Islet Cell Proliferation is Inappropriately Maintained in the Pancreas of Children with Congenital Hyperinsulinism in Infancy

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Background

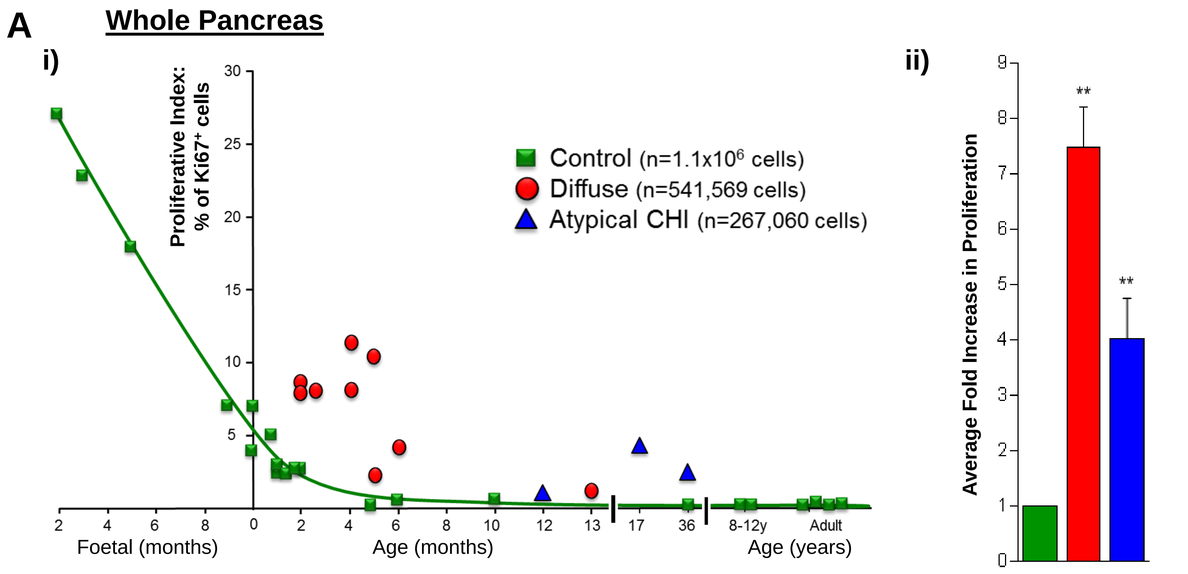
Congenital Hyperinsulinism of Infancy (CHI) is a potentially lethal condition of profound hypoglycaemia caused by unregulated insulin release in the neonatal period and early infancy. CHI mainly arises due to mutations in ATP-sensitive K-channel genes (*ABCC8* and *KCNJ11*) which can manifest in all islets cells – diffuse CHI (CHI-D), or can be localised to a focal lesion, focal CHI (CHI-F). Increased rates of cell proliferation have been reported in the CHI-D and this may be liked to *ABCC8* and *KCNJ11* defects. Here, we examined the proliferative index (PI) of islet cells in CHI-D patients and compared this with focal CHI (CHI-F), which is caused by loss of cell cycle repression in β-cells specifically within the focal domain. We also examined islet PI in patient tissues with severe CHI unrelated to defects in *ABCC8* and *KCNJ11,* atypical CHI (CHI-A).

