Tissues, Pathology, and Diagnostic Microscopy

LS.2.P066
PDX1 expression in rat pancreas induced by sucrose-rich diet

M. Ukropina[1], V. Petković[1], M. Čakić-Milošević[1]

[1]University of Belgrade, Faculty of Biology, Belgrade, Serbia

mirela@bio.bg.ac.rs
Keywords: pancreas, PDX-1, acinar cells, cell reprogramming

Pancreas is an organ with endocrine and exocrine function. Endocrine cells clustered in islets of Langerhans secrete depending on type, various hormones involved mainly in maintaining glucose homeostasis. Prevailing type are glucose sensitive β-cells, whose product insulin lowers blood sugar level. The dominant, exocrine part of an organ comprises of ductal cells, and relatively homogenous population of acinar cells which are dedicated to digestive enzyme production.

Both endocrine and exocrine cells originate from the same progenitor cell. During the organ development many transcription factors are necessary for determining the cells fate and lineage differentiation: pancreatic and duodenal homeobox 1 (PDX1) homeodomain protein is essential for initiation of all pancreatic lineages. In mature pancreas PDX1 is highly expressed in insulin-producing β-cells, while its downregulation in the exocrine pancreas is required for normal acinar cell function [1].

In the β-cell nucleus, PDX1 is an activator of insulin gene transcription, thus PDX1 localization correlates with β-cell function and survival [2]. It has been reported that acute hyperglycemia induces β-cell proliferation and insulin production, while chronically elevated glucose concentrations deteriorate β-cell function, inducing apoptosis [3], and thus create a perfect milieu for onset of diabetes. In present study we wanted to investigate how both endocrine and exocrine component of the pancreas respond to sucrose-rich diet, in terms of PDX1 expression.

Young adult male Wistar rats fed with commercial rat food, were divided into two groups: control animals (C, n=6) had access to tap water, and sucrose group (S, n=8) to 10% sucrose solution in tap water, both *ad lib*. Animals were raised and kept under 12:12 light:dark cycle, at 21±1 °C. Experimental treatment lasted for three weeks and was approved by the local ethic committee. Pancreas tissue samples were routinely processed for light microscopy, and 5 μm thick sections were obtained. Presence of PDX1 protein was detected by LSAB immunohistochemistry method, using rabbit polyclonal anti-PDX1 antibody acording to manufacturers recommendations (Abcam). Photographs of approximately 30 islets per group, orig. magn. 40x were analyzed using Image J 1.45 software: data on islets profile area, and number of both PDX1-positive and -negative nuclei were collected.

In endocrine pancreas of control animals moderately strong reaction for PDX1 protein was detected, while in sucrose group PDX1 positivity was more intense and abundant (Figure 1, a and b). In both groups PDX1-positive nuclei were predominantly positioned in the central part of the islet, corresponding to location of insulin-secreting cells. Moreover, in S group in distinct portions of exocrine lobes, clusters of PDX1-positive nuclei were observed; PDX1 expression was mostly evident in peculiar binucleate population of acinar cells, with usually one nucleus remaining negative (Figure 1c). Results of morphometric analyzes revealed (Table 1) statistically significant increase of PDX1-positive nuclei number relative to number of all nuclei in S group. In addition, total number of nuclei per unit area of islet of Langerhans showed statistically greater value for S group also.

Glucose is one of the determinants of β-cell growth, thus principally this population of all endocrine cells within the islets undergoes accelerated growth in response to elevated glucose level [4]. This corresponds to our findings documenting the rise of PDX1-positive nuclei in S group, per unit area, most probably at the expense of proliferation of PDX1-positive, functional β-cells, since the number of negative nuclei in both groups remained unchanged. In exocrine portion of pancreas distinct population of PDX1 +/- binucleate acinar cells suggests the involvement of these cells in both insulin and digestive hormone production. In S group these specific acinar cells underwent fate switching, shifting towards PDX1 expression, and possibly insulin producing surogate β-cells. Similar cell reprogramming was achieved in different, non diet induced experimental setting [5]. We suggest that this gene reprogramming represents a good layout for insulin secretion in the case of more serious β-cell functional overload. In conclusion, moderately applied sugar-rich diet makes the pancreatic acinar cells alert to possible environmental challenges.

1. J. Jonsson, L. Carlsson, T. Edlund, and H. Edlund, Nature 371(1994), pp. 606-609.
2. A . Ardestani, N. S. Sauter, F. Paroni, G. Dharmadhikari, J. H. Cho, R. Lupi, P. Marchetti, J. Oberholzer, J. Kerr, Conte, and K. Maedler, J Biol Chem. 286 (2011), pp. 17144–17155.
3. K. Maedler, G. A. Spinas, R. Lehmann, P. Sergeev, M. Weber, A. Fontana, N. Kaiser, and M. Y. Donath, Diabetes 50 (2001), pp.1683-1690.
4. K. Fujimoto, and K. S. Polonsky, *Diabetes Obes. Metab.* 11 (*2009*), pp. 30-37.
5. Q. Zhou, J. Brown, A. Kanarek, J. Rajagopal, and D. A. Melton, Nature 455 (2008), pp. 627-632.

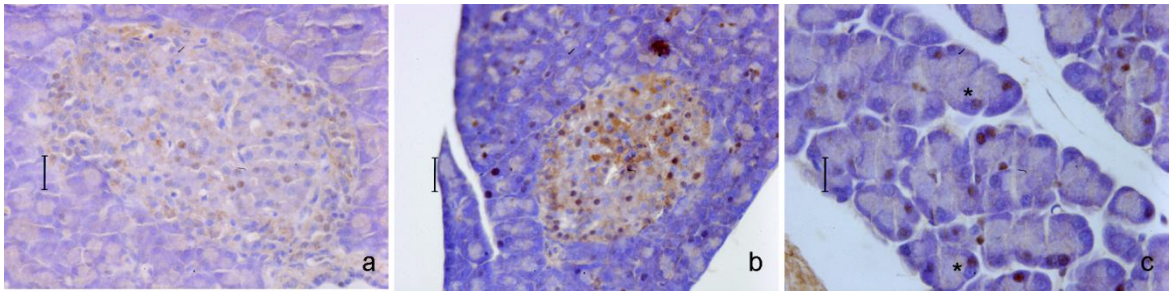

Figure 1. PDX1 localization is present in nuclei of islets of Langerhans in both control (a) and sucrose-fed rats with more prominent reaction in later (b). In sucrose group PDX1 +/- binucleate acinar cells was also detected (c, asterisk) (orig. magn. a and b – 40x, c – 63x).

	% of nuclei		Number of nuclei/μm^2		
	PDX1-	PDX1+	PDX1-	PDX1+	Total
Control group	55.08±3.44	44.92±3.44	0.0035±0.0003	0.0030±0.0003	0.0065±0.0003
Sucrose-fed group	45.70±2.96*	54.30±2.96*	0.0035±0.0004	0.0043±0.0004*	0.0078±0.0006*

Table 1. Results of morphometric analyzes of some parameters in islets of Langerhans in control and animals on sucrose-rich diet. Values are expressed as mean±standard error, statistical significance was determined with Student's t-test, * $p<0.05$.