

# **CIT ISLET-ALONE PROTOCOLS**

## **CIT-04: LABORATORY MANUAL FOR CENTRAL LABORATORY ASSESSMENTS**

**VERSION 7.0  
JANUARY 18, 2012**

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## 1. ADDITIONAL STUDY CONTACT INFORMATION

|   |   |   |
|---|---|---|
| <b>PPD Development</b><br>Attn: Maria Edwards<br>2244 Dabney Rd<br>Richmond, VA 23230<br>Phone: 804-359-1900<br>Fax: 804-253-1104 | <b>Tandem Labs</b><br>Attn: Dania Yaskanin<br>115 Silvia Street<br>West Trenton, NJ 08628<br>Phone: 609-228-0243<br>Fax: 609-434-1028 | <b>Protocol Coordinator</b><br>Cynthia Diltz, RN, BSN, CCRC<br>University of Iowa<br>Department of Biostatistics<br>Clinical Trials Statistical and Data<br>Management Center<br>2400 University Capitol Centre<br>Iowa City, IA USA 52242-5500<br>Phone: +1-319-353-4982<br>Fax: +1-319-353-3960<br><a href="mailto:cynthia-diltz@uiowa.edu">cynthia-diltz@uiowa.edu</a> |
|---|---|---|

## 2. CIT-04 CENTRAL LABORATORY SPECIMEN SCHEDULE

| Central Laboratory Assessments                         |                            |  |              |   |  |
|--|----------------------------|--|--------------|---|--|
| Assessment   | Laboratory                 | Visit / Time-point   | Volume       | Collection Container                            | Shipping Instructions  |
| Albumin/Creatinine Ratio                               | University of Minnesota    | V08,10,21<br>(Subs Tx Day 75 <sup>3</sup> , Day 365) <sup>2</sup><br>[V56, 63] <sup>6</sup>  | 5 mL Urine   | Sterile Urine Container                         | Ship in batches weekly, frozen on dry ice. Ship on Monday – Thursday.  |
| Alloantibodies   | University of Pennsylvania | V03,10,15,18,21<br>(V24,27,30) <sup>1</sup><br>(Subs Tx Day 0, 75 <sup>3</sup> , 365) <sup>2</sup><br>[V56, 63] <sup>6</sup>   | 2 mL Blood   | (1) 3-mL Red-top Vacutainer                     | Ship on dry ice in batches at least quarterly. Ship Monday – Thursday only.  |
| Atherogenic Profile                                    | University of Washington   | V21<br>(Sub Tx Day 365) <sup>2</sup><br>[V63] <sup>6</sup>   | 8.5 mL Blood | (1) 8.5 mL Gold SST                             | Ship on dry ice in batches at least weekly. Ship Monday – Thursday only.   |
| Autoantibodies   | Barbara Davis Center       | V10,15,18,21<br>(V24,27,30) <sup>1</sup><br>(Subs Tx Day 75 <sup>3</sup> , 365) <sup>2</sup><br>[V56, 63] <sup>6</sup>   | 2 mL Blood   | (1) 3-mL Red-top OR<br>(1) 3.5 mL Tiger top SST | Ship on dry ice in batches at least quarterly. Ship Monday – Wednesday only. If collected on Thursday or Friday, freeze serum at -20°C or -70°C until Monday, Tuesday, or Wednesday. |
| Belatacept Trough Level                                | PPD Development            | V04,05,06,07,08,09,10,11,12,13,14,15,16,17,18,19,20,21<br>(V22,23,24,25,26,27,28,29,30) <sup>1</sup><br>(Subs Tx Day 4, 7, 75 <sup>3</sup> , 365) <sup>2</sup><br>[V51,52,53,54,55,56,57,58,59,60,61,62,63] <sup>6</sup> | 2 mL Blood   | (1) 3.5 mL Gold SST                             | Ship in batches bi-monthly on dry ice. Ship Monday – Thursday.   |
| Fasting serum glucose and c-peptide / serum creatinine | University of Washington   | V08,09,10 <sup>1</sup> ,12,13,14,15,16,17,18,19,20,21<br>(V22,23,24,25,26,27,28,29,30) <sup>1</sup><br>(Subs Tx Day 75 <sup>3</sup> , 365) <sup>2</sup><br>[V56,63,64] <sup>6</sup>                                      | 2 mL Blood   | (1) 3.5-mL Gold SST                             | Ship on dry ice in batches at least weekly. Ship Monday – Thursday only.   |

| Central Laboratory Assessments                     |   |  |   |   |  |
|--|---|--|---|---|--|
| Assessment   | Laboratory                                      | Visit / Time-point   | Volume  | Collection Container                        | Shipping Instructions  |
| GFR  | University of Minnesota                         | V08,10,21<br>(Subs Tx Day 75 <sup>3</sup> , 365) <sup>2</sup><br>[V63] <sup>6</sup>  | 10 mL Blood<br>2 mL each at 120, 150, 180, 210<br>and 240 minutes   | (5) 2-mL Na<br>Heparin Tube                 | Ship in batches weekly on dry ice. Ship<br>Monday – Thursday                   |
| Hemoglobin A1c<br>(HbA1c)                          | University of Washington                        | V10,15,18,21,<br>(V24,27,30) <sup>1</sup><br>(Subs Tx Day 75 <sup>3</sup> , 365) <sup>2</sup><br>[V53, 56, 60, 63, 64] <sup>6</sup>      | 2 mL Blood  | (1) 2-mL Lavender<br>top EDTA<br>Vacutainer | Ship on cold pack within 24 hours of<br>collection. Ship Monday-Thursday only. |
| Immunogenicity<br>Sample                           | Tandem Labs                                     | V02,14,21<br>(V27) <sup>1</sup><br>(Subs Tx Day 365) <sup>2</sup><br>[V63, 64] <sup>6</sup>  | 4 mL Blood  | (1) 4-mL Gold SST                           | Ship in batches quarterly on dry ice. Ship<br>Monday – Wednesday               |
| Insulin Modified<br>FSIGT                          | University of Washington                        | V10 <sup>4</sup> ,21<br>(Subs Tx Day 75 <sup>3</sup> , 365) <sup>2</sup>   | 48 mL Total Blood<br>2 mL each at -10, -5, and -1<br>minutes pre-injection of glucose<br>2 mL each at 1, 2, 3, 4, 5, 7, 10, 12,<br>14, 16, 18, 20, 22, 25, 30, 40, 50,<br>70, 100, 140, and 180 minutes post<br>injection of glucose  | (24) 3.5-mL Gold<br>SST                     | Ship on dry ice in batches at least weekly.<br>Ship Monday – Thursday only.    |
| MMTT:<br>Stimulated serum<br>glucose and c-peptide | University of Washington                        | V10 <sup>4</sup> ,15,18,21<br>(V24,27,30) <sup>1</sup><br>(Subs Tx Day 75 <sup>3</sup> , 365) <sup>2</sup><br>[V56, 63, 64] <sup>6</sup> | 4 mL Total Blood<br><ul style="list-style-type: none"> <li>• 2 mL at 60 minutes (<i>ONLY if checking for graft failure</i>)</li> <li>• 2 mL at 90 minutes</li> </ul> (Note: the Fasting serum glucose<br>and c-peptide/serum creatinine is<br>the 0 hour sample for the MMTT) | (2) 3.5-mL Gold<br>SST                      | Ship on dry ice in batches at least weekly.<br>Ship Monday – Thursday only.    |
| PBMC / Plasma to<br>Archive                        | ITN Central Cell<br>Processing Core<br>Facility | V10,15,18,21<br>(V24,27,30) <sup>1</sup><br>(Subs Tx Day 75 <sup>3</sup> , 365) <sup>2</sup>   | 30 mL Blood   | (3) 10-mL Na<br>Heparin Tube                | Ship ambient daily.  |
| RNA to Archive                                     | Expression<br>Analysis                          | V10,15,18,21<br>(V24,27,30) <sup>1</sup><br>(Subs Tx Day 75 <sup>3</sup> , 365) <sup>2</sup>   | 9 mL Blood  | (3) 3-mL Tempus<br>RNA Tube                 | Ship in batches quarterly on dry ice.  |

