

Electronic microscopic evaluation of the beta insular cells in type 2 diabetes

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Abstract

In this work it has been done an electronic microscopic evaluation of the intra-cytoplasmic organelle devices of the pancreatic beta insular cells in the type 2 diabetes mellitus (insulin independent). The motivation of this study is the lesions noticed within another study, photonic microscopic, reason why we have considered it important to show up as well the interest modifications of the constitutive organelles of the beta insular cells.

Keywords: pancreas, type 2 diabetes, organelles.

Introduction

The present paper is part of a complex study owing to the high incidence in the appearance of the type II insulin-independent diabetes, mainly within the last years, being well-known that diabetes is the 6th cause of death, enhanced by the cardiovascular diseases which lead to 66–75% of the diabetic patients' deaths.

Works drawn up between 1997 and 2004 have shown that diabetes occurs at 7.6‰ men and 6.7‰ women. It is known that type 2 diabetes, which represents 90–96% of the total cases of diabetes occurs following to the organism's incapability to respond in producing insulin, obesity being considered a major factor in its development.

Recent studies have determined the implication of a specific inherited genotype of the interleukin 6 (IL-6) gene in the increase of susceptibility of the appearance of type 2 diabetes on men, while on women and more obese persons no modification of the genotype could be associated with the susceptibility of the appearance of type 2 diabetes.

Starting from these data from the literature corroborated with the photonic microscopic and morphometric aspects, as well as those immuno-histochemical aspects obtained in our personal studies, we decided to realize an electronic microscopic study on beta insular cells, both on men and women with type 2 diabetes.

Material and methods

The studied material, of human origin, was represented by fragments of pancreas collected from the necropsies of 91 subjects with type 2 diabetes, over 5 years (1999–2004).

Their deaths occurred as the result of conditions of hepatic, pulmonary and/or cardiac etiology. The collection of the pieces was done in the Pathological Anatomy Laboratory of the "Filantropia" Hospital of Craiova, and the preparation of the

fragments for the electronic microscopic study was done in the Electronic Microscopy Laboratory of the "Carol Davila" University of Medicine and Pharmacy of Bucharest.

Transmission electronic microscopy (TEM) method

The fragments of pancreas were fixed in a 2% glutaraldehyde plus 2% paraformaldehyde solution in phosphate buffer of 0.1 M, pH 7.4 at 4° C, for 4–12 hours. Eventually we washed the fragments in phosphate buffer, post-fixation in 1% osmium tetroxide (OsO₄) in phosphate buffer for 60 minutes at 4° C, followed by dehydration in 60% solution of acetone in bi-distilled water 5 × 5 minutes, and inclusion in resins, and eventually inclusion in formel for 12 hours at 60°C.

Then they were sliced with the ultra-microtome, examined and photographed with the Philips CM 10 electronic microscope.

Results

The electronic microscopic examination of the beta insular cells allowed us to notice that the rough endoplasmic reticulum is very well-represented, maintaining its structural individuality from a beta cell to another.

In the beta insular cells, as it can be noticed in the picture, the rough endoplasmic reticulum is formed by a network of endoplasmic ducts with an almost parallel layout, with rare linking ducts between them, of relatively equal diameter, with numerous ribosomes attached to the external surface (Figure 1).

In the inter-duct space free ribosomes and poliribosomes can be noticed, with quasi-constant dimensions. The inter-duct hyaloplasma contains rare ribosomes of variable dimensions and structured material, less electronically dense.

In some of the beta cell we noticed the hypertrophy of the smooth endoplasmic reticulum, while the rough

endoplasmic reticulum appears less evident, up to its disappearance.

The mitochondria of the beta insular cells present significant modifications, to the effect that the internal membrane, the mitochondrial cristae appeared disorganized, with anarchic arrangement, and the mitochondrial matrix with homogenous aspect was arranged all over the surface of the mitochondrion.

This structural aspect makes us consider that the first organelles to suffer in the type 2 diabetes are the mitochondria of the beta insular cells, but even these suffer variably from a beta cell to another. Thus, the mitochondria presented various degrees of alteration, with mainly three types of lesions: significant swelling, ballooning with shortening and disorganizing of the cristae, accompanied by degeneration, and vacuolization (Figures 2 and 3).

Variations of dimension and density in the electron flux of the intracytoplasmic granules were noticed on some beta insular cells, compared to other near beta insular cells which showed minimal modifications of the intracytoplasmic granules, lamellar concentric aspects alternating with the presence of some endomembrane residues (Figure 4).

However, the beta insular cellularity in its whole was characterized by emphasized granular depletive aspects.

We also noticed significant modifications at the level of the capillary walls in the vicinity of the beta insular cells (Figure 5).

