**ESM (Electronic Supplementary Material)**

# Human Islet Amyloid Polypeptide Aggregation Upregulates the Mitochondrial Cholesterol Transport Protein, StAR, and Induces Mitochondrial Dysfunction in Beta Cells

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**ESM Table 1**: Human islet donor characteristics

| Donor | Accession # | Gender | Age years | BMI kg/m$^2$ | HbA1c mmol/mol (%) | Source | Used in Figure |
|---|---|---|---|---|---|---|---|
| 1 | SAMN08769836 | F | 56 | 33.5 | 33 (5.2) | Scharp / Lacy | 1a, b, S1 |
| 2 | SAMN08769831 | F | 51 | 20.1 | 37 (5.5) | U. Miami | 1a, b, S1 |
| 3 | SAMN08769819 | F | 45 | 30.5 | 49 (6.6) | SC-ICRC | 1a, b |
| 4 | SAMN08769837 | M | 48 | 43.7 | 49 (6.6) | Scharp/ Lacy | 1a, b |
| 5 | SAMN08612554 | F | 51 | 32.0 | 34 (5.3) | U. Miami | 1a, c, S1 |
| 6 | SAMN08612553 | M | 51 | 25.5 | 36 (5.4) | U. Pennsylvania | 1a, c |
| 7 | SAMN18092805 | M | 56 | 21.6 | 32 (5.1) | SC-ICRC | 1c |
| 8 | SAMN18528866 | F | 32 | 25.7 | 32 (5.1) | Scharp / Lacy | 1c |
| 9 | SAMN18875704 | M | 37 | 28.4 | 38 (5.6) | Scharp / Lacy | 1c |
| 10 | SAMN09228907 | M | 62 | 36.1 | 40 (5.8) | Scharp / Lacy | 1a, S1 |
| 11 | SAMN09370567 | M | 32 | 28.5 | 33 (5.2) | U. Wisconsin | 1a, S1 |
| 12 | SAMN09432880 | M | 51 | 35.7 | 36 (5.4) | SC-ICRC | 1a, S1 |
| 13 | SAMN09844981 | M | 56 | 29.4 | 60 (7.6) | SC-ICRC | 1a |
| 14 | SAMN09862214 | M | 31 | 31.8 | 34 (5.3) | SC-ICRC | 1a, S1 |
| 15 | SAMN10372788 | F | 50 | 35.4 | 85 (9.9) | SC-ICRC | 1a |
| 16 | SAMN11791244 | M | 47 | 36.1 | 39 (5.7) | SC-ICRC | 1a, e, f, g, S1 |
| 17 | SAMN12339206 | M | 45 | 32.9 | 36 (5.4) | SC-ICRC | 1a, e, f, g, S1 |
| 18 | SAMN12534700 | M | 29 | 26.0 | 43 (6.1) | U. Pennsylvania | 1a, e, f, g, S1 |
| 19 | SAMN40619409 | M | 63 | 34.1 | 32 (5.1) | Prodo Labs | 1d, e, f, g, h, S1 |
| 20 | SAMN40709610 | F | 55 | 28.6 | 39 (5.7) | U. Miami | 1a, e, f, g, h, S1 |
| 21 | SAMN41657868 | M | 38 | 25.5 | 36 (5.4) | U. Wisconsin | 1a, e, f, g |
| 22 | SAMN42008301 | M | 36 | 30.0 | 31 (5) | U. Pennsylvania | 1d, e, f, g, h |
| 23 | SAMN48072591 | M | 62 | 31.4 | 34 (5.3) | Prodo Labs | 1g, h, S1 |
| 24 | SAMN48430443 | M | 50 | 25.8 | 33 (5.2) | Prodo Labs | 1g, h |

Scharp-Lacy Research Institute (Prodo labs)
SC-ICRC: Southern California Islet Cell Resource Center (City of Hope)
HbA1c in red = confirmed T2D donor