| Central Laboratory Assessments |                       |  |   |   |  |
|--------------------------------|-----------------------|--|---|---|--|
| Assessment                     | Laboratory            | Visit / Time-point   | Volume  | Collection Container                        | Shipping Instructions  |
| Serum to Archive               | NIDDK Repository      | V10,15,18,21<br>(V24,27,30) <sup>1</sup><br>(Subs Tx Day 75 <sup>3</sup> , 365) <sup>2</sup> | 4 mL Blood  | (1) 4-mL Gold SST                           | Ship in batches at least quarterly.                              |
| TAT Complex, C3a,<br>c-peptide | Rudbeck<br>Laboratory | V03 <sup>5</sup><br>(Subs Tx Day 0) <sup>2</sup>   | 10 mL Total Blood @ V03<br>2 mL at Pre-immunosuppression,<br>immediately pre-tx, 15, 60,<br>180 min post-tx | (5) 2-mL Lavender<br>top EDTA<br>Vacutainer | Ship on dry ice (minimum 5 kg) in batches<br>at least quarterly. |

<sup>1</sup>Visits as needed for subsequent islet transplant(s). See Appendix 2 – Year Two Schedule of Events

<sup>2</sup>Endpoint assessments for subsequent transplant(s). See Appendix 3 – Subsequent Transplant Schedule of Events

<sup>3</sup>If third transplant occurs less than 75 days after the second transplant, the 75 day endpoint data for the second transplant will not be collected

<sup>4</sup>Do not collect these samples at Day 75 for subjects with confirmed graft failure

<sup>5</sup>Collect these samples beginning on Day -2 (pre-IS) for Visit 03

<sup>6</sup>1-Year Additional Follow-up

### 3. THROMBIN-ANTITHROMBIN COMPLEX (TAT), C3a, AND C-PEPTIDE

- Refer to the General Lab manual for instructions regarding blood draws, processing and shipping
- Subjects randomized to CIT-04 do not receive immunosuppression on Day -2. For those subjects, the Pre Immunosuppression sample, labeled Pre IS, is drawn on day 0 prior to the administration of Belatacept, Daclizumab or MMF; the Pre-Transplant sample, labeled Pre-Tx, is drawn immediately prior to the start of the islet infusion

### 4. BELATACEPT TROUGH LEVEL

- Complete test **prior to** Belatacept infusion.
- Collect 2 ml Blood into 3.5 ml Gold Serum Separator (SST) tube
- Allow to clot at room temperature for 15 – 30 minutes
- Centrifuge for 15 minutes at 1000 xg (approximately 3000 RPM) in a refrigerated centrifuge
- Transfer at least 0.5 ml serum into a purple cap cryovial
- Immediately freeze at -20° C or colder
- Follow packing instructions outlined in section 4 of General Lab Manual
- Ship samples monthly on dry ice. If shipping from Canada to U.S., please complete an Expanded Service International Air Waybill (With Dry Ice) (Appendix 5 in General Lab Manual)
- On the day of the shipment, print the Specimen List by clicking the View Shipping Report button in the Specimen Tracking System and place a copy in the shipping box. NOTE: you will need to expand the columns in the specimen list to show all the data before printing. An automatic email will be sent to:
  - [Susan.Steever@bms.com](mailto:Susan.Steever@bms.com)
  - [MG-PK-Manifest@bms.com](mailto:MG-PK-Manifest@bms.com)
  - [maria.edwards@richmond.ppd.com](mailto:maria.edwards@richmond.ppd.com)
  - [Kathy.zelinsky@bms.com](mailto:Kathy.zelinsky@bms.com)
  - [Mehmooda.shaikh@bms.com](mailto:Mehmooda.shaikh@bms.com)
- If you are unable to print the Specimen List from the Specimen Tracking System on the day of shipment, complete a Specimen Submission Form and fax a copy of the form including the airbill tracking number to the laboratory at +1-804-253-1104.
- Shipments must be made Monday through Thursday.

PPD Development  
Attn: Maria Edwards  
2244 Dabney Rd  
Richmond, VA 23230  
Tel: 804-359-1900  
Fax: 804-253-1104

## 5. IMMUNOGENICITY SAMPLE

- Collect immunogenicity samples **prior to** belatacept infusion. For subjects who discontinue belatacept, collect additional immunogenicity samples at 4 weeks and 8 weeks post last dose.
- Collect 4 ml Blood into 4ml Gold Serum Separator (SST) tube
- Allow to clot at room temperature for 15 – 30 minutes
- Centrifuge for 15 minutes at 1000 xg (approximately 3000 RPM) in a refrigerated centrifuge
- Transfer at least 2 ml serum into a white cap cryovial
- Immediately freeze at -20° C or colder
- Follow packing instructions outlined in section 4 of General Lab Manual
- Ship samples quarterly on dry ice. If shipping from Canada to U.S., please complete an Expanded Service International Air Waybill (With Dry Ice) (Appendix 5 in General Lab Manual)
- On the day of the shipment, print the Specimen List by clicking the View Shipping Report button in the Specimen Tracking System and place a copy in the shipping box. NOTE: you will need to expand the columns in the specimen list to show all the data before printing. An automatic email will be sent to:  
[Susan.Steever@bms.com](mailto:Susan.Steever@bms.com)  
[zoe.tzogas@bms.com](mailto:zoe.tzogas@bms.com)  
[Leec4@labcorp.com](mailto:Leec4@labcorp.com)  
[albanej@labcorp.com](mailto:albanej@labcorp.com)  
[mcgrata@labcorp.com](mailto:mcgrata@labcorp.com)  
[Mehmooda.shaikh@bms.com](mailto:Mehmooda.shaikh@bms.com)  
[Yaskand@Labcorp.com](mailto:Yaskand@Labcorp.com)
- If you are unable to print the Specimen List from the Specimen Tracking System on the day of shipment, complete a Specimen Submission Form and fax a copy of the form including the airbill tracking number to the laboratory at +1-804-253-1104.
- Shipments must be made Monday through Wednesday.

Tandem Labs  
Attn: Dania Yaskanin  
115 Silvia Street  
West Trenton, NJ 08628  
Phone: 609.228.0243  
Fax: 609.434.1028

**Note: On visits 14 and 21 you will need to draw both the Belatacept Trough Level and the Immunogenicity sample. On these 2 visits you can draw 5 mL of blood in 1 – 5mL SST tube. Then follow the processing directions in sections 3&4, aliquot 0.5 mL serum in the purple cap cryovial and 2 mL into the white cap cryovial. Shipper above instructions.**

