Transmission microscopical investigation of the dark and light pancreatic acinar beta-cells of young-domesticated pig (Sus Suidae, Erxleben 1777)

Authors: M. Elghoul, R. Kandyle, K. Morsy, M. M.A. Abumandour

DOI: 10.5603/FM.a2021.0104
Article type: Original article
Submitted: 2021-08-23
Accepted: 2021-09-29
Published online: 2021-10-07

This article has been peer reviewed and published immediately upon acceptance. It is an open access article, which means that it can be downloaded, printed, and distributed freely, provided the work is properly cited. Articles in "Folia Morphologica" are listed in PubMed.
Transmission microscopical investigation of the dark and light pancreatic acinar beta-cells of young-domesticated pig (Sus Suidae, Erxleben 1777)

M. Elghoul et al., Pancrease of young-domesticated pig

M. Elghoul¹, R. Kandyle², K. Morsy³,⁴, M.M.A. Abumandour⁵

¹Department of Histology and Cytology, Faculty of Veterinary Medicine, Alexandria University, Egypt
²Zoology Department, Faculty of Science, Tanta University, Tanta, Egypt
³Biology Department, Faculty of Science, King Khalid University, Abha, Saudi Arabia
⁴Zoology Department, Faculty of Science, Cairo University, Cairo, Egypt
⁵Department of Anatomy and Embryology, Faculty of Veterinary Medicine, Alexandria University, Egypt

Address for correspondence: Prof. Mohamed Abumandour, Anatomy and Embryology Department, Faculty of Veterinary Medicine, Alexandria, Egypt, Post Box: 22758, tel: +201282586488, fax: +20452960450, e-mail: m.abumandour@yahoo.com or M.abumandour@alexu.edu.eg

Abstract

The current study was designed to give transmission microscopical focus on the dark and light pancreatic acinar β-cells of young-domesticated pig (Sus Suidae). This study depends on the fresh pancreatic specimens from five healthy young pig of two months ages that collected immediately after their slaughtered at the abattoir of Abdelkader Alexandria, Egypt. In our findings, the acinar pancrease formed of pyramidal pancreatic acinar cells with large spherical nuclei of condensed heterochromatin at the periphery and prominent eccentric nucleoli. Zymogen granules observed at the apical region of the acinar cells, and they appear as electron dense bodies. Numerous mitochondria and Golgi complexes observed in the acinar cell cytoplasm. The electron dense acinar cells were joined by junctional Complexes. The rough
endoplasmic reticulum was more prominent in the electron-dense acinar cells than did electron-lucent acinar cells. There was no connective tissue capsule separate the acinar portion of pancreas from the pancreatic islets. The pancreatic islets mainly formed of Beta cells. The irregular α-cells possess numerous small granules. The cytoplasmic B-cells granules were surrounded by hallow area and enclosed by a limiting membrane. Delta cells were generally polygonal in shape and found in clumps throughout the islet and they were also identified in between β-cells. Their granules were of moderate electron density and were generally smaller than β-cells’ granules. The limiting membrane was tightly enclosed the delta cells granules and the hallow area around the granule were found similar to the granules of β-cells.

**Key words:** pancrease, sus suidae, transmission electron microscope, pancreatic islets, pancreatic cells

**INTRODUCTION**

The omnivorous domesticated pig (*Sus Suidae, Erxleben 1777*) species belonged to order Artiodactyla, family suidae, and genus sus. This omnivorous domesticated pigs bred mostly for the meat consumption plus they feed on plants and sometimes on insects and fish (1).

Pancreas described anatomically as an accessory portion of the digestive system and has a clinical perspective as it is the objective of two significant sicknesses that are diabetes mellitus and pancreatic cancer. It is to be trusted that a superior comprehension of the morphology and histology of this organ will in the long run add to the improvement of novel treatments for the treatment of either or both of the Above infections (2). The pancreas is a composite organ comprising of exocrine and endocrine parts. The exocrine part comprises of acini that discharge stomach related proteins into pipes. The endocrine part, the pancreatic (islets of Langerhans), comprises of masses of endocrine cells inserted inside the exocrine pancreas, where pancreatic chemicals of insulin, glucagon, somatostatin, and pancreatic polypeptide (PP) are circulated into the entire body by means of veins (3).

