested as favouring a cell fusion hypothesis (Kobayashi, 1966) but our survey shows that membranous partitions are confined to acinar intermediate cells and have not been observed in islet or duct intermediate types. Moreover, if cell fusion were the major cause of intermediate cell formation it is difficult to explain why the phenomenon seems to have been detected almost exclusively in the pancreas (cf. Benedeczky & Lapis, 1970).

The propensity of cells at the interface of different pancreatic cell types to have a hybrid structure and presumably overlapping functions supports the generally accepted hypothesis of a common primitive foregut origin for all the pancreatic parenchymal cells and argues against a neuroectodermal derivation for the islet cells, in spite of certain histochemical staining characteristics they share with proven neuroectodermal derivatives (Pearse, 1969). It would also seem from this study of intermediate cell types that the specificity or degree of differentiation of function in the pancreas is determined principally by the position of the primitive precursor cells.
Intermediate cells of pancreas

within the developing organ (see Wolpert, 1969), their ultimate differentiation possibly being determined in part by their proximity to some focal stimulus such as a nutritional gradient created by the more vascular islet areas (see Crick, 1970). The reported morphological differences between the peri-islet acini and those further removed from the islets (Hellman, Wallgren & Peterson, 1962; Kramer & Tan, 1968) is also consistent with such a possibility.

We thank the Fleming Memorial Fund for Medical Research for providing a Philips EM 200 electron microscope, the Cancer Research Campaign for supporting one of us (C.J.B.) and the Medical Research Council for their generous support of this project.

REFERENCES


Intermediate cells of pancreas


Figs. 1–26 are electron micrographs of ultrathin sections of intermediate cells in the pancreas of several species. All are stained with uranyl acetate and lead citrate.

Fig. 1. Rat acinar-β cell. The field shows the typical admixture of endocrine (b) and exocrine (α) granules found in these cells. A crinophagic vacuole (crv) containing endocrine granules is present in the upper right corner. A membranous septum apparently continuous with the plasma membrane (pm) is indicated by arrows. × 34,000.

Fig. 2. Rat intermediate cell, predominantly acinar-β in character but also containing a few δ granules (d). Both zymogen (α) and β-granules (b) are situated close to the acinar lumen (l). Microvilli (mv) are normally abundant in the lumen formed by acinar-intermediate cells. A portion of the nucleus is seen at the lower left. × 25,000.
Intermediate cells of pancreas
Fig. 3. Part of a rat acinar-α cell showing α-granules (a) surrounded by an extensive rough-surfaced endoplasmic reticulum characteristic of the acinar cell. The arrows point to ribosome-free membranous septa. × 25,000.

Fig. 4. This field shows the coexistence of zymogen (z) and δ-granules (d) in the cytoplasm of a rat acinar-δ cell. × 25,000.

Fig. 5. Rat acinar-α, β cell. α- and β-granules (a, b) are predominant in the left half of the field and are separated from a group of zymogen granules (z) on the right by a tract of extensively convoluted rough-surfaced endoplasmic reticulum (rer). This is in striking contrast to the sparse rough endoplasmic reticulum typical of islet cells, seen in the predominantly endocrine part of this cell. Note the proximity of endocrine granules to the plasma membrane (pm) and basal lamina (bl) in the upper left of the field. × 19,000.
Intermediate cells of pancreas

3

4

5

pm

bl

a

b

l

r

rer

z

d

Fig. 6. Rat β-acinar cell showing numerous zymogen granules (z) and acinar type rough endoplasmic reticulum (rer) in a β-cell. Numerous β-granules (b) are seen throughout the field. × 10,500.

Fig. 7. Rat duct-β cell. Numerous β-granules (b) are close to the plasma membrane of the cell except on the luminal surface. An extensive Golgi system (g) is present in this cell. Profiles of cilia (c) and membrane-bound vesicles are seen in the duct lumen. × 12,000.
Intermediate cells of pancreas
Fig. 8. Guinea-pig acinar-α cell showing α-granules (a) adjacent to the plasma membrane (pm) and separated from zymogen granules (z) by the rough-surfaced endoplasmic reticulum (rer) containing intracisternal granules (ig) typical of this species. × 26000.