## 6. CIT-04 KIT COMPONENTS

|   |                      |   |   |
|---|----------------------|---|---|
| <p><b>VISIT 03</b><br/><b>Day 0</b></p>   | <p><b>KIT #3</b></p> | <p><b>TAT, C3a, C-Peptide</b><br/>(5) 2-mL EDTA Vacutainer Tube<br/>(5) 1.8-mL Cryogenic Vial<br/><b>Alloantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial</p>  | <p><b>Immunogenicity sample</b><br/>(1) 4-mL Gold SST<br/>(1) 4.0-mL white cap Cryogenic Vial</p>   |
| <p><b>VISIT 04 – 07</b><br/><b>Day 4, 7, 14, 21</b><br/><b>[Subs Tx Day 4, 7]</b><br/><b>(if needed)</b></p>              | <p><b>KIT #4</b></p> | <p><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |   |
| <p><b>VISIT 08</b><br/><b>Day 28</b></p>  | <p><b>KIT #5</b></p> | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials<br/><b>Albumin/Creatinine Ratio</b><br/>(1) Urine Specimen Container<br/>(1) 4.0-mL Cryogenic Vial</p>  | <p><b>GFR</b><br/>(5) 2-mL Na Heparin Tubes<br/>(5) 1.8-ml Cryogenic Vials<br/><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |
| <p><b>VISIT 09</b><br/><b>Days 56</b></p>   | <p><b>KIT #6</b></p> | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials</p>   | <p><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |
| <p><b>VISIT 10</b><br/><b>Day 75</b><br/><b>Secondary Endpoint</b><br/><b>[Subs Tx Day 75]</b><br/><b>(if needed)</b></p> | <p><b>KIT #7</b></p> | <p><b><sup>1</sup>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials<br/><b><sup>1,2</sup>MMTT : Stimulated Glucose and C-peptide</b><br/>(2) 3.5-mL Gold SST<br/>(4) 1.8-mL Cryogenic Vials<br/><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube<br/><b>Albumin/Creatinine Ratio</b><br/>(1) Urine Specimen Container<br/>(1) 4.0-mL Cryogenic Vial<br/><b><sup>1</sup>FSIGT</b><br/>(24) 3.5-mL Gold SST<br/>(58) 1.8-mL Cryogenic Vials</p> | <p><b>GFR</b><br/>(5) 2-mL Na Heparin Tubes<br/>(5) 1.8 ml Cryogenic Vials<br/><b>Alloantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Autoantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Serum to Archive</b><br/>(1) 4-mL Gold SST Tube<br/>(3) 1.8-mL Cryogenic Vials<br/><b>RNA to Archive (NIDDK)</b><br/>(3) 3-mL Tempus RNA Tube<br/><b>PBMC and Plasma to Archive</b><br/>(3) 10-mL Na Heparin Vacutainer tubes<br/><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p> |
| <p><b>VISIT 11</b><br/><b>Day 84</b></p>  | <p><b>KIT #4</b></p> | <p><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |   |

|   |                      |   |  |
|---|----------------------|---|--|
| <p><b>VISIT 12,13</b><br/><b>Day 112, 140</b></p>   | <p><b>KIT #6</b></p> | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials</p>   | <p><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>   |
| <p><b>VISIT 14</b><br/><b>Day 168</b></p>           | <p><b>Kit #8</b></p> | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials</p>   | <p><b>Belatacept Trough Level /Immunogenicity sample</b><br/>(1) 5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial<br/>(1) 4.0-mL white cap Cryogenic Vial</p>  |
| <p><b>VISIT 15</b><br/><b>Day 196</b></p>           | <p><b>KIT #9</b></p> | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials<br/><b><sup>2</sup>MMTT: Stimulated Glucose and C-peptide</b><br/>(2) 3.5-mL Gold SST<br/>(4) 1.8-mL Cryogenic Vials<br/><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube</p> | <p><b>Alloantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Autoantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Serum to Archive</b><br/>(1) 4-mL Gold SST Vacutainer Tube<br/>(3) 1.8-mL Cryogenic Vials<br/><b>RNA to Archive (NIDDK)</b><br/>(3) 3-mL Tempus RNA Tube<br/><b>PBMC and Plasma to Archive</b><br/>(3) 10 mL Na Heparin Vacutainer tubes<br/><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p> |
| <p><b>VISIT 16, 17</b><br/><b>Days 224, 252</b></p> | <p><b>KIT #6</b></p> | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials</p>   | <p><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>   |
| <p><b>VISIT 18</b><br/><b>Day 280</b></p>           | <p><b>KIT #9</b></p> | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials<br/><b><sup>2</sup>MMTT: Stimulated Glucose and C-peptide</b><br/>(2) 3.5-mL Gold SST<br/>(4) 1.8-mL Cryogenic Vials<br/><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube</p> | <p><b>Alloantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Autoantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Serum to Archive</b><br/>(1) 4-mL Gold SST Vacutainer Tube<br/>(3) 1.8-mL Cryogenic Vials<br/><b>RNA to Archive (NIDDK)</b><br/>(3) 3-mL Tempus RNA Tube<br/><b>PBMC and Plasma to Archive</b><br/>(3) 10 mL Na Heparin Vacutainer tubes<br/><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p> |
| <p><b>VISIT 19, 20</b><br/><b>Days 308, 336</b></p> | <p><b>KIT #6</b></p> | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials</p>   | <p><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>   |

|   |                       |  |  |
|---|-----------------------|--|--|
| <p><b>VISIT 21</b><br/><b>Day 365 / Month 12</b><br/><i>Secondary Endpoint</i><br/><b>Subs Tx Day 365 (if needed)</b></p> | <p><b>KIT #10</b></p> | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials<br/><b><sup>2</sup>MMTT: Stimulated Glucose and C-peptide</b><br/>(2) 3.5-mL Gold SST<br/>(4) 1.8-mL Cryogenic Vials<br/><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube<br/><b>Albumin/Creatinine Ratio</b><br/>(1) Urine Specimen Container<br/>(1) 4.0-mL Cryogenic Vial<br/><b>FSIGT</b><br/>(24) 3.5-mL Gold SST<br/>(58) 1.8-mL Cryogenic Vials<br/><b>Atherogenic Profile</b><br/>(1) 8.5 mL Gold SST<br/>(4) 1.8 mL Cryogenic Vials</p> | <p><b>GFR</b><br/>(5) 2-mL Green-top Vacutainer Tubes<br/>(5) 1.8-ml Cryogenic Vials<br/><b>Alloantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Autoantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Serum to Archive</b><br/>(1) 4-mL Gold SST Vacutainer Tube<br/>(3) 1.8-mL Cryogenic Vials<br/><b>RNA to Archive (NIDDK)</b><br/>(3) 3-mLTempus RNA Tube<br/><b>PBMC and Plasma to Archive</b><br/>(3) 10-mL Na Heparin Vacutainer tubes<br/><b>Belatacept Trough Level /Immunogenicity sample</b><br/>(1) 5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial<br/>(1) 4.0-mL white cap Cryogenic Vial</p> |
| <p><b>VISIT 22, 23</b><br/><b>Day 392, 420</b></p>  | <p><b>KIT #6</b></p>  | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials</p>  | <p><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1)1.8-mL purple cap Cryogenic Vial</p>  |
| <p><b>VISIT 24</b><br/><b>Day 448</b></p>   | <p><b>KIT #9</b></p>  | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials<br/><b><sup>2</sup>MMTT: Stimulated Glucose and C-peptide</b><br/>(2) 3.5-mL Gold SST<br/>(4) 1.8-mL Cryogenic Vials<br/><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube</p>  | <p><b>Alloantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Autoantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Serum to Archive</b><br/>(1) 4-mL Gold SST Vacutainer Tube<br/>(3) 1.8-mL Cryogenic Vials<br/><b>RNA to Archive (NIDDK)</b><br/>(3) 3-mLTempus RNA Tube<br/><b>PBMC and Plasma to Archive</b><br/>(3) 10 mL Na Heparin Vacutainer tubes<br/><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |
| <p><b>VISIT 25, 26</b><br/><b>Day 476, 504</b></p>  | <p><b>KIT #6</b></p>  | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials</p>  | <p><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1)1.8-mL purple cap Cryogenic Vial</p>  |