The pancreas consists of exocrine acini that produce digestive enzymes and endocrine islets and that produces hormones. The exocrine acini of the pancreas consists mainly of tubuloacinaiar gland (4). The digestive enzymes of the secretory acini are circulated to the intestine through a branched network of ducts. Cells in the endocrine islets produce hormones that
regulate the body metabolism via the blood circulation (5). The endocrine portion of the pancreas composed of cellular aggregations that are scattered among the pancreatic acini known as islets of Langerhans (6). The endocrine part of the pancreas represents one or two percent of the pancreatic mass (7). The mammalian endocrine pancreas composed of Alpha cells (α-cells or glucagon-producing cells), Beta cells (β-cells, insulin-producing cells), Delta cells (Δ-cells, somatostatin-producing cells), and F-cells (PP-cells, pancreatic polypeptide-producing cells) as described by (6).

The available data on the structure of the pancreas generally focus on the histological features of the pancreas in the adult stage. In addition, there is a lack of available data on the structure of the pancreas in newborn animals, especially on the domesticated pig. Therefore, the current work describes the light and electron microscopic structure of the exocrine and endocrine pancreas of the newborn domesticated pig (Sus Suidae). Finally, our findings presented in this study, together with those presented in the previous study, provide a comprehensive report of the light and ultrastructural structure of the pancreas of newly born pig.

**MATERIALS AND METHODS**

**Sample’s collection**

Fresh specimens of pancreas from five apparently healthy of two months age domesticated pig (Sus Suidae) were collected immediately after their slaughtered at the abattoir of Abdelkader Alexandria, Egypt. The young, domesticated pig (Sus Suidae) were slaughtered for human food, and consequently had been examined prior to slaughter by the slaughterhouse veterinarian to obtain approval for human consumption. The present investigation was prepared according to the guidelines for the using and caring of the laboratory animals and follow the animal Ethics and welfare in the Faculty of Veterinary Medicine, Alexandria University and according to the Egyptian’s laws.

**Transmission electron microscopy (SEM) description**

Fresh pancreatic tissue samples of 1 mm thickness were obtained, fixed at 4°C in a solution of 2.5% glutaraldehyde for 24 h, rinsed in 0.1 M cacodylate buffer at pH 7.4,
then post-fixed for 1 h at room temperature with 1% osmium tetroxide. Then, these samples were put at 4°C in washing solution formed from 0.1 M sodium cacodylate containing 5% sucrose, processed by the tannic acid. Samples were dehydrated with ethyl alcohol in a graded sequence and eventually embedded in epoxy resin (8). For tissue examination, semithin sections were cut and stained with toluidine blue. Ultrathin sections (50-60 μm) were cut and stained with uranyl acetate dihydrate and lead citrate saturated solution (9). Finally, the sections were photographed by JEOL JEM-2100 TEM at Faculty of Science, Alexandria University, Egypt.

RESULTS

The acinar pancrease is formed of pyramidal pancreatic acinar cells which had a relatively large spherical nuclei with condensated heterochromatin at the periphery and prominent nucleoli that found eccentric to the nucleus (Fig.1). The cytoplasm contains cisternae of the rough endoplasmic reticulum (Fig.1/ER). Zymogen granules were found at the apical region of the acinar cells, and they appear as electron dense bodies (Fig.1/green arrowheads). Numerous mitochondria and Golgi complexes were found in the cytoplasm of each acinar cell. The acinar lumen was continuous with the intercalated duct (Fig.1B/MT). There were two types of acinar cells; the first cellular type is the electron lucent acinar cells that characterized by few numbers of zymogen granules, and the second cellular type is the electron dense acinar cells that characterized by the presence of numerous zymogen granules and electron dense nucleus (Fig.2/Dr). The electron dense acinar cells were joined by junctional Complexes (Fig.2/Li). The centro-acinar cells have centrally located nucleus (Fig.2/NU).

The rough endoplasmic reticulum was more prominent in the electron-dense acinar cells than did electron-lucent acinar cells (Fig.2B-D/Dr, ER). There was no connective tissue capsule separate the acinar portion of pancrease from the pancreatic islets. The distribution of pancreatic islets does not uniform, but they observed scattered and have different electron density of granules. The pancreatic islets mainly formed of Beta cells (Fig.1/β). The α-cells characterized by an irregular outline and possessed numerous small granules that characterized by a moderate to high density surrounded by a membrane. The nuclei of the cells were ovoid. Cytoplasmic vacuoles were found and the rough endoplasmic reticulum that consisted of narrow tubules (Fig.1-2/ER). Golgi apparatus appeared as small irregular lamina and their cytoplasmic granules were spherical and were larger than those of α-cells. The cytoplasmic B-cells granules were
surrounded by hallow area and enclosed by a limiting membrane (Fig.2). Delta cells were
generally polygonal in shape and found in clumps throughout the islet and they were also
identified in between β-cells (Fig.2). Their granules were of moderate electron density and were
generally smaller than β-cells’ granules. The limiting membrane was tightly enclosed the delta
cells granules and the hallow area around the granule were found similar to the granules of Beta
cells.