We consider necessary to mention that the modifications of the beta insular organelles did not show sex specificity.

☐ Discussions

Starting from the idea that the cell is a “system” constituted of numerous cellular organelles, as cellular sub-systems, we considered important to evaluate the beta insular organelles in the type I diabetes.

Thus it is known that the mitochondria are the most numerous organelles, whose position in the cell is determined by the large amount of ATP needed, regionally associated with the rough endoplasmic reticulum, generating the ATP necessary to synthesize the proteins, reason why they can change their position in cytosol [1, 2].

It is known that the dimensions of the mitochondria are within 7 μm of length. In our study we noticed the presence of mitochondria with dimensions near the above mentioned values, but also mitochondria whose shape modified rapidly. Thus they were able to decrease or increase the volume and even to fusion into elongated mitochondria, with their fragmenting into globular mitochondria [3, 4].

The mitochondrial external membrane is doubled on the inner side by an internal membrane with the same thickness, fitted with creases, the two membranes enclosing an inter-membrane space of 60–80 Å , extended in the cristae too.

In type 2 diabetes the mitochondria of the beta insular cells suffered modifications highly accentuated

for the internal membrane, with local regional destructions, with the growth and widening of the inter membrane space, with the dilatation of the internal cristae, and even with their disappearance [5].

The very significant mitochondrial ballooning can evolve towards degeneration, consisting in the total disappearance of the cristae and the internal membrane, the mitochondrion becoming but a vesicle, bordered by a membrane, with a high content of transparency to the electron flux.

Yet the content can also consist of local agglomerations of dense material or of a finer material, or of a more rough material, reticularly arranged. The cristae can fragment, forming small vesicles inside the main vesicle, which eventually transform into dense bodies, aspects characteristic to an irreversible degenerative process.

In the study of the rough endoplasmic reticulum we noticed the direct link with the nucleus of the beta insular cell, as it is known that the membrane external to the nuclear covering continues with that of the rough endoplasmic reticulum and the perinuclear space with the lumen of the reticulum. The smooth endoplasmic reticulum is found in the beta insular cells, in the extension of the rough endoplasmic reticulum, being disposed towards the external areas of the cytoplasm [6].

In the casuistry we have studied, the modifications of the rough endoplasmic reticulum, with distant ducts and ribosomes sporadic up to their disappearance in certain areas characterized the beta cellularity, as it is known that the linking of the ribosome to the reticulum is done by means of the I and II ribophorines, which, along with its recognition, also has the role of “tunnel” in the transport of the newly synthesized polypeptide chain, in the lumen of the reticulum. Probably the detachment of the ribosomes from the reticulum is owed to the alteration of the “insertion signal” in the beta insular cells [7].

No matter in which stage the granule was observed, either it was pre-pro-granule to pro-granule and then to the mature granule, it showed a dense content at the electron flux, with delimitation at the exterior from the endomembranes [8].

The electron microscopic evaluation showed modifications, with variations in dimensions and density on the electron flux of the granules contained by the beta insular cells in type 2 diabetes [9].

Of course it cannot be neglected the aspect of the endothelial wall of the insular capillaries, the thickening.

☐ Conclusions

We consider that the electron microscopical evaluation of some of the beta insular organelles allowed us to ascertain that the hypertrophy of the smooth endoplasmic reticulum with the detachment of the ribosomes, mitochondrial ballooning, modifications in the electron flux of the granules contained are non-specific, yet at the same time characteristic to type 2 diabetes.

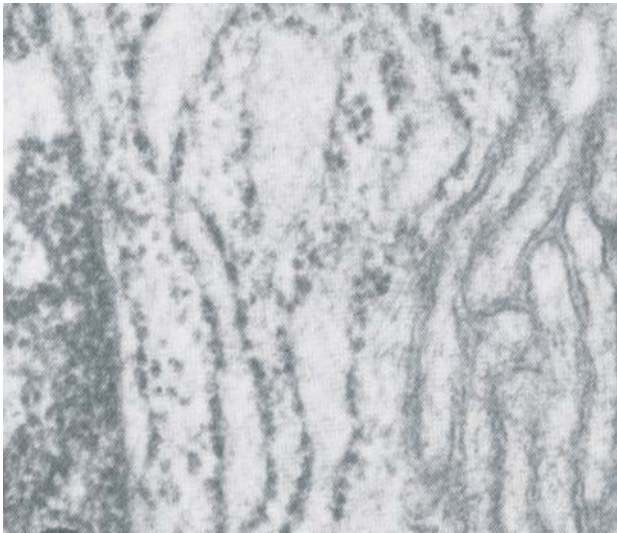


Figure 1 – Rough endoplasmic reticulum less evident.
It is to observe the disorganized network of ducts, far one from another, of different caliber on their trajectory, with ribosomes, highly dispersed in certain areas (TEM, $\times 35\ 000$)