|   |                       |   |   |
|---|-----------------------|---|---|
| <p><b>VISIT 27</b><br/><br/><b>Day 532</b></p>  | <p><b>Kit #11</b></p> | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials<br/><b><sup>2</sup>MMTT: Stimulated Glucose and C-peptide</b><br/>(2) 3.5-mL Gold SST<br/>(4) 1.8-mL Cryogenic Vials<br/><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube</p> | <p><b>Alloantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Autoantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Serum to Archive</b><br/>(1) 4-mL Gold SST Vacutainer Tube<br/>(3) 1.8-mL Cryogenic Vials<br/><b>RNA to Archive (NIDDK)</b><br/>(3) 3-mL Tempus RNA Tube<br/><b>PBMC and Plasma to Archive</b><br/>(3) 10-mL Na Heparin Vacutainer tubes<br/><b>Belatacept Trough Level /Immunogenicity sample</b><br/>(1) 5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial<br/>(1) 4.0-mL white cap Cryogenic Vial</p> |
| <p><b>VISIT 28, 29</b><br/><br/><b>Day 560, 588</b></p>                                   | <p><b>KIT #6</b></p>  | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials</p>   | <p><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |
| <p><b>VISIT 30</b><br/><br/><b>Day 616</b></p>  | <p><b>KIT #9</b></p>  | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials<br/><b><sup>2</sup>MMTT: Stimulated Glucose and C-peptide</b><br/>(2) 3.5-mL Gold SST<br/>(4) 1.8-mL Cryogenic Vials<br/><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube</p> | <p><b>Alloantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Autoantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Serum to Archive</b><br/>(1) 4-mL Gold SST Vacutainer Tube<br/>(3) 1.8-mL Cryogenic Vials<br/><b>RNA to Archive (NIDDK)</b><br/>(3) 3-mL Tempus RNA Tube<br/><b>PBMC and Plasma to Archive</b><br/>(3) 10 mL Na Heparin Vacutainer tubes<br/><b>Belatacept Trough Level</b><br/>(1) 3.5 mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |
| <p><b>Visit 51, 52</b><br/><b>M13 (post final tx),</b><br/><b>M14 (post final tx)</b></p> | <p><b>KIT #4</b></p>  | <p><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |   |
| <p><b>Visit 53</b><br/><b>M15 (post final tx)</b></p>                                     | <p><b>KIT #13</b></p> | <p><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  | <p><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube</p>   |
| <p><b>Visit 54, 55</b><br/><b>M16 (post final tx),</b><br/><b>M17 (post final tx)</b></p> | <p><b>KIT #4</b></p>  | <p><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |   |

|  |                       |   |  |
|--|-----------------------|---|--|
| <p><b>Visit 56</b><br/><b>M18 (post final tx)</b></p>  | <p><b>KIT #14</b></p> | <p><b>Albumin/Creatinine Ratio</b><br/>(1) Urine Specimen Container<br/>(1) 4.0-mL Cryogenic Vial<br/><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube<br/><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials<br/><b><sup>2</sup>MMTT: Stimulated Glucose and C-peptide</b><br/>(2) 3.5-mL Gold SST<br/>(4) 1.8-mL Cryogenic Vials<br/><b>Alloantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Autoantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial</p>   |
| <p><b>Visit 57, 58, 59</b><br/><b>M19 (post final tx),</b><br/><b>M20 (post final tx)</b><br/><b>TBD (extra visit)</b></p> | <p><b>KIT #4</b></p>  | <p><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |  |
| <p><b>Visit 60</b><br/><b>M21 (post final tx)</b></p>  | <p><b>KIT #13</b></p> | <p><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  | <p><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube</p>  |
| <p><b>Visit 61, 62</b><br/><b>M22 (post final tx),</b><br/><b>M23 (post final tx)</b></p>                                  | <p><b>KIT #4</b></p>  | <p><b>Belatacept Trough Level</b><br/>(1) 3.5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial</p>  |  |
| <p><b>Visit 63</b><br/><b>M24 (post final tx)</b></p>  | <p><b>KIT #15</b></p> | <p><b>Albumin/Creatinine Ratio</b><br/>(1) Urine Specimen Container<br/>(1) 4.0-mL Cryogenic Vial<br/><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube<br/><b>GFR</b><br/>(5) 2-mL Green-top Vacutainer Tubes<br/>(5) 1.8-ml Cryogenic Vials<br/><b>Atherogenic Profile</b><br/>(1) 8.5 mL Gold SST<br/>(4) 1.8 mL Cryogenic Vials<br/><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials</p> | <p><b><sup>2</sup>MMTT: Stimulated Glucose and C-peptide</b><br/>(2) 3.5-mL Gold SST<br/>(4) 1.8-mL Cryogenic Vials<br/><b>Alloantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Autoantibody</b><br/>(1) 3-mL Red-top Vacutainer Tube<br/>(1) 1.8 mL Cryogenic Vial<br/><b>Belatacept Trough Level /Immunogenicity sample</b><br/>(1) 5-mL Gold SST<br/>(1) 1.8-mL purple cap Cryogenic Vial<br/>(1) 4.0-mL white cap Cryogenic Vial</p> |
| <p><b>Visit 64</b><br/><b>Y2 (post initial tx)</b></p>   | <p><b>KIT#16</b></p>  | <p><b>HBA1C</b><br/>(1) 2-mL EDTA Vacutainer Tube<br/><b>Immunogenicity sample</b><br/>(1) 4-mL Gold SST<br/>(1) 4.0-mL white cap Cryogenic Vial</p>  | <p><b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br/>(1) 3.5-mL Gold SST<br/>(2) 1.8-mL Cryogenic Vials<br/><b><sup>2</sup>MMTT: Stimulated Glucose and C-peptide</b><br/>(2) 3.5-mL Gold SST<br/>(4) 1.8-mL Cryogenic Vials</p>   |

|   |                 |  |  |
|---|-----------------|--|--|
| <b>4 or 8 weeks post last dose of Belatacept</b>                    | <b>KIT #12</b>  | <b>Immunogenicity sample _____ week post last dose</b><br>(1) 4-mL Gold SST<br>(1) 4.0-mL white cap Cryogenic Vial<br><b>(note which visit on the requisition)</b>   |  |
| <b>Reduced Follow-Up<br/>(Year 1 and 2 post Initial transplant)</b> | <b>KIT #50</b>  | <b>90 min C-peptide post MMTT, Serum Creatinine</b><br>(1) 3.5-mL Gold SST<br>(2) 1.8-mL Cryogenic Vials   | <b>HBA1C</b><br>(1) 2-mL EDTA Vacutainer Tube<br><b>Alloantibody</b><br>(1) 3-mL Red-top Vacutainer Tube<br>(1) 1.8-mL Cryogenic Vial  |
| <b>Reduced Follow-Up<br/>(Monthly and Quarterly)</b>                | <b>KIT #50X</b> | <b>Alloantibody</b><br>(1) 3-mL Red-top Vacutainer Tube<br>(1) 1.8-mL Cryogenic Vial   |  |
| <b>Subs Tx Day 0<br/>(if needed)</b>                                | <b>KIT #51</b>  | <b>TAT, C3a, C-Peptide</b><br>(5) 2-mL EDTA Vacutainer Tube<br>(5) 1.8-mL Cryogenic Vial   | <b>Alloantibody</b><br>(1) 3-mL Red-top Vacutainer Tube<br>(1) 1.8 mL Cryogenic Vial   |
| <b>Suspected Graft Failure</b>                                      | <b>Kit 50Z</b>  | <b>Fasting Serum Glucose, C-Peptide, Creatinine</b><br>(1) 3.5-mL Gold SST<br>(2) 1.8-mL Cryogenic Vials<br><b>MMTT: Stimulated Glucose and C-peptide</b><br>(2) 3.5-mL Gold SST<br>(4) 1.8-mL Cryogenic Vials | <b>Alloantibody</b><br>(1) 3-mL Red-top Vacutainer Tube<br>(1) 1.8 mL Cryogenic Vial<br><b>Autoantibody</b><br>(1) 3-mL Red-top Vacutainer Tube<br>(1) 1.8 mL Cryogenic Vial |

<sup>1</sup>Do not collect these samples at Day 75 for subjects with confirmed graft failure

<sup>2</sup>The kit contains two 3.5mL Gold SST tubes for the MMTT. Only one of these two tubes will be filled (at 90 minutes), unless it is suspected that the subject has suffered graft failure (in which case, the second tube should be filled at 60 minutes). If there is no suspicion of graft failure, the 60 minute tube and aliquots associated with the 60 minute draw can be discarded