DISCUSSION

The small clusters of pancreatic endocrine cells were first depicted by Paul Langerhans in
1869, after whom they were named as "Islets of Langerhans". From that point forward,
numerous morphological examinations have been completed utilizing the different histological
and ultrastructural techniques in numerous animal species such as camel (10-13), bovine (14,
15), feline (16), monkey (17), rabbit (18), rodent, canines and Guinea pig (19), mice (20), and rat
(21-23). The endocrine pancreas has likewise been concentrated in non mammalian species like
some avian species (24, 25), teleost fish (26), lizard (27), and frog (28).

The acinar pancreas formed of pancreatic acinar cells which had a relatively large
spherical nuclei with condensed heterochromatin at the periphery and prominent nucleoli that
found eccentric to the nucleus. The shape of the acinar cells had some variation in which the
current work reported the pyramidal cells, similar to that reported by (29) in large white
Yorkshire pigs (Sus scrofa) and (11) in Egyptian one-humped camel Camelus dromedarius,
while (15) described that there are some variation of acinar cell shape from columnar to
pyramidal in buffalo.

The current ultrastructural observations reported that the Zymogen granules appeared as
electron dense bodies, similar to that reported by. Zymogen granules were reported also by (15)
in buffalo, (11) in Egyptian one-humped camel Camelus dromedarius and (30) in human
pancrease of fetus and at 12 weeks fetus by (31). Moreover, the current work reported that there
are numerous mitochondria and Golgi complexes were found in the cytoplasm of each acinar
cell, in addition to the acinar lumen was continuous with the intercalated duct, similar to that
described by (11, 15, 32).

There are minor variations about the position of the Zymogen granules in acinar cells.
The current observations reported that the Zymogen granules were found at the apical region of
the acinar cells, similar to that reported by (29) in large white Yorkshire pigs (Sus scrofa) and (11) in Egyptian one-humped camel Camelus dromedarius and confirmed in the different animal species by (3) in text book, while (15) in buffalo reported that the zymogenic granules were observed in the supranuclear cellular part of the cell.

The current ultrastructural observations reported that there were two types of a cinar cells; the first cellular type is the electron lucent acinar cells that characterized by few numbers of zymogen granules, and the sconed cellular type is the electron dense acinar cells that characterized by the presence of numerous zymogen granules and electron dense nucleus. The current ultrastructural observations reported that the electron dense acinar cells were joined by junctional Complexes, in addition to the centro-acinar cells have centrally located nucleus, similar to that reported by (11) in Egyptian one-humped camel Camelus dromedarius and (14) in Holstein cattle pancreas.

The current ultrastructural observations reported that the rough endoplasmic reticulum was more prominent in the electron-dense acinar cells than did electron-lucent acinar cells, similar to that reported by (11) in Egyptian one-humped camel Camelus dromedarius. The current ultrastructural observations reported that there was no connective tissue capsule separate the acinar portion of pancrease from the pancreatic islets. The distribution of pancreatic islets does not uniform, but they observed scattered and have different electron density of granules.

There are three significant cell types in the islets of Langerhans: α-cell for the creation of glucagon, β-cell for creation of insulin, and δ-cell for creation of somatostatin; an arrangement dependent on the pioneer works of (33, 34). Likewise, other cell types (C, E, F, V , and X or PP) for the creation of pancreatic polypeptides were seldom seen in a couple of animal varieties like camel (11), canine, Guinea pig, rabbit (35) and mice (20). The current work described the presence of mainly three well-defined cells in the pancreatic islets and the islets mainly formed of Beta cells, similar to that observed by (10, 12, 13) in a single hump camel (Camelus dromedarius). The cytoplasmic B-cells granules were surrounded by hallow area and enclosed by a limiting membrane, similar to that reported by (11) in Egyptian one-humped camel Camelus dromedarius.

The current ultrastructural observations reported that the α-cells characterized by an irregular outline and possessed numerous small granules that characterized by a moderate to high density surrounded by a membrane. The nuclei of the cells were ovoid. Cytoplasmic vacuoles
were found and the rough endoplasmic reticulum that consisted of narrow tubules. Golgi apparatus appeared as small irregular lamina and their cytoplasmic granules were spherical and were larger than those of α-cells. These findings were similar to that reported by (11) in Egyptian one-humped camel Camelus dromedarius. Also, (10) in a single hump camel (Camelus dromedarius) confirmed these findings but differ in that the presence of the spherical nuclei of the polyhedral shaped cells.