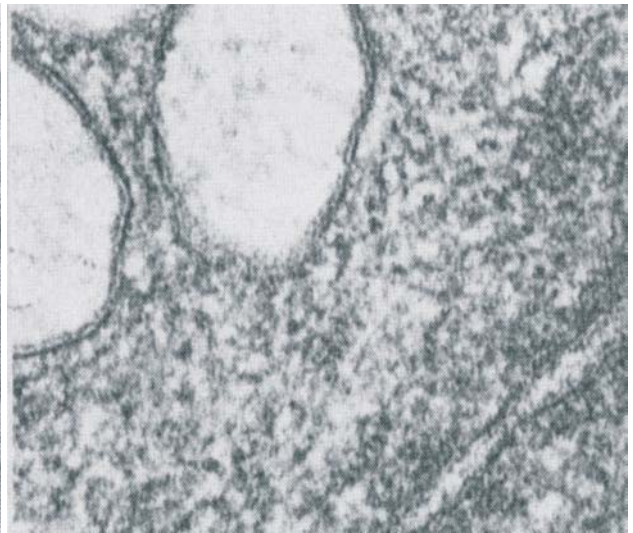


Figure 2 – Mitochondria in various stages of degeneration.
It is to observe mitochondria with vacuolization, significant swelling, erasing of the internal cristae (TEM, $\times 35\ 000$)

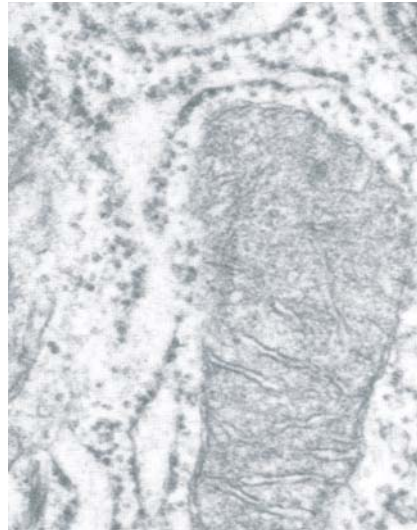


Figure 3 – Mitochondria with completely changed structures (TEM, $\times 35\ 000$)

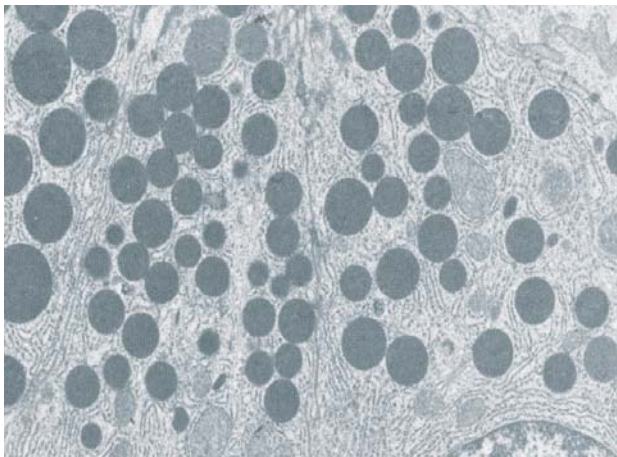


Figure 4 – Granules of secretion of variable dimensions and densities, with perinuclear topography. It is to observe the presence of endo-membranous residues (TEM, $\times 12\ 500$)

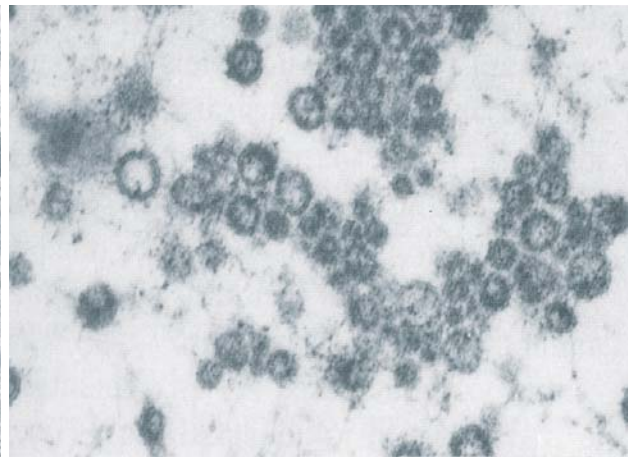


Figure 5 – Image similar to the previous one (TEM, $\times 12\ 500$)

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