See Appendix 1 for Kit Supply Order Form

### 6. CIT-04 MAXIMUM RESEARCH BLOOD VOLUME TABLE

| CIT04 - MAXIMUM RESEARCH BLOOD VOLUME TABLE   |              |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
|---|--------------|--------------|-------------|------------|------------|-------------|-------------|-------------|-------------|-------------|--------------|------------|-------------|-------------|-------------|-------------|------------|------------|-------------|------------|------------|--------------|
| TIME POINTS/VISITS                            |              |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| TIMING OF STUDY PARTICIPATION                 | Days         |              |             |            |            | Weeks       |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
|   | SCRN         | BL           | TX 0        | 3          | 4          | 1           | 2           | 3           | 4           | 8           | (Day 75)     | 12         | 16          | 20          | 24          | 28          | 32         | 36         | 40          | 44         | 48         | 52           |
| VISIT   | 1            | 2            | 3           | 3a         | 4          | 5           | 6           | 7           | 8           | 9           | 10           | 11         | 12          | 13          | 14          | 15          | 16         | 17         | 18          | 19         | 20         | 21           |
| BLOOD VOLUMES                                 |              |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| LOCAL LABORATORY ASSESSMENTS                  |              |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| CBC (WBC + Diff & Plat)                       | 5            | 5            | 5           |            |            | 5           | 5           | 5           | 5           | 5           | 5            | 5          | 5           | 5           | 5           | 5           | 5          | 5          | 5           | 5          | 5          | 5            |
| Chemistry (P18 + Mg or P20)                   | 4            | 4            | 4           |            |            | 4           | 4           | 4           | 4           | 4           | 4            | 4          | 4           | 4           | 4           | 4           | 4          | 4          | 4           | 4          | 4          | 4            |
| Lipids  | 4            | 4            |             |            |            |             |             |             |             |             | 4            |            |             |             |             | 4           |            |            | 4           |            |            | 4            |
| Thyroid Function                              | 4            | 4            |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| Serum b-HCG (females)                         | 4            | 4            |             |            | 4          |             | 4           |             | 4           | 4           |              | 4          | 4           | 4           | 4           | 4           | 4          | 4          | 4           | 4          | 4          | 4            |
| PSA   | 4            | 4            |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            | 4            |
| Serology                                      | 7            | 7            |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| EBV IgG                                       | 2            |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| CMV IgG, CMV IgM                              |              | 4            |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| Coagulation (PT, PTT, INR)                    | 5            | 5            | 5           |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| Blood Type & HLA                              |              | 11           |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| Crossmatch                                    |              | 10           |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| PRA by flow cytometry (Sub Tx)                |              | 10           |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| Fasting and 2 post-prandial (1-3 hrs) c-pep   |              |              |             | 9          |            | 9           |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| CMV by PCR                                    |              | 4            |             |            |            |             |             |             |             |             | 4            |            |             |             | 4           |             |            |            |             |            |            |              |
| EBV py PCR                                    |              | 4            |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| CENTRAL LABORATORY AND METABOLIC ASSESSMENTS  |              |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| GFR (5 timed specimens/timept, 2 ml each)     | 10           | 10           |             |            |            |             |             |             | 10          |             | 10           |            |             |             |             |             |            |            |             |            |            | 10           |
| HbA1c (central + local)                       | 4            | 4            |             |            |            |             |             |             |             |             | 4            |            |             |             |             | 4           |            |            | 4           |            |            | 4            |
| Fasting glucose & c-pep / serum creatinine    | 2            | 2            |             |            |            |             |             | 2           | 2           | 2           | 2            | 2          | 2           | 2           | 2           | 2           | 2          | 2          | 2           | 2          | 2          | 2            |
| Insulin modified FSIGT (c-pep, insulin, gluc) |              | 48           |             |            |            |             |             |             |             |             | 48           |            |             |             |             |             |            |            |             |            |            | 48           |
| 60, 90 min c-pep, gluc (MMIT) / serum creat   | 4            |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| 90 min c-pep, gluc (MMIT) / serum creat       |              |              |             |            |            |             |             |             |             |             | 2            |            |             |             |             | 2           |            |            | 2           |            |            | 2            |
| Belatacept Trough Level                       |              |              |             |            | 2          | 2           | 2           | 2           | 2           | 2           | 2            | 2          | 2           | 2           | 2           | 2           | 2          | 2          | 2           | 2          | 2          | 2            |
| Atherogenic Profile                           |              | 8.5          |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            | 8.5          |
| CENTRAL MECHANISTIC ASSAYS                    |              |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| Alloantibody                                  | 2            | 2            |             |            |            |             |             |             |             |             | 2            |            |             |             |             | 2           |            |            | 2           |            |            | 2            |
| Autoantibody                                  |              | 2            |             |            |            |             |             |             |             |             | 2            |            |             |             |             | 2           |            |            | 2           |            |            | 2            |
| TAT, c-peptide & C3a                          |              |              | 10          |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| Immunogenicity samples                        |              |              | 4           |            |            |             |             |             |             |             |              |            |             | 4           |             |             |            |            |             |            |            | 4            |
| CENTRAL ARCHIVED SAMPLES                      |              |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| Serum   |              | 4            |             |            |            |             |             |             |             |             | 4            |            |             |             |             | 4           |            |            | 4           |            |            | 4            |
| PBMC & Plasma                                 |              | 30           |             |            |            |             |             |             |             |             | 30           |            |             |             |             | 30          |            |            | 30          |            |            | 30           |
| RNA   |              | 9            |             |            |            |             |             |             |             |             | 9            |            |             |             |             | 9           |            |            | 9           |            |            | 9            |
| <b>TOTALS (mls)</b>                           | <b>61.0</b>  | <b>199.5</b> | <b>28.0</b> | <b>9.0</b> | <b>6.0</b> | <b>20.0</b> | <b>15.0</b> | <b>11.0</b> | <b>27.0</b> | <b>17.0</b> | <b>132.0</b> | <b>6.0</b> | <b>17.0</b> | <b>17.0</b> | <b>16.0</b> | <b>74.0</b> | <b>8.0</b> | <b>8.0</b> | <b>74.0</b> | <b>8.0</b> | <b>8.0</b> | <b>148.5</b> |
| <b>BL - WK 6 TOTAL (mls)</b>                  | <b>315.5</b> |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |
| <b>YEAR TOTAL (mls)</b>                       | <b>910.0</b> |              |             |            |            |             |             |             |             |             |              |            |             |             |             |             |            |            |             |            |            |              |

## Appendix 1: Kit Supply Order Form

Protocol CIT- 04: Please complete form and fax to University of Iowa @+1-319-353-3960

|                   |       |                    |       |
|-------------------|-------|--------------------|-------|
| Site Name & #:    | _____ | CIT Protocol:      | _____ |
| Shipping Address: |       |                    |       |
| Order Date:       | _____ | Due Date @ Site:   | _____ |
| Requested By:     | _____ | Requestor's phone: | _____ |
| Requestor's FAX:  | _____ | Requestor's email: | _____ |

| Kit(s) #       | QUANTITY |
|----------------|----------|
| Kit(s) # _____ | _____    |

You will receive an initial supply of kits for 10 participants upon notice of your site activation. The initial supply of kits will include (1) Kit #1 through Kit # 5, per subject.

