DIABETES

A multi-omics roadmap of β-cell failure in type 2 diabetes mellitus

Gaowei Wang and Maike Sander

Pancreatic β-cell dysfunction contributes to type 2 diabetes mellitus (T2DM) pathogenesis. However, human islet studies have revealed few consistent molecular changes in T2DM, owing partly to technical and biological confounding factors. A new multi-omics study of islets from well-phenotyped living donors attempts to overcome these limitations to catalogue T2DM-associated molecular changes.

Refers to Wigger, L. et al. Multi-omics profiling of living human pancreatic islet donors reveals heterogeneous beta cell trajectories towards type 2 diabetes. *Nat. Metab.* **3**, 1017–1031 (2021).

Failure of pancreatic β-cells to secrete insulin in sufficient amounts to maintain blood glucose homeostasis is a hallmark feature of type 2 diabetes mellitus (T2DM). Insights into the molecular causes of β-cell failure in T2DM are mostly derived from rodent models and human islets from cadaveric donors. These studies suggest that altered mitochondrial metabolism and loss of β-cell identity (dedifferentiation) are the predominant drivers of insulin insufficiency in T2DM, rather than β-cell death[1]. However, the molecular events driving β-cell failure in T2DM remain unclear and numerous biological and technical confounding factors make human islet studies highly challenging.

To overcome these limitations, a study by Wigger et al.[2] analysed human islets collected from 133 metabolically phenotyped patients across the glycaemic spectrum (FIG. 1). Samples were snap-frozen upon collection from living donors undergoing pancreatectomy for pancreatic disease, thereby eliminating confounding artefacts, such as effects of medications and metabolic stress in the terminal condition, known to affect islets from brain-dead donors. Furthermore, to avoid induction of stress response genes by the enzymatic islet isolation procedure[3], islets were extracted using laser-capture microdissection. The authors conducted RNA sequencing as well as proteomic analysis on a subset of samples. In addition, plasma lipidomic and sphingolipid profiles were generated from the same cohort of patients. To minimize confounding factors, transcriptomic analysis was restricted to islet samples that highly expressed insulin. The authors used network analysis to infer gene modules (highly co-regulated genes) and then identified gene modules and lipid species most strongly associated with plasma HbA_{1c} levels[2].

The principal finding in the present study[2] was that genes that encode proteins involved in processes affecting insulin secretion were dysregulated in T2DM (FIG. 1). These processes include oxidative phosphorylation, protein transport to the endoplasmic reticulum and cytokine signalling. Analysis of transcript levels across the glycaemic spectrum showed that dysregulation of these genes was correlated with HbA_{1c} levels, suggesting progressive changes as glucose control deteriorates. As shown in mouse islets[4], overall concordance between transcript and protein levels in human islets was poor[2]. Nevertheless, the transcript and protein levels of the glycolytic enzyme ALDOB and the glucose transporter SLC2A2 were identified as having strong positive and negative associations, respectively, with HbA_{1c} levels. Furthermore, mitochondrial proteins were less abundant in T2DM islet samples than islets from people without T2DM, consistent with decreased expression of genes involved in oxidative phosphorylation[2]. These indications of

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apparent mitochondrial dysfunction agree with prior observations in human islets from smaller T2DM cohorts[5]. The study also uncovered associations between plasma ceramide levels (C20 and C18) and ether-linked phosphatidylcholines and HbA_{1c} levels, suggesting the potential value of these lipids as biomarkers for glucose intolerance and perhaps β-cell dysfunction[2].

One, perhaps surprising, finding is that genes related to β-cell identity (such as *PDX1*, *MAFA*, *NKX6.1* and *UCN3*) were not altered in T2DM islet samples, arguing against β-cell dedifferentiation, at least at the transcriptional level[2]. The authors further argue that islet samples from different patients with T2DM should exhibit molecular similarity if dedifferentiation was a common occurrence during T2DM pathogenesis. They instead observed substantially greater heterogeneity of transcriptomic and proteomic profiles among islet samples from individuals with T2DM than among samples from individuals without diabetes mellitus. However, the role of β-cell dedifferentiation in human T2DM remains controversial. Some studies, including single-cell transcriptomic studies, have found evidence supportive of β-cell dedifferentiation[6,7], while others have not[8]. These discrepancies illustrate the necessity for sufficient sample sizes, rigorous accounting for confounding factors and application of informative computational tools in human islet studies.