**ESM Table 2**: Primary and secondary antibodies and buffers for protein detection by western blotting, immunohistochemistry, or immunofluorescence

| Antibody | Vendor | Catalogue # | Species | RRID | Dilution for Western Blotting | Dilution for IHC / IF |
|---|---|---|---|---|---|---|
| **Primary Antibody** | | | | | | |
| Insulin | Millipore Sigma | I2018 | Mouse | AB_260137 | | 1:2000 |
| StAR | Cell Signaling Technology | 8449S | Rabbit | AB_10889737 | 1:1000 | 1:100 |
| Human StAR | LifeSpan Biosciences | C185729 | Rabbit | AB_3740098 | 1:500 | |
| StAR | Invitrogen | MA5-47013 | Mouse | AB_2938085 | | 1:100 |
| HSP90 | Cell Signaling Technology | 4874 | Rabbit | AB_2121214 | 1:1000 | |
| ATP5a | Invitrogen | 459240 | Mouse | AB_2532234 | 1:1000 | 1:100 |
| TOMM20 | Abcam | ab289670 | Rat | AB_2943038 | | 1:100 |
| **Secondary Antibody** | | | | | | |
| anti-Rat AF488 | Invitrogen | A11006 | Goat | AB_2534074 | | 1:250 |
| anti-Mouse AF568 | Invitrogen | A11004 | Goat | AB_2534072 | | 1:250 |
| anti-Rabbit AF568 | Invitrogen | A11011 | Goat | AB_143157 | | 1:250 |

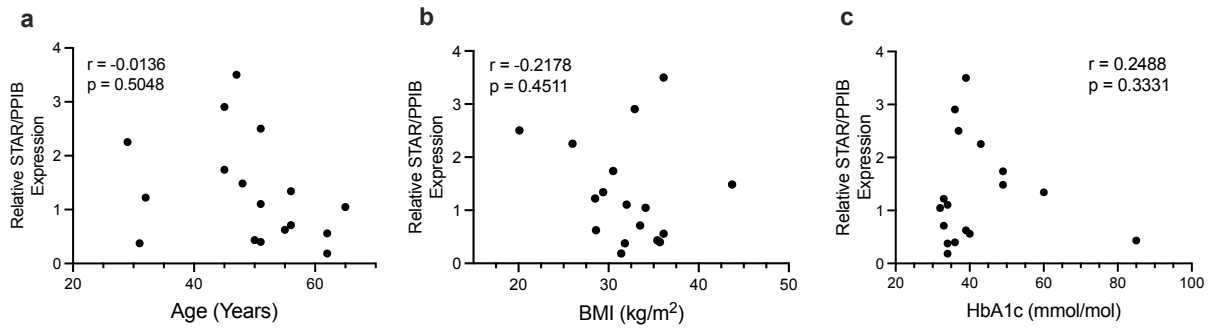
| **Buffers** | |
|---|---|
| **Purpose** | **Buffer** |
| Western blotting | Running Buffer: 0.3% (w/vol) Tris base (Sigma T1503), 1.44% (w/vol) glycine (Sigma G7126), pH 8.3, 0.1% (w/vol) SDS (Invitrogen 15553027) |

| | |
|---|---|
| Western blotting | Transfer Buffer: 0.3% (w/vol) Tris base, 1.44% (w/vol) glycine, pH 8.3, 0.1% (w/vol) SDS, 20% (vol/vol) Methanol (Sigma 179337) |
| IHC/IF | Blocking buffer: 1x PBS (Invitrogen AM9625), 0.2% (vol/vol) Triton X-100 (Thermo Fisher 85111), 0.01% (w/vol) sodium azide (Sigma S2002), 1% (w/vol) bovine serum albumin (Sigma A7888), 2% (vol/vol) normal goat serum (Gibco 16210064) |
| IHC/IF | Antibody incubation buffer: 1X PBS, 0.2% (vol/vol) Triton X-100, 0.01% (w/vol) sodium azide, 1% (w/vol) bovine serum albumin |
| IHC/IF | Wash buffer: 1X PBS, 0.2% (vol/vol) Triton X-100, 0.01% (w/vol) sodium azide |
| IHC/IF | Thioflavine S staining solution: 0.5% (w/vol) Thioflavine S (Sigma T1892) in water |