Methods

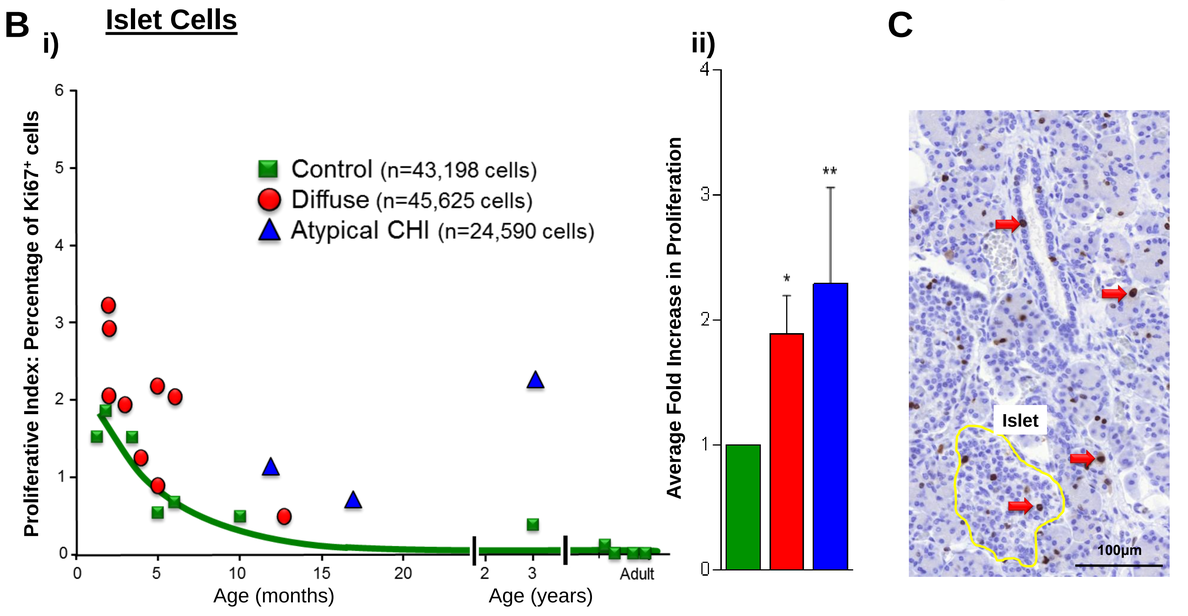
Tissue was obtained following surgery from patients with CHI-D (n=10 patients with *ABCC8* gene defects), CHI-F (n=6 patients with *ABCC8* gene defects), and CHI-A (n=3 patients with unknown genetic causes of disease). Neonatal control (n=12, 2-days to 36-months of age), foetal tissues (n=5, 10 to 35 weeks post conception) and adult tissues (n=4) were included as control groups. Immunohistochemistry staining with Ki67 on histological sections (5 µm) and high-content analysis were used to quantify the proliferation index (PI) and changes in PI expressed as average fold-changes across the various groups. Data were analysed with One-way ANOVA followed by Tukey's post hoc test.

Results 1

A **Whole Pancreas**



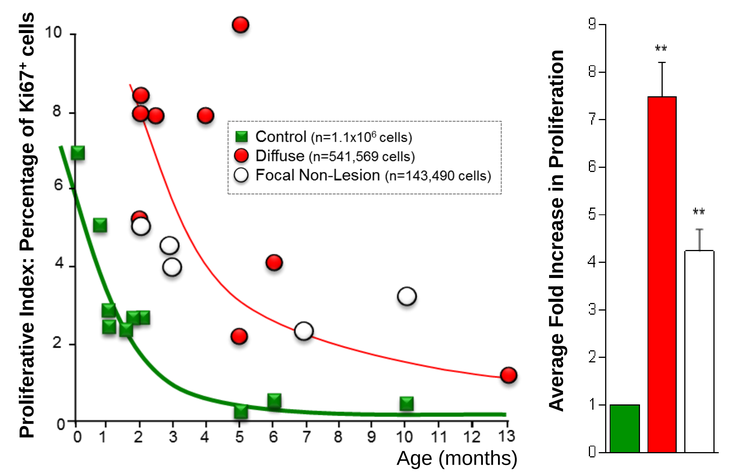
B **Islet Cells**



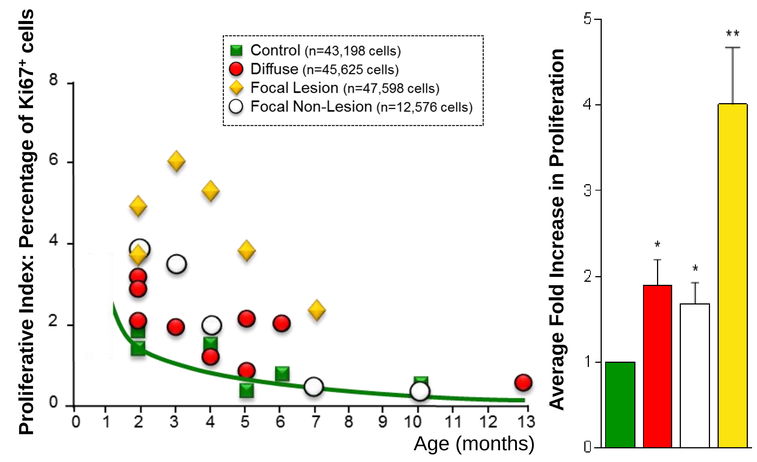
Graphical representations of proliferation index (PI) and fold changes in patients groups compared with age matched controls, foetal and adult control samples. Scatter plots of the differences in percentage of Ki67⁺ cells in CHI-D, CHI-A and age matched controls across whole pancreatic tissue (shown in **A-i**) and in islet specific regions (shown in **B-i**). Average fold changes in proliferation among CHI samples and age matched controls; data has been normalised to controls (**A-ii** and **B-ii**). Immunohistochemistry staining of Ki67 in a 10-week post-natal control pancreatic tissue. A single islet has been annotated in yellow and Ki67⁺ cells have been indicated by red arrows (**C**). P*<0.05; P**<0.01.