**Please check your kits' expiration dates and DO NOT order more than a 6 month supply of kits.**

## Appendix 2: Year One Schedule of Events

### Appendix 1: Year One Schedule of Events

| Time points (in days relative to transplant) | SCR            | WL / BL <sup>1</sup> | 0 <sup>2</sup> | 3   | 4              | 7    | 14             | 21   | 28             | 56             | 75   | 84             | 112            | 140            | 168            | 196            | 224            | 252            | 280            | 308            | 336            | 365            |   |
|--|----------------|----------------------|----------------|-----|----------------|------|----------------|------|----------------|----------------|------|----------------|----------------|----------------|----------------|----------------|----------------|----------------|----------------|----------------|----------------|----------------|---|
| Visit Number                                 | 01             | 02                   | 03             | 03a | 04             | 05   | 06             | 07   | 08             | 09             | 10   | 11             | 12             | 13             | 14             | 15             | 16             | 17             | 18             | 19             | 20             | 21             |   |
| Visit Windows (in days)                      | N/A            | N/A                  | N/A            | N/A | N/A            | +/-2 | +/-2           | +/-2 | +/-3           | +/-3           | +/-5 | +/-3           | +/-3           | +/-3           | +/-3           | +/-5           | +/-5           | +/-5           | +/-5           | +/-5           | +/-5           | +/-5           |   |
| Equivalent Week                              | N/A            | N/A                  | N/A            | N/A | N/A            | W1   | W2             | W3   | W4             | W8             | N/A  | W12            | W16            | W20            | W24            | W28            | W32            | W36            | W40            | W44            | W48            | W52            |   |
| <b>GENERAL ASSESSMENTS</b>                   |                |                      |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                |                |   |
| Informed Consent                             | X <sup>3</sup> | X <sup>4</sup>       |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                |                |   |
| Med/Diabetes Hx & Demographics               | X              |                      |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                |                |   |
| Eval of Inclusion / Exclusion                | X              | X                    |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                |                |   |
| Mammogram (females >35)                      | X              | X-yrly               |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                |                |   |
| Retinopathy Evaluation <sup>5</sup>          | X              | X-yrly <sup>6</sup>  |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                | X              |   |
| Physical Exam                                | X              | X-yrly               | X              |     |                | X    | X              | X    | X              | X              | X    |                | X              | X              |                | X              |                | X              |                | X              |                | X              |   |
| QOL  |                | X-q <sup>3</sup> mo  |                |     |                |      |                |      |                |                | X    |                |                |                |                | X              |                |                | X              |                |                | X              |   |
| Chest X-Ray                                  | X              | X-yrly               |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                | X              |   |
| Abdominal US (Pelvis/Liver)                  | X              | X-yrly               |                |     |                | X    |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                | X              |   |
| ECG  | X              | X-yrly               |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                | X              |   |
| Cardiac Stress Test or Angiogram             | X              |                      |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                |                |   |
| PPD  | X              | X-yrly               |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                | X              |   |
| AE/Hypo Event/Toxicity Assess                |                | X                    | X              |     | X              | X    | X              | X    | X              | X              | X    | X              | X              | X              | X              | X              | X              | X              | X              | X              | X              | X              | X |
| <b>LOCAL LABORATORY ASSESSMENTS</b>          |                |                      |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                |                |   |
| CBC (WBC + Diff & Plat)                      | X              | X-q <sup>6</sup> mo  | X              |     |                | X    | X              | X    | X              | X              | X    |                | X              | X              |                | X              |                |                | X              |                |                | X              |   |
| Chemistry <sup>7</sup>                       | X              | X-yq <sup>6</sup> mo | X              |     |                | X    | X              | X    | X              | X              | X    |                | X              | X              |                | X              |                |                | X              |                |                | X              |   |
| Lipids                                       | X              | X-q <sup>6</sup> mo  |                |     |                |      |                |      |                |                | X    |                |                |                |                | X              |                |                | X              |                |                | X              |   |
| Thyroid Function (TSH)                       | X              | X-yrly               |                |     |                |      |                |      |                |                |      |                |                |                |                |                |                |                |                |                |                |                |   |
| Pregnancy test (WOCBP)                       | X              | X <sup>8</sup>       |                |     | X <sup>9</sup> |      | X <sup>9</sup> |      | X <sup>9</sup> | X <sup>9</sup> |      | X <sup>9</sup> |   |

| Time points (in days relative to transplant)         | SCR | WL/ BL               | 0 <sup>2</sup>  | 3   | 4   | 7    | 14   | 21   | 28   | 56   | 75   | 84   | 112  | 140  | 168  | 196  | 224  | 252  | 280  | 308  | 336  | 365             |
|--|-----|----------------------|-----------------|-----|-----|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|-----------------|
| Visit Number   | 01  | 02                   | 03              | 03a | 04  | 05   | 06   | 07   | 08   | 09   | 10   | 11   | 12   | 13   | 14   | 15   | 16   | 17   | 18   | 19   | 20   | 21              |
| Visit Windows (in days)                              | N/A | N/A                  | N/A             | N/A | N/A | +/-2 | +/-2 | +/-2 | +/-3 | +/-3 | +/-5 | +/-3 | +/-3 | +/-3 | +/-3 | +/-5 | +/-5 | +/-5 | +/-5 | +/-5 | +/-5 | +/-5            |
| Equivalent Week                                      | N/A | N/A                  | N/A             | N/A | N/A | W1   | W2   | W3   | W4   | W8   | N/A  | W12  | W16  | W20  | W24  | W28  | W32  | W36  | W40  | W44  | W48  | W52             |
| <b>LOCAL LABORATORY ASSESSMENTS (Con't)</b>          |     |                      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| Serology <sup>10</sup> (HepB/C,HIV)                  | X   | X-yrly               |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      | X               |
| EBV IgG  | X   |                      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| CMV IgG, CMV IgM                                     |     | X-yrly <sup>11</sup> |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      | X <sup>11</sup> |
| Coagulation (PT, PTT, INR)                           | X   | X-yrly               | X               |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| Blood Type   |     | X <sup>12</sup>      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| HLA  |     | X                    |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| Crossmatch   |     | X <sup>13</sup>      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| Fasting & post-prandial c-pep <sup>14</sup>          |     |                      |                 | X   |     | X    |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| Glucose (immediately post-bx)                        |     |                      | X <sup>15</sup> |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| PRA by flow cytometry                                |     | X <sup>16</sup>      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| CMV by PCR   |     | X                    |                 |     |     |      |      |      |      |      | X    |      |      |      | X    |      |      |      |      |      |      |                 |
| EBV by PCR <sup>17</sup>                             |     | X                    |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| <b>CENTRAL LABORATORY ASSESSMENTS</b>                |     |                      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| First morning spot urine <sup>18</sup>               | X   | X                    |                 |     |     |      |      |      | X    |      | X    |      |      |      |      |      |      |      |      |      |      | X               |
| GFR  | X   | X-yrly               |                 |     |     |      |      |      | X    |      | X    |      |      |      |      |      |      |      |      |      |      | X               |
| HbA1c  | X   | X-qsmo               |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X               |
| Fasting serum gluc/c-pep & creat <sup>19</sup>       | X   | X                    |                 |     |     |      |      |      | X    | X    | X    |      | X    | X    | X    | X    | X    | X    | X    | X    | X    | X               |
| Insulin modified FSIGT <sup>19</sup>                 |     | X-yrly <sup>6</sup>  |                 |     |     |      |      |      |      |      | X    |      |      |      |      |      |      |      |      |      |      | X               |
| 90 min <sup>20</sup> c-pep/gluc (MMIT) <sup>19</sup> | X   |                      |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      |      | X    |      | X               |
| Atherogenic Profile <sup>21</sup>                    |     | X                    |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      | X               |
| <b>LOCAL METABOLIC ASSESSMENTS</b>                   |     |                      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |                 |
| Glycemic Stability (CGMS) <sup>19</sup>              |     | X <sup>6</sup>       |                 |     |     |      |      |      |      |      | X    |      |      |      |      |      |      |      |      |      |      | X               |