**ESM Table 3**: TaqMan probes for gene expression analysis

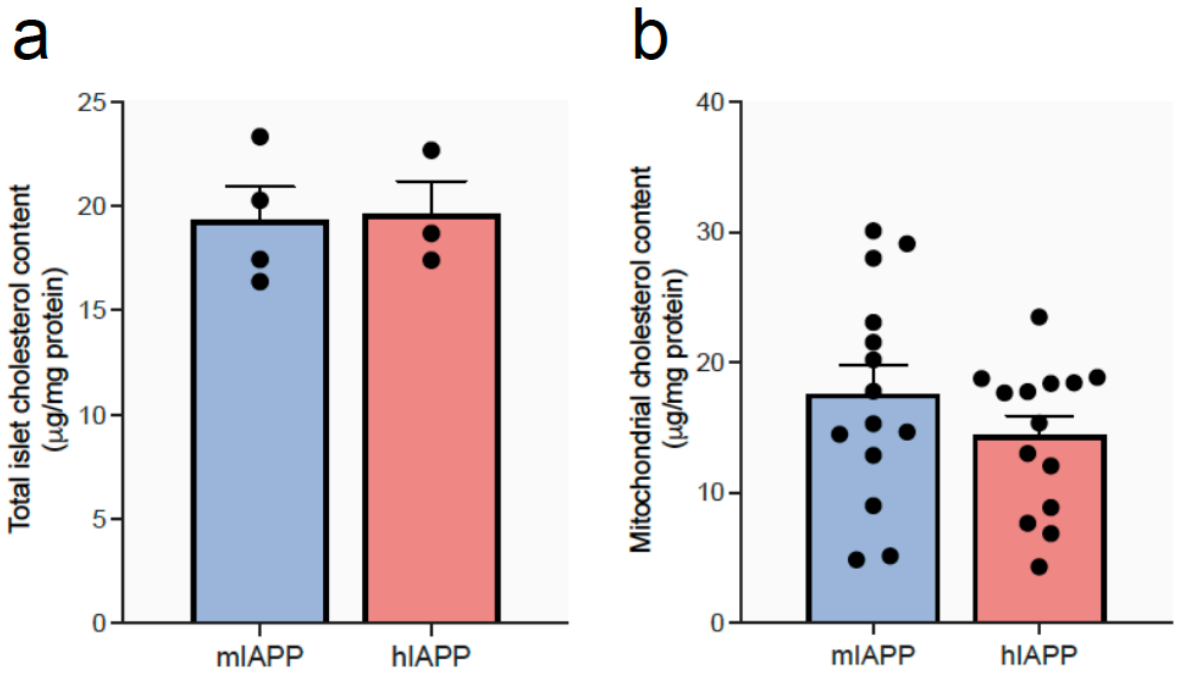
| Gene Name | Species | TaqMan Probe ID |
|---|---|---|
| Ppib (Cyclophilin) | Mouse | Mm00478295 |
| PPIB (Cyclophilin) | Human | Hs00168719 |
| Star (Stard1) | Mouse | Mm00441558 |
| StAR (Stard1) | Human | Hs00986559 |
| Stard3 | Mouse | Mm00445524 |
| Tspo | Mouse | Mm00437828 |

**ESM Figure 1: Relationship of StAR mRNA expression in human islets with age, BMI and haemoglobin A1c levels:**




StAR mRNA expression in human islets does not correlate with (a) age, (b) BMI, and (c) haemoglobin A1c levels from individual donors without diabetes (n=13, see Supplemental Table 1).

**ESM Figure 2: Islet and mitochondrial cholesterol content**




(a) Total islet cholesterol content relative to protein; n=3-4. (b) Mitochondrial cholesterol content relative to protein; n=14, in non-transgenic (mIAPP; blue bars) and RIP-hIAPP transgenic (red bars) islets.

