Blocking Interleukin-1 beta Signaling Protects Pancreatic Islet Beta Cells From Intracellular and Extracellular Amyloid – Implications in Childhood Type 2 Diabetes

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INTRODUCTION

- The incidence of type-2 Diabetes (T2D; adult-onset diabetes), characterized by reduced β-cell mass and function, is progressively increasing in children and adolescents.
- A key contributing factor to the β-cell death in T2D is the intracellular and extracellular aggregation of the toxic protein, amyloid, in pancreatic islets. Amyloid also forms in cultured and transplanted islet grafts in type 1 diabetes (T1D).
- Amyloid formation plays a key role in islet inflammation by stimulating the production of interleukin-1ß (IL-1β), a pro-inflammatory cytokine, in islets which in turn further promotes its aggregation.

Fig 1. Pathogenesis of T2D.

- IL-1 receptor antagonist (IL-1RA, anakinra) and IL-1β neutralizing monoclonal antibody (nAb) block IL-1 β signaling by targeting the receptor and IL-1β, respectively.

AIMS

We examined if:

1. Blocking IL-1β signalling can reduce the intracellular and/or extracellular amyloid-induced ß-cell death.
2. Blocking IL-1β signalling can enhance β-cell survival in the presence of intracellular and extracellular amyloid formation.

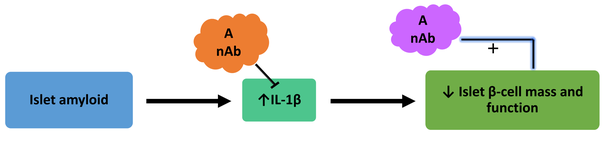
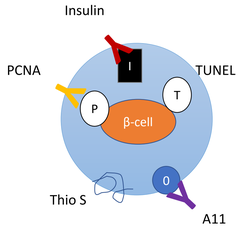


Fig 2. The mechanism of amyloid-induced β-cell toxicity and proposed protecting mechanisms of anakinra (A) and neutralizing antibody (nAb).

METHOD

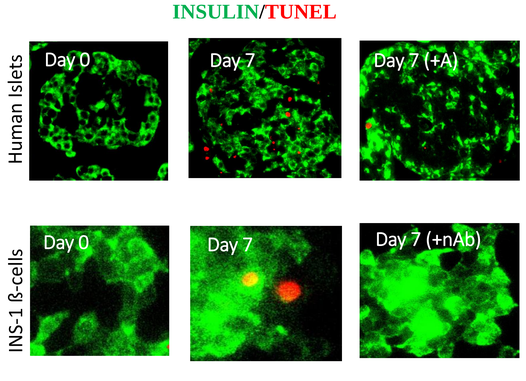
- Human islets (n= 4 donors) were cultured free-floating in CMRL (11.1 mmol/l glucose; 7 days) to form amyloid.
- INS-1 β-cells (n= 3 independent studies) were cultured in RPMI-1640 medium after transduction with prohIAPP-adenovirus to induce intracellular amyloid formation.
- Human islets and INS-1 ß-cells were treated with anakinra (10 µg/ml) or nAb (1 µg/ml), respectively.
- Quantitative immunohistochemistry was performed on paraffin-embedded human islet sections and INS-1 β-cells for insulin and Thioflavin S (large aggregates), A11 (small aggregates), TUNEL (apoptosis), or PCNA (proliferation).

Fig 3. Paraffin imbedded human islet sections or INS-1 ß-cells were immunolabelled for Insulin, thioflavin S, A11, TUNEL or PCNA.



RESULTS

Treatment of human islets with anakinra or INS-1 β-cells with nAb significantly reduced the number of TUNEL-positive and amyloid-positive β-cells. PCNA-positive β-cells were also increased post treatment



RESULTS, continued

Fig 4. Paraffin-embedded human islet sections and INS-1 β-cells from control, non-treated and treated [with anakinra (A) or neutralizing IL-1ß antibody (nAb)] were immunolabelled for insulin and TUNEL. Micrographs represent three independent studies.