| Time points (in days relative to transplant) | SCR | WL / BL <sup>1</sup> | 0 <sup>2</sup>  | 3   | 4   | 7    | 14   | 21   | 28   | 56   | 75   | 84   | 112  | 140  | 168  | 196  | 224  | 252  | 280  | 308  | 336  | 365  |
|--|-----|----------------------|-----------------|-----|-----|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|
| Visit Number                                 | 01  | 02                   | 03              | 03a | 04  | 05   | 06   | 07   | 08   | 09   | 10   | 11   | 12   | 13   | 14   | 15   | 16   | 17   | 18   | 19   | 20   | 21   |
| Visit Windows (in days)                      | N/A | N/A                  | N/A             | N/A | N/A | +/-2 | +/-2 | +/-2 | +/-3 | +/-3 | +/-5 | +/-3 | +/-3 | +/-3 | +/-3 | +/-5 | +/-5 | +/-5 | +/-5 | +/-5 | +/-5 | +/-5 |
| Equivalent Week                              | N/A | N/A                  | N/A             | N/A | N/A | W1   | W2   | W3   | W4   | W8   | N/A  | W12  | W16  | W20  | W24  | W28  | W32  | W36  | W40  | W44  | W48  | W52  |
| ESR eCRFs <sup>19,22</sup>                   | X   | X-q6mo               |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X    |
| <b>CALCULATED METABOLIC ASSESSMENTS</b>      |     |                      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |
| MAGE   |     | X-q6mo               |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X    |
| LI   | X   | X-q6mo               |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X    |
| Clarke Score                                 | X   | X-q6mo               |                 |     |     |      |      |      |      |      |      |      |      |      |      | X    |      |      |      |      |      | X    |
| HYP0   | X   | X-q6mo               |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X    |
| Beta Score                                   |     | X                    |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X    |
| C-peptide (gluc X creat) ratio               | X   | X                    |                 |     |     |      |      |      | X    | X    | X    |      | X    | X    | X    | X    | X    | X    | X    | X    | X    | X    |
| <b>IMMUNOSUPPRESSION LEVELS</b>              |     |                      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |
| Belatacept trough levels <sup>23</sup>       |     |                      |                 |     | X   | X    | X    | X    | X    | X    | X    | X    | X    | X    | X    | X    | X    | X    | X    | X    | X    | X    |
| <b>MECHANISTIC ASSAYS</b>                    |     |                      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |
| Alloantibody                                 | X   | X-q6mo <sup>24</sup> |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X    |
| Autoantibody <sup>25</sup>                   |     | X                    |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X    |
| TAT, C3a, & c-peptide                        |     | X                    | X <sup>26</sup> |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |
| Immunogenicity samples <sup>27</sup>         |     |                      | X <sup>28</sup> |     |     |      |      |      |      |      |      |      |      |      | X    |      |      |      |      |      |      | X    |
| <b>ARCHIVED SAMPLES</b>                      |     |                      |                 |     |     |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |      |
| Serum  |     | X                    |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X    |
| PBMC & Plasma                                |     | X                    |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X    |
| RNA  |     | X                    |                 |     |     |      |      |      |      |      | X    |      |      |      |      | X    |      |      | X    |      |      | X    |

<sup>1</sup> WL = Waiting List. BL = Baseline. Repeat assessments as indicated (i.e. yrly, q3mo), while subject is on the waiting list. All one-time WL/BL assessments should be completed on Day -2 whenever possible, but always prior to start of immunosuppression. For WL/BL repeat assessments, record results from test done closest to the start of immunosuppression will be used as the baseline value.

<sup>2</sup> Day 0 = the day of transplant.

<sup>3</sup> *Informed consent #1* includes information on CIT04 and the multi-center Phase 3 protocol (CIT07)

<sup>4</sup> *Informed Consent #2* includes information specific to CIT04. IC # 2 must be signed immediately after randomization.

<sup>5</sup> Retinopathy eval includes fundoscopic pictures for WL/BL assessments and Y1. Screening retinopathy evaluation should be done per site-specific standards. If pupils cannot be dilated, then a manual ophthalmologic evaluation can be substituted.

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- <sup>6</sup> These can be collected after subject is considered protocol eligible and has been moved to the transplant wait list, as time allows.
- <sup>7</sup> Chemistry includes: Sodium, albumin, magnesium, chloride, potassium, alk phosphatase, total bilirubin, CO<sub>2</sub>, creatinine, ALT (SGPT), BUN, gamma GT, glucose, AST (SGOT), calcium, phosphorus
- <sup>8</sup> Complete serum pregnancy tests within 72 hours prior to initiation of study medication.
- <sup>9</sup> Complete test prior to Belatacept infusion. Confirm negative result prior to administering Belatacept
- <sup>10</sup> Serology includes: HBc Ab, HBs Ab, HBs Ag, HCV Ab, and HIV. Do not repeat Hepatitis B tests if HBs Ab was previously positive.
- <sup>11</sup> Repeat only if previous test was negative.
- <sup>12</sup> Repeat for subsequent transplant(s)
- <sup>13</sup> Sample used for crossmatch may be obtained up to 60 days prior to the start of immunosuppression, as long as there is no evidence of infections or transfusions since the time the sample was drawn. Repeat crossmatch for subsequent transplants.
- <sup>14</sup> C-peptide should be done locally and drawn fasting, and twice between 1-3 hrs post-prandial on Day 3 and Day 7 post-transplant.
- <sup>15</sup> Finger stick glucose should be done locally and drawn every hour for the first 6 hours immediately post-transplant.
- <sup>16</sup> Subsequent transplants only. Local result used to determine eligibility for subsequent transplants only.
- <sup>17</sup> EBV by PCR should only be done post-randomization if reactivation is suspected.
- <sup>18</sup> First morning spot urine includes: albumin, protein, and creatinine
- <sup>19</sup> Do not collect for participants with graft failure. Results of tests performed at the time of graft failure will be used for day 75 endpoint calculations.
- <sup>20</sup> MMTT should include 60 and 90 minute c-peptide and glucose measurements for the screening visit and as necessary when determining graft failure.
- <sup>21</sup> Atherogenic profile consisting of fasting lipid panel (TG, TC, HDL, LDL, non-HDL), C reactive protein, serum amyloid A, apolipoprotein A1 and apolipoprotein B. If blood is drawn locally, sample should be sent from local lab to study site and then shipped to the central laboratory (Univ of Washington).
- <sup>22</sup> Blood Sugar Record (BSR) eCRF is completed using information gathered from subject diary logs, glucometer download data, and insulin requirements
- <sup>23</sup> Complete test prior to Belatacept infusion.
- <sup>24</sup> For each transplant, complete alloantibody assessment every 6 months and again on Day -2, regardless of the most recent draw. Central PRA result, current within 6 months, is used to determine subject eligibility for first transplant
- <sup>25</sup> Autoantibody testing includes GAD, IA-2, and IAA.
- <sup>26</sup> TAT & c-peptide: pre-tx, 15, 60, 180 min post-tx
- <sup>27</sup> Collect immunogenicity samples prior to belatacept infusion. For subjects that discontinue belatacept, collect additional immunogenicity samples at 4 weeks and 8 weeks post last dose.
- <sup>28</sup> Collect immunogenicity sample prior to dosing with immunosuppression medications.