Diabetologia

# Checklist for reporting human islet preparations used in research

Adapted from Hart NJ, Powers AC (2018) Progress, challenges, and suggestions for using human islets to understand islet biology and human diabetes. Diabetologia https://doi.org/10.1007/s00125-018-4772-2

| **Islet preparation** | **1** | **2** | **3** | **4** | **5** | **6** | **7** | **8[a]** |
|---|---|---|---|---|---|---|---|---|
| **MANDATORY INFORMATION** | | | | | | | | |
| Unique identifier | SAMN08769836 | SAMN08769831 | SAMN08769819 | SAMN08769837 | SAMN08612554 | SAMN08612553 | SAMN18092805 | SAMN18528866 |
| Donor age (years) | 56 | 51 | 45 | 48 | 51 | 51 | 56 | 32 |
| Donor sex (M/F) | F | F | F | M | F | M | M | F |
| Donor BMI ($kg/m^2$) | 33.5 | 20.1 | 30.5 | 43.7 | 32 | 25.5 | 21.6 | 25.7 |
| Donor $HbA_{1c}$ or other measure of blood glucose control | 33 mol/mol (5.2%) | 37 mol/mol (5.5%) | 49 mol/mol (6.6%) | 49 mol/mol (6.6%) | 34 mol/mol (5.3%) | 36 mol/mol (5.4%) | 32 mol/mol (5.1%) | 32 mol/mol (5.1%) |
| Origin/source of islets[b] | IIDP | IIDP | IIDP | IIDP | IIDP | IIDP | IIDP | IIDP |
| Islet isolation centre | Sharp / Lacy | U. Miami | SC-ICRC | Sharp / Lacy | U. Miami | U. Pennsylvania | SC-ICRC | Sharp / Lacy |
| Donor history of diabetes? Please select yes/no from drop down list | No | No | Yes | Yes | No | No | No | No |
| **If Yes, complete the next two lines if this information is available** | | | | | | | | |
| Diabetes duration (years) | | | | | | | | |
| Glucose-lowering therapy at time of death[c] | | | | | | | | |

| RECOMMENDED INFORMATION | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Donor cause of death | Head Trauma | CV / Stroke | Head Trauma | Anoxia | CV / Stroke | CV / Stroke | CV / Stroke | CV / Stroke |
| Warm ischaemia time (h) | | | | | | | | |
| Cold ischaemia time (h) | 11.3 | 14.9 | 0 | 7.5 | 14.5 | 15.5 | 5.7 | 8.5 |
| Estimated purity (%) | 90 | 90 | 80 | 90 | 90 | 90 | 90 | 85 |
| Estimated viability (%) | 95 | 95 | 97 | 95 | 90 | 93 | 96 | 95 |
| Total culture time (h)[d] | 84 | 75 | 92 | 61 | 94 | 45 | 36 | 91 |
| Glucose-stimulated insulin secretion or other functional measurement[e] | | | | | | | | |
| Handpicked to purity? Please select yes/no from drop down list | Yes | Yes | Yes | Yes | Yes | Yes | Yes | Yes |
| Additional notes | | | | | | | | |

[a]If you have used more than eight islet preparations, please complete additional forms as necessary
[b]For example, IIDP, ECIT, Alberta IsletCore

| **Islet preparation** | **9** | **10** | **11** | **12** | **13** | **14** | **15** | **16** |
|---|---|---|---|---|---|---|---|---|

| MANDATORY INFORMATION | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Unique identifier | SAMN18875704 | SAMN09228907 | SAMN09370567 | SAMN09432880 | SAMN09844981 | SAMN09862214 | SAMN10372788 | SAMN11791244 |
| Donor age (years) | 37 | 62 | 32 | 51 | 56 | 31 | 50 | 47 |
| Donor sex (M/F) | M | M | M | M | M | M | F | M |
| Donor BMI ($kg/m^2$) | 28.4 | 36.1 | 28.5 | 35.7 | 29.4 | 31.8 | 35.4 | 36.1 |
| Donor $HbA_{1c}$ or other measure of blood glucose control | 38 mmol / mol (5.6%) | 40 mmol / mol (5.8%) | 33 mmol / mol (5.2%) | 36 mmol / mol (5.4%) | 60 mmol / mol (7.6%) | 34 mmol / mol (5.3%) | 85 mmol / mol (9.9%) | 39 mmol / mol (5.7%) |
| Origin/source of islets[b] | IIDP | IIDP | IIDP | IIDP | IIDP | IIDP | IIDP | IIDP |
| Islet isolation centre | Scharp / Lacy | Scharp / Lacy | U. Wisconsin | SC-ICRC | SC-ICRC | SC-ICRC | SC-ICRC | SC-ICRC |
| Donor history of diabetes? Please select yes/no from drop down list | No | No | No | No | Yes | No | Yes | No |
| **If Yes, complete the next two lines if this information is available** | | | | | | | | |
| Diabetes duration (years) | | | | | | | | |
| Glucose-lowering therapy at time of death[c] | | | | | | | | |
| **RECOMMENDED INFORMATION** | | | | | | | | |
| Donor cause of death | Head Trauma | Anoxia | Head Trauma | CV / Stroke | CV / Stroke | Head Trauma | CV / Stroke | CV / Stroke |
| Warm ischaemia time (h) | | | | | | | | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Cold ischaemia time (h) | 11.5 | 10.7 | 11.5 | 6 | 6.5 | 5.5 | 8.5 | 6.5 |
| Estimated purity (%) | 90 | 90 | 95 | 80 | 75 | 80 | 80 | 95 |
| Estimated viability (%) | 95 | 95 | 91 | 96 | 93 | 95 | 96 | 95 |
| Total culture time (h)[d] | 65 | 92 | 42 | 102 | 44 | 45 | 67 | 389 |
| Glucose-stimulated insulin secretion or other functional measurement[e] | | | | | | | | |
| Handpicked to purity? Please select yes/no from drop down list | Yes | Yes | Yes | Yes | Yes | Yes | Yes | Yes |
| Additional notes | | | | | | | | |