Delta cells were generally polygonal in shape and found in clumps throughout the islet and they were also identified in between β-cells. Their secretory granules were of moderate electron density and were generally smaller than β-cells’ granules. The limiting membrane was tightly enclosed the delta cells granules and the hallow area around the granule were found similar to the granules of Beta cells, similar to that reported by (10) in a single hump camel (Camelus dromedarius). These findings were similar to that reported by (11) in Egyptian one-humped camel Camelus dromedarius but different in that the no hallow area around the granules. The presence of the Delta-cells was also described in the endocrine part of the pancreas of numerous animals species such as cats (36), rats (21-23), camels (11, 12, 15).

Acknowledgements

The authors extend their appreciation to the Deanship of Scientific Research at King Khalid University for funding this work through Research Group Project under Grant number (R.G.P.2/40/40).

Conflict of interest: None declared

REFERENCES

1. Kongsted AG, Horsted K, and Hermansen JE. Free-range pigs foraging on Jerusalem artichokes (Helianthus tuberosus L.)—effect of feeding strategy on growth, feed conversion and animal behaviour. Acta Agriculturae Scandinavica, Section A–Animal Science, 2013; 63(2): p. 76-83.
2. Seymour PA, Bennett WR, and Slack JM. Fission of pancreatic islets during postnatal growth of the mouse. Journal of Anatomy, 2004; 204(2): p. 103-116.
3. Eurell JA and Frappier BL. Dellmann's textbook of veterinary histology. 2013: John Wiley & Sons.
4. Frappier B. Digestive System. In: Eurell J, Frappier B (eds). Dellmann's Textbook of Veterinary Histology, 6th ed. Wiley-Blackwell. 2007.
5. Hafez SA, Zaghloul D, and Caceci T. Immunohistochemical identification of the endocrine cells in the pancreatic islets of the camel, horse and cattle. Eur J Anat, 2015; 19(1): p. 27-35.
6. Gartner L. Digestive System: Glands. In: Color Textbook of Histology, 3rd ed. Saunders Elsevier, Philadelphia, PA, pp 413. 2006.
7. Johnston C, et al. Anatomy and Physiology of Pancreatic Islets. In: Besser G, Bodansky H, Cudworth A (eds). Clinical Diabetes. An Illustrated text. Gower Medical Publishing, London, pp 1.1-1.14. 1988.
8. Hayat M. Basic Techniques for Transmission Electron Microscopy. 2nd Edn., Academic Press, Baltimore. 1986.
9. Kandyel RM, et al. Comparative ultrastructural-functional characterizations of the skin in three reptile species; Chalcides ocellatus, Uromastyx aegyptia aegyptia, and Psammophis schokari aegyptia (FORSKAL, 1775): Adaptive strategies to their habitat. Microscopy Research and Technique, 2021.
10. Bsoul M, et al. Ultrastructure of pancreatic endocrine cells of the single hump camel (Camelus dromedarius). Annals of Microscopy, 2013; 13: p. 36-42.
11. Hafez S and Zaghloul D. Light and electron microscopy of the pancreas of the Egyptian one-humped camel (Camelus dromedarius). Eur. j. anat, 2017; 21(1): p. 37-45.
12. Khatim M, et al. The structure and hormone content of the endocrine pancreas of the one-humped camel (Camelus dromedarius). Anatomischer Anzeiger, 1985; 159(1-5): p. 181-186.
13. Al-Adhhami A. Ultrastructure of Pancreatic Endocrine Cells of the Single Hump Camel (Camelus dromedarius). SCOPE OF THE JOURNAL: p. 36.
14. Bonner-Weir S and Like A. A dual population of islets of Langerhans in bovine pancreas. Cell and tissue research, 1980; 206(1): p. 157-170.
15. Gupta D, et al. Light and electron microscopic studies on prenatal differentiation of exocrine pancreas in buffalo. Veterinary medicine international, 2016; 2016.
16. Sato T, Herman L, and Fitzgerald PJ. The comparative ultrastructure of the pancreatic islet of Langerhans. General and Comparative Endocrinology, 1966; 7(1): p. 132-157.
17. Winborn WB. Light and electron microscopy of the islets of Langerhans of the Saimiri monkey pancreas. The Anatomical Record, 1963; 147(1): p. 65-93.
18. Williamson J, Lacy P, and Taylor K. Electron microscopy of islets Of langerhans in rabbit pancreas slices incubated in vitro. Biochemical Journal, 1967; 102(3): p. 928.
19. Fujita T. The cells and hormones of the GEP endocrine system-The current of studies. Gastro-entero-pancreatic endocrine system. A cell-biological approach, 1973: p. 1-16.
20. Al-ANI I, Histochimecal and ultrastructural studies on the islets of langerhans of lean and obese hyperglycaemic mice with age. 1978, Aston University.
21. Elayat AA, el-Naggar MM, and Tahir M. An immunocytochemical and morphometric study of the rat pancreatic islets. Journal of anatomy, 1995; 186(Pt 3): p. 629.
22. Goldsmith P, et al. Ultrastructural localization of somatostatin in pancreatic islets of the rat. Endocrinology, 1975; 97(4): p. 1061-1064.
23. Kocamis H, et al. Immunohistochemical distribution of insulin-, glucagon-, and somatostatin-containing cells in the pancreas of the rat (Wistar albino). Kafkas Univ Vet Fak Derg, 2009; 15: p. 611-614.
24. MIKAMI S-I and ONO K. Glucagon deficiency induced by extirpation of alpha islets of the fowl pancreas. Endocrinology, 1962; 71(3): p. 464-473.
25. McClISH RD and Eglitis JA. Distribution of the A and B Cells and of the Islets (Langerhans) in the Duck Pancreas. 1969.
26. Klein C and Van Noorden S. Pancreatic polypeptide (PP)-and glucagon cells in the pancreatic islet of Xiphophorus helleri H.(Teleostei). Cell and tissue research, 1980; 205(2): p. 187-198.
27. Epple A. Islet cytology in urodele amphibians. General and Comparative Endocrinology, 1966; 7(2): p. 207-214.
28. Etayo J, et al. Characterization of pancreatic endocrine cells of the European common frog Rana temporaria. General and comparative endocrinology, 2000; 117(3): p. 366-380.
29. Iniyah K, et al. Histomorphology of exocrine pancreas of large white Yorkshire pigs (Sus scrofa). Journal of Entomology and Zoology Studies, 2020; 8(3): p. 1484-1486.
30. Conklin JL. Cytogenesis of the human fetal pancreas. American journal of Anatomy, 1962; 111(2): p. 181-193.
31. Laitio M, Lev R, and Orlic D. The developing human fetal pancreas: an ultrastructural and histochemical study with special reference to exocrine cells. Journal of anatomy, 1974; 117(Pt 3): p. 619.
32. Caro LG and Palade GE. Protein synthesis, storage, and discharge in the pancreatic exocrine cell: an autoradiographic study. The Journal of cell biology, 1964; 20(3): p. 473-495.
33. Lacy PE. Electron microscopic identification of different cell types in the islets of Langerhans of the guinea pig, rat, rabbit and dog. The Anatomical Record, 1957; 128(2): p. 255-267.
34. Munger BL, Caramia F, and Lacy PE. The ultrastructural basis for the identification of cell types in the pancreatic islets. Zeitschrift für Zellforschung und Mikroskopische Anatomie, 1965; 67(6): p. 776-798.
35. Volk BW and wellmen KF. The Diabetic pancreas. Baillière Tindall, London. 1977.
36. Legg PG. The fine structure and innervation of the beta and delta cells in the islet of Langerhans of the cat. Zeitschrift für Zellforschung und mikroskopische Anatomie, 1967; 80(3): p. 307-321.

