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Islet-like cell clusters occur naturally in human gall bladder and are retained in diabetic conditions†

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A 53-year-old woman was admitted for persistent stomach ache. Ultrasonography examination revealed presence of gallstones and the patient electively decided to go for cholecystectomy. She had been diabetic for past 12 years and diabetes was controlled with combinations of drug, exercise and/or insulin injection as advised by her physician. Following cholecystectomy, gall bladder was taken to the laboratory and processed for routine histochemistry and immunostaining. The gall bladder showed normal morphology with columnar epithelial cells (Fig. 1A). We also observed presence of a large number of ('islet-like') cell aggregates (Fig. 1B) in epithelial cells isolated from the gall bladder. These aggregates were seen to be immunopositive for pancreatic islet hormones (Fig. 1C): insulin (green) and glucagon (red). The presence of such hormone-producing cells in hepato-biliary epithelium has been documented in mice [1]. In our laboratory, we observe the presence of such hormone-containing islet-like cell aggregates even in non-diabetic individuals. Such aggregates show presence of islet hormones and also contain (pro-) hormone transcript in high abundance (Supporting Fig. S1). Here, we present an unusual case wherein insulin-producing cells were seen to be retained in the gall bladder of a diabetic woman. Co-expression of insulin and glucagon hormones in several of these epithelial cells suggests that these hormone-containing cells may be similar to the 'immature' islet cells observed during pancreas development. Pro-insulin and pro-glucagon transcripts were detectable in these cells by TaqMan based duplex real-time PCR (Fig. 1C). These observations demonstrate for the first time that hormone-containing cell aggregates present in non-diabetic human beings may be retained in diabetic individuals. Though we presently do not understand the mechanisms that contribute to generation of hormone-producing cells in human gall bladder, these observations reveal that gall bladder epithelial cells can be a potentially important source of islet progenitors for cell replacement therapy in diabetes.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Fig. S1. Pancreatic hormone-containing cells are present in human gall bladder of non-diabetic individual. Epithelial cells

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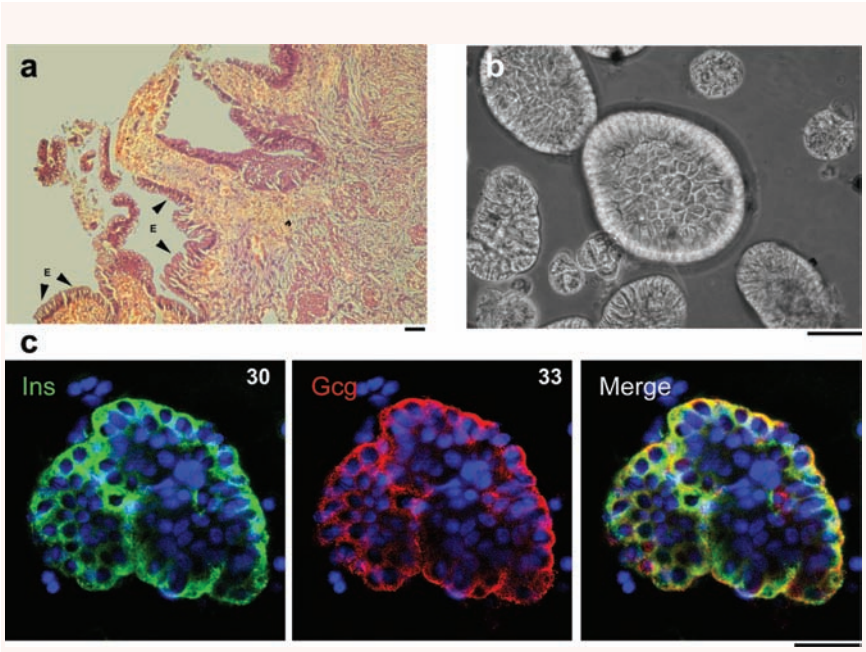

Fig. 1 Gall bladder epithelial cells in a diabetic woman. Paraffin sections obtained from gall bladder of a 53-year-old woman were stained with haematoxylin and eosin. Sections show epithelial cells lining the inner layer of gall bladder (**A**). Isolated epithelial cells form cell aggregates in culture dishes (**B**) and were seen to be immunopositive (**C**) for insulin (green) and Glucagon (red). Nuclei (blue) are labelled with Hoechst 33342 dye. Quantitative duplex TaqMan-based real-time PCR was carried out on these cells for transcript analysis of islet pro-hormones. The Δ cycle threshold (Ct) values obtained after normalization for 18S rRNA carried out in duplex reactions in each well are indicated in upper right corner of each panel (**C**). A Ct value of 39 is considered to be undetectable. Bar = 50 μm. Negative controls included RNA isolated from outer wall of gall bladder and human skin showed undetectable levels (Ct $>$ 39) of insulin and glucagon.

lining the gall bladder were cytospun and immunostained for insulin (green), glucagon (red) and pancreatic polypeptide (magenta) and visualized by confocal microscopy. Nuclei (Nuc) are co-stained with Hoechst 33342 dye. Quantitative duplex TaqMan-based real-time PCR was carried out for transcript analysis of islet pro-hormones. The Ct-values obtained after normalization for 18S rRNA carried out in duplex reactions in each well are indicated in upper right corner of each panel (ND: not done). Quantitative PCR in these cells confirmed presence of insulin transcript at levels that were 800-fold lower than adult human islets. A Ct value of 39 is considered to be undetectable. Bar = 50 μm.

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