| Islet preparation | 17 | 18 | 19 | 20 | 21 | 22 | 23 | 24 |
|---|---|---|---|---|---|---|---|---|
| **MANDATORY INFORMATION** | | | | | | | | |
| Unique identifier | SAMN12339206 | SAMN12534700 | SAMN40619409 | SAMN40709610 | SAMN41657868 | SAMN42008301 | SAMN48072591 | SAMN48430443 |
| Donor age (years) | 45 | 29 | 63 | 55 | 38 | 36 | 62 | 50 |
| Donor sex (M/F) | M | M | M | F | M | M | M | M |
| Donor BMI ($kg/m^2$) | 32.9 | 26 | 34.1 | 28.6 | 25.5 | 30 | 31.4 | 25.8 |
| Donor $HbA_{1c}$ or other measure of blood glucose control | 36 mmol / mol (5.4%) | 43 mmol / mol (6.1%) | 32 mmol / mol (5.1%) | 39 mmol / mol (5.7%) | 36 mmol / mol (5.4%) | 31 mmol / mol (5%) | 34 mmol / mol (5.3%) | 33 mmol / mol (5.2%) |
| Origin/source of islets[b] | IIDP | IIDP | IIDP | IIDP | IIDP | IIDP | IIDP | IIDP |
| Islet isolation centre | SC-ICRC | U. Pennsylvania | Prodo Labs | U. Miami | U. Wisconsin | U. Pennsylvania | Prodo Labs | Prodo Labs |
| Donor history of diabetes? Please select yes/no from drop down list | | | | | | | | |
| **If Yes, complete the next two lines if this information is available** | | | | | | | | |
| Diabetes duration (years) | | | | | | | | |
| Glucose-lowering therapy at time of death[c] | | | | | | | | |
| **RECOMMENDED INFORMATION** | | | | | | | | |
| Donor cause of death | CV / Stroke | Anoxia | Anoxia | CV / Stroke | Head Trauma | Anoxia | CV / Stroke | Head Trauma |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Warm ischaemia time (h) | | | | | | | | |
| Cold ischaemia time (h) | 6.5 | 18.5 | 11.2 | 11.8 | 4 | 14 | 12 | 10 |
| Estimated purity (%) | 85 | 90 | 90 | 85 | 90 | 80 | 90 | 90 |
| Estimated viability (%) | 96 | 93 | 95 | 92 | 98 | 97 | 95 | 95 |
| Total culture time (h)[d] | 392 | 384 | 441 | 382 | 403 | 407 | 425 | 427 |
| Glucose-stimulated insulin secretion or other functional measurement[e] | | | | | | | | |
| Handpicked to purity? Please select yes/no from drop down list | Yes | Yes | Yes | Yes | Yes | Yes | Yes | Yes |
| Additional notes | | | | | | | | |

[c]Please specify the therapy/therapies
[d]Time of islet culture at the isolation centre, during shipment and at the receiving laboratory
[e]Please specify the test and the results