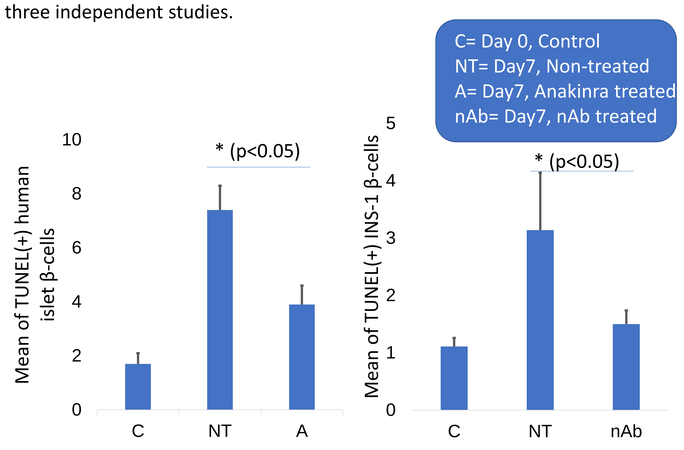


Fig 5. The proportion of TUNEL-positive (apoptotic) β-cells after 7-day treatment with anakinra (human islets; left) or nAb (INS-1cells; right). Data are expressed as mean±SEM of three independent studies.

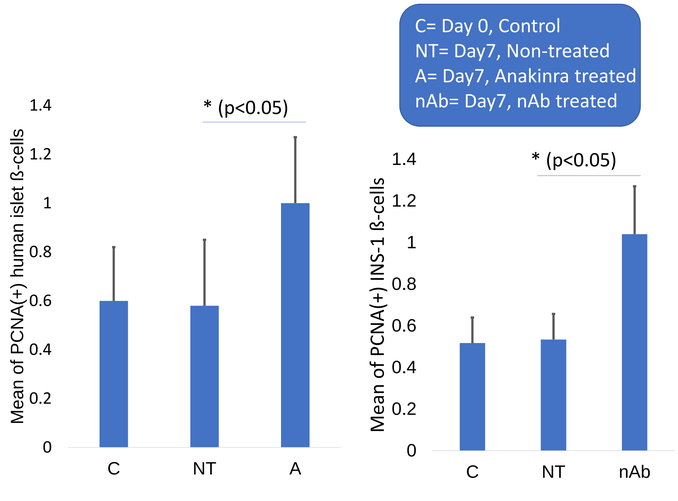


Fig 6. The proportion of PCNA-positive (proliferative) β-cells after 7-day treatment with anakinra (human islets; left) or nAb (INS-1 cells; right). Data are expressed as mean±SEM of three independent studies.

RESULTS, continued

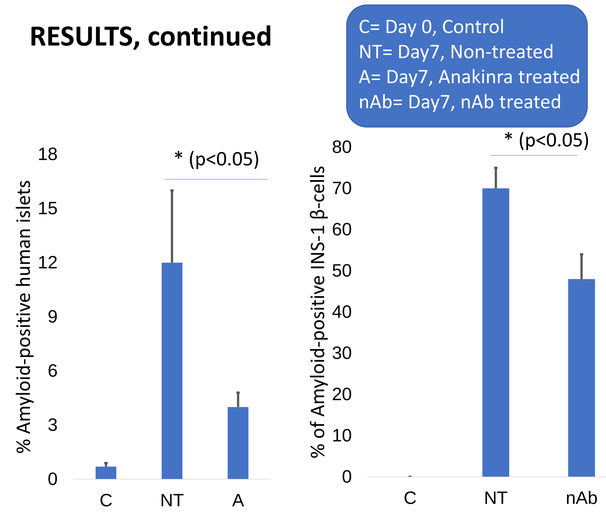


Fig 7. The proportion of amyloid-positive human islets (left) or amyloid-positive transduced INS-1 ß-cells (right) with and without treatment with anakinra or nAb, respectively. Data are expressed as mean±SEM of three independent studies.

CONCLUSION

- Treatment with anakinra or nAb significantly reduced intracellular and extracellular amyloid formation, respectively, decreased amyloid-induced ß-cell death, and enhanced ß-cell survival (proliferation).
- Reducing amyloid formation by blocking IL-1ß signalling may provide an effective approach to slow down the process of ß-cell loss in both children and adults with T2D.
- Blocking amyloid-induced IL-1ß signalling may also be of benefit in increasing the longevity of islet grafts in patients with T1D.

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