Results 2

A **Whole Pancreas**



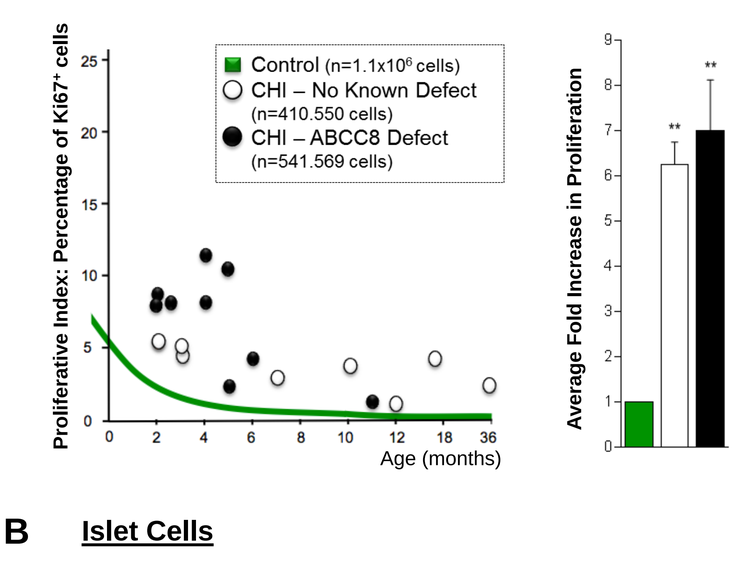
B **Islet Cells**



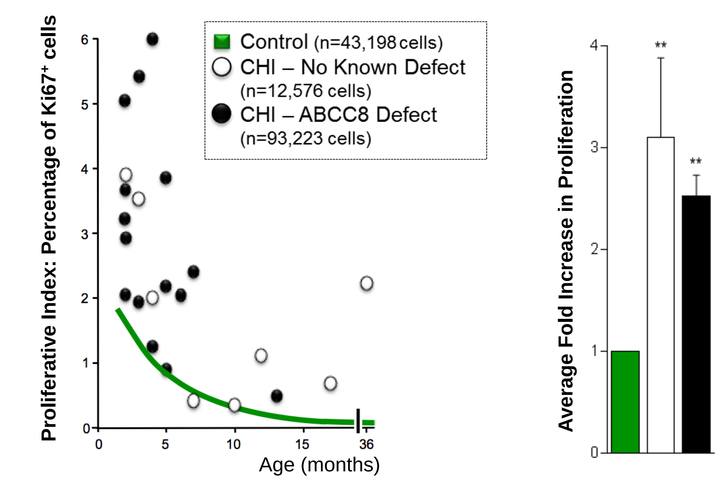
Graphical representations of PI and fold change in patients with various forms of CHI disease compared with age matched controls. Scatter plots of the differences in percentage of Ki67⁺ cells in CHI-D, CHI-F and age matched controls across whole pancreatic tissue (**A**) and in islet specific regions (**B**). Average fold changes in proliferation among CHI samples and age matched controls; data has been normalised to controls. P*<0.05; P**<0.01.

Results 3

A **Whole Pancreas**



B **Islet Cells**



Graphical representations of PI and fold change in tissues with *ABCC8*⁺ gene defect vs. no known mutations compared with age matched controls. Scatter plots of the differences in percentage of Ki67⁺ cells in CHI tissues and age matched controls across whole pancreas (**A**) and within islet specific regions (**B**). Average fold changes in proliferation between CHI samples and age matched controls; data has been normalised to controls. P*<0.05; P**<0.01.

Conclusions

1. There is a negative correlation between age and cell proliferation in control pancreas. 2. All three forms of CHI are associated with increased rates of cell proliferation. 3. Although there is a negative correlation between age and cell proliferation in CHI tissue (islets and pancreas), it is quantitatively different and not directly associated with *ABCC8* mutations.

Poster Template from www.manchester.ac.uk/photographics