While this study begins to address some of these challenges, the broad applicability of the findings still needs to be demonstrated across the spectrum of people with T2DM, including those without pre-existing pancreatic diseases. A caveat of the current study is its analysis of whole islets, which are composed of different endocrine cell types as well as endothelial, immune and stromal cells. Moreover, substantial heterogeneity exists within the β-cell population at functional, transcriptional and epigenetic levels[9,10] that cannot be accounted for with bulk analysis. Large-scale analyses of human islets with single-cell technologies should help pinpoint changes specific to β-cells,

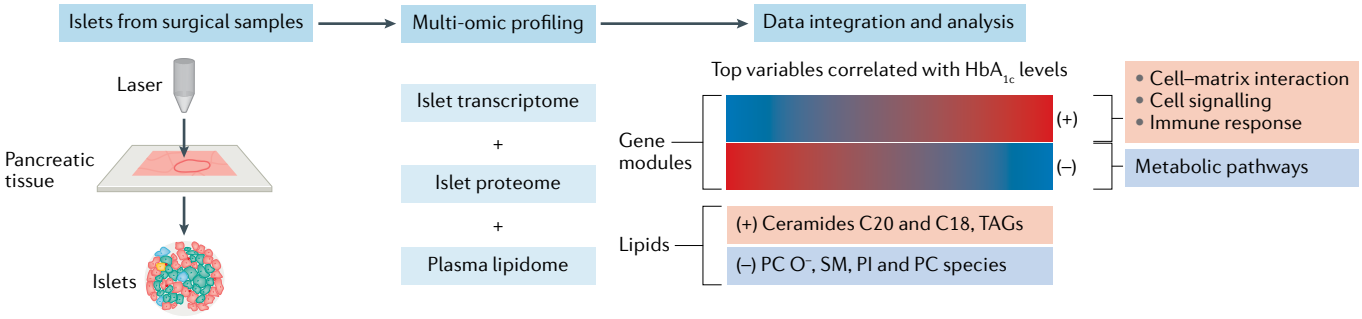

Fig. 1 | **Relationships between islet gene and protein expression, plasma lipids and plasma HbA_{1c} levels.** Multi-omic profiling of well-phenotyped living islet donors (without diabetes mellitus, $n = 18$; impaired glucose tolerance, $n = 41$; type 3c diabetes mellitus (diabetes due to diseases of the exocrine pancreas), $n = 35$; type 2 diabetes mellitus, $n = 39$) reveals gene expression, protein expression and plasma lipid profiles correlated with progression to type 2 diabetes mellitus. (+), positive correlation; (–), negative correlation; PC O^-, phosphatidylcholine O-class; PI, phosphatidylinositol; SM, sphingomyelin; TAGs, triacylglycerols.

or subtypes thereof, and elucidate mechanisms of β-cell failure. Heterogeneity in the aetiology of T2DM is probably another important contributor to the molecular heterogeneity of islets. Incorporation of genetic data in multi-omic profiling studies could help identify subtypes of T2DM with similar molecular and clinical features. Finally, a complete understanding of the stepwise events leading to β-cell failure in T2DM will require validation in longitudinal studies. Such studies are severely limited in feasibility by the relative inaccessibility of the pancreas, so complementary approaches in human cell models and animal models of T2DM will be useful. We could well learn that there is more than one path to β-cell dysfunction in T2DM.

One interesting observation from this study is that upregulated genes substantially outnumbered downregulated genes in islets from donors with prediabetes and donors with T2DM. T2DM islet samples exhibited a global increase in histone acetylation, which is an epigenetic modification associated with gene activation. These findings suggest an epigenetic component to altered islet gene regulation in T2DM. The authors propose a possible mechanism: the downregulation of two mitochondrial enzymes (ACADS and ACADSB), which are involved in inhibition of histone deacetylases, leads to alterations of the epigenome in T2DM islets via regulation of histone acetylation. These epigenome alterations, in turn, induce broad gene expression changes. This model is intriguing, as it suggests that several molecular and functional phenotypes observed in β-cells during T2DM progression result from changes in mitochondrial function. Of course, the model still needs validation.

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In conclusion, this study provides a rich starting point for unravelling the mystery of the path to β-cell dysfunction in T2DM. Moving forwards, the field will need to integrate multi-modal data, including at single-cell level, from even larger cohorts of genotyped and phenotyped islet donors. Datasets of increasing complexity will probably require new analytical tools to identify prognostic classifiers and disease mechanisms. Such approaches will help define subtypes of T2DM, and biomarkers thereof, and could eventually enable precision approaches to the treatment of T2DM.

Gaowei Wang and Maike Sander ✉

Department of Pediatrics, Pediatric Diabetes Research Center, University of California San Diego, La Jolla, CA, USA.

✉*e-mail: masander@ucsd.edu*

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Competing interests
The authors declare no competing interests.