Fig. 9. Guinea-pig β-α, acinar cell showing β- and α-granules (b, a) in the lower half of the field, the upper half of which is occupied by rough-surfaced endoplasmic reticulum (rer) with intracisternal granules (ig) characteristic of the acinar cell in this species. × 26000.
Intermediate cells of pancreas
Fig. 10. α-acinar cell in the rhesus monkey showing zymogen granules (z) and extensive rough endoplasmic reticulum (rer) characteristic of the acinar cell, together with numerous α-granules (a). × 26,000.

Fig. 11. The left third of the field is occupied by part of an α, β-acinar cell. This contains a zymogen granule (z) in addition to both α- and β- endocrine granules (α, β). The acinar cell on the right contains α-granules (α) in addition to zymogen (z). The 2 cells are separated by an obliquely sectioned nerve bundle (nb). × 26,000.
Intermediate cells of pancreas
Figs. 12, 13. These show respectively α- and β-granules in goat islet cells. Note the relative absence of rough-surfaced endoplasmic reticulum in cells of these types. Fig. 12, × 37 500; Fig. 13, × 31 000.

Fig. 14. α-Granules (a) and extensive rough endoplasmic reticulum (rer) in a goat acinar cell. × 31 000.

Fig. 15. β-granules (b) and extensive rough endoplasmic reticulum (rer) in a goat acinar cell. × 36 000.

Fig. 16. A goat islet intermediate cell containing predominantly β-granules (b) and some α-granules (a). The sparseness of the rough endoplasmic reticulum is typical of the islet cells of this species. × 24 000.
Intermediate cells of pancreas
Fig. 17. An acinar-α cell in the chick. Several dense α-granules (a) and a zymogen granule (z) are present in the cytoplasm which possesses the extensive rough endoplasmic reticulum (rer) characteristic of the acinar cell in this species. The prominent Golgi apparatus (g) contains material with a density comparable to that of the zymogen granule. × 20000.

Fig. 18. A chick acinar-β cell. Zymogen granules (z) and extensive rough endoplasmic reticulum (rer) are accompanied by a number of β-granules (b) recognized by their characteristic inclusions. Granule d appears to resemble the chick δ-granule (see Fig. 19). × 20000.

Fig. 19. A chick acinar-δ cell showing zymogen granules (z) and a group of pleomorphic δ-granules (d), together with extensive tracts of rough endoplasmic reticulum (rer). The organelle above the central zymogen granule appears to be a small autophagic vacuole (av). × 25000.

Fig. 20. This chick α-acinar cell contains a typical zymogen granule (z), together with numerous denser α-granules (a). A centriole (ce) is present just below the centre of the field and a portion of a δ-cell (dc) in the islet is seen at the lower left. × 20000.
Fig. 21. A group of Type I granules in the frog pancreas, some of which resemble the α-granules of other species. × 30,000.

Fig. 22. Type II granules in the frog pancreas, some of which closely resemble β-granules in the rat pancreas. × 30,000.

Fig. 23. Numerous Type III granules fill this portion of a cell in the frog pancreas. They are larger and paler than Type I granules, pleomorphic in character and similar to δ-granules in the islets of other species, especially the chick. × 30,000.

Fig. 24. In the frog the Type IV granules are smaller than the others. They vary in shape but frequently have elongated profiles. The body with the appendage in the lower centre of the field is probably an autophagic vacuole (av). × 30,000.

Fig. 25. This cell in the frog pancreas, characterized as acinar by the presence of an extensive rough-surfaced endoplasmic reticulum (rer) (cf. Figs. 21–24) and a zymogen granule (z), also contains granules of Types I–IV. × 30,000.

Fig. 26. This acinar cell in the frog pancreas shows a group of Type III granules in addition to a number of zymogen granules (z). A Golgi complex (g) with 2 small condensing vacuoles (cv) is seen in the lower left of the field. × 22,500.
Intermediate cells of pancreas