**Figure 1.** Transmission electron micrographs of young-domesticated pig (Sus Suidae) pancreas (Views A, B, C, and D) showing light (LβC) & dark β-cells (DβC) with slightly oval nuclei, abundant zymogen secretory granules of variable sizes (white arrowheads), numerous mitochondria (MT), rough endoplasmic reticulum (rER), and few vacuoles (V). (View A: X2500) and (View B: X 4000), and (view C and D: 6000).

**Figure 2.** Transmission electron micrographs of pancreatic acinar (Views A, B, C, and D) of young-domesticated pig (Sus Suidae) showing the junction between the light (LβC) & dark β-cells (DβC) at the microvillus surface (Red arrow). Note the presence of zymogen granules (white arrowheads), numerous mitochondria (MT), rough endoplasmic reticulum (rER), and cytoplasmic vacuoles (V) in the cytoplasm of both light and dark β-cells. (X 2000).

**Figure 3:** Transmission electron micrograph of pancreatic acinar β-cells young-domesticated pig (Sus Suidae) pancrease; light (LβC) & dark β-cells (DβC) of pig. Note the abundant mitochondria (MT), rough endoplasmic reticulum (rER), a well-developed Golgi apparatus (GA), zymogen granules (white arrowheads), and several variable sized vesicles (Zigzag arrows).