### Appendix 3: Continuation of Appendix 1 Schedule of Events (Subjects with Subsequent CIT Transplants)

#### Appendix 2: Continuation of Appendix 1 Schedule of Events (Subjects with Subsequent CIT Transplants)

Subjects who receive a subsequent islet transplant should continue their study visits according to this Schedule of Events until one year (365 +/- 5 days) after the final islet transplant. Additional visits - outlined in Appendix 3 - will also need to be completed after each subsequent transplant. After day 365 post final transplant, subjects should stop following this schedule, and complete the follow-up visits outlined in Appendix 4: *Schedule of Events for 1-Year Additional Follow-Up*.

| Time points (in days relative to 1 <sup>st</sup> transplant) | 392                                 | 420 | 448 | 476 | 504 | 532 | 560 | 588 | 616 |
|--|-------------------------------------|-----|-----|-----|-----|-----|-----|-----|-----|
| Visit Number   | 22                                  | 23  | 24  | 25  | 26  | 27  | 28  | 29  | 30  |
| Visit Windows (in days)                                      | ±5                                  | ±5  | ±5  | ±5  | ±5  | ±5  | ±5  | ±5  | ±5  |
| Equivalent Week  | W56                                 | W60 | W64 | W68 | W72 | W76 | W80 | W84 | W88 |
| <b>GENERAL ASSESSMENTS</b>                                   |                                     |     |     |     |     |     |     |     |     |
| Physical Exam  |                                     |     | X   |     |     | X   |     |     | X   |
| QOL  |                                     |     | X   |     |     | X   |     |     | X   |
| AE/Hypo Event/Toxicity Assessment                            | X                                   | X   | X   | X   | X   | X   | X   | X   | X   |
| <b>LOCAL LABORATORY ASSESSMENTS<sup>1</sup></b>              |                                     |     |     |     |     |     |     |     |     |
| CBC (WBC + Diff & Plat)                                      |                                     |     | X   |     |     | X   |     |     | X   |
| Chemistry <sup>2</sup>                                       |                                     |     | X   |     |     | X   |     |     | X   |
| Lipids   |                                     |     | X   |     |     | X   |     |     | X   |
| Pregnancy test (WOCBP) <sup>3</sup>                          | X                                   | X   | X   | X   | X   | X   | X   | X   | X   |
| <b>CENTRAL LABORATORY ASSESSMENT</b>                         |                                     |     |     |     |     |     |     |     |     |
| HbA1c  |                                     |     | X   |     |     | X   |     |     | X   |
| Fasting serum glucose & c-peptide & serum creat <sup>4</sup> | X                                   | X   | X   | X   | X   | X   | X   | X   | X   |
| 90 min <sup>5</sup> c-peptide/glucose (MMIT) <sup>4</sup>    |                                     |     | X   |     |     | X   |     |     | X   |
| Atherogenic Profile <sup>6</sup>                             | <i>1 year post-final transplant</i> |     |     |     |     |     |     |     |     |
| <b>LOCAL METABOLIC ASSESSMENTS</b>                           |                                     |     |     |     |     |     |     |     |     |
| BSR eCRFs <sup>4,7</sup>                                     |                                     |     | X   |     |     | X   |     |     | X   |
| <b>CALCULATED METABOLIC ASSESSMENTS</b>                      |                                     |     |     |     |     |     |     |     |     |
| MAGE   |                                     |     | X   |     |     | X   |     |     | X   |
| LI   |                                     |     | X   |     |     | X   |     |     | X   |
| Clarke Score   |                                     |     |     |     |     | X   |     |     |     |
| HYP0   |                                     |     | X   |     |     | X   |     |     | X   |

**Appendix 2: Continuation of Appendix 1 Schedule of Events (Subjects with Subsequent CIT Transplants)**

Subjects who receive a subsequent islet transplant should continue their study visits according to this Schedule of Events until one year (365 +/- 5 days) after the final islet transplant. Additional visits - outlined in Appendix 3 - will also need to be completed after each subsequent transplant. After day 365 post final transplant, subjects should stop following this schedule, and complete the follow-up visits outlined in Appendix 4: *Schedule of Events for 1-Year Additional Follow-Up*.

| Time points (in days relative to 1 <sup>st</sup> transplant) | 392                                 | 420 | 448 | 476 | 504 | 532 | 560 | 588 | 616 |
|--|-------------------------------------|-----|-----|-----|-----|-----|-----|-----|-----|
| Visit Number   | 22                                  | 23  | 24  | 25  | 26  | 27  | 28  | 29  | 30  |
| Visit Windows (in days)                                      | ±5                                  | ±5  | ±5  | ±5  | ±5  | ±5  | ±5  | ±5  | ±5  |
| Equivalent Week  | W56                                 | W60 | W64 | W68 | W72 | W76 | W80 | W84 | W88 |
| <b>GENERAL ASSESSMENTS</b>                                   |                                     |     |     |     |     |     |     |     |     |
| Physical Exam  |                                     |     | X   |     |     | X   |     |     | X   |
| QOL  |                                     |     | X   |     |     | X   |     |     | X   |
| AE/Hypo Event/Toxicity Assessment                            | X                                   | X   | X   | X   | X   | X   | X   | X   | X   |
| <b>LOCAL LABORATORY ASSESSMENTS<sup>1</sup></b>              |                                     |     |     |     |     |     |     |     |     |
| CBC (WBC + Diff & Plat)                                      |                                     |     | X   |     |     | X   |     |     | X   |
| Chemistry <sup>2</sup>                                       |                                     |     | X   |     |     | X   |     |     | X   |
| Lipids   |                                     |     | X   |     |     | X   |     |     | X   |
| Pregnancy test (WOCBP) <sup>3</sup>                          | X                                   | X   | X   | X   | X   | X   | X   | X   | X   |
| <b>CENTRAL LABORATORY ASSESSMENT</b>                         |                                     |     |     |     |     |     |     |     |     |
| HbA1c  |                                     |     | X   |     |     | X   |     |     | X   |
| Fasting serum glucose & c-peptide & serum creat <sup>4</sup> | X                                   | X   | X   | X   | X   | X   | X   | X   | X   |
| 90 min <sup>5</sup> c-peptide/ glucose (MMTT) <sup>4</sup>   |                                     |     | X   |     |     | X   |     |     | X   |
| Atherogenic Profile <sup>6</sup>                             | <i>1 year post-final transplant</i> |     |     |     |     |     |     |     |     |
| <b>LOCAL METABOLIC ASSESSMENTS</b>                           |                                     |     |     |     |     |     |     |     |     |
| BSR eCRFs <sup>4,7</sup>                                     |                                     |     | X   |     |     | X   |     |     | X   |
| <b>CALCULATED METABOLIC ASSESSMENTS</b>                      |                                     |     |     |     |     |     |     |     |     |
| MAGE   |                                     |     | X   |     |     | X   |     |     | X   |
| LI   |                                     |     | X   |     |     | X   |     |     | X   |
| Clarke Score   |                                     |     |     |     |     | X   |     |     |     |
| HYP0   |                                     |     | X   |     |     | X   |     |     | X   |

|  |     |     |     |     |     |     |     |     |     |
|--|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Time points (in days relative to 1 <sup>st</sup> transplant) | 392 | 420 | 448 | 476 | 504 | 532 | 560 | 588 | 616 |
| Visit Number   | 22  | 23  | 24  | 25  | 26  | 27  | 28  | 29  | 30  |
| Visit Windows (in days)                                      | ±5  | ±5  | ±5  | ±5  | ±5  | ±5  | ±5  | ±5  | ±5  |
| Equivalent Week  | W56 | W60 | W64 | W68 | W72 | W76 | W80 | W84 | W88 |
| <b>CALCULATED METABOLIC ASSESSMENTS (continued)</b>          |     |     |     |     |     |     |     |     |     |
| Beta Score   |     |     | X   |     |     | X   |     |     | X   |
| C-peptide (glucose X creatinine) ratio                       | X   | X   | X   | X   | X   | X   | X   | X   | X   |
| <b>IMMUNOSUPPRESSION LEVELS</b>                              |     |     |     |     |     |     |     |     |     |
| Belatacept trough levels <sup>8</sup>                        | X   | X   | X   | X   | X   | X   | X   | X   | X   |
| <b>MECHANISTIC ASSAYS</b>                                    |     |     |     |     |     |     |     |     |     |
| Alloantibodies <sup>9</sup>                                  |     |     | X   |     |     | X   |     |     | X   |
| Autoantibodies <sup>10</sup>                                 |     |     | X   |     |     | X   |     |     | X   |
| Immunogenicity samples <sup>11</sup>                         |     |     |     |     |     | X   |     |     |     |
| <b>ARCHIVED SAMPLES</b>                                      |     |     |     |     |     |     |     |     |     |
| Serum  |     |     | X   |     |     | X   |     |     | X   |
| PBMC & Plasma  |     |     | X   |     |     | X   |     |     | X   |
| RNA  |     |     | X   |     |     | X   |     |     | X   |

<sup>1</sup> EBV by PCR should only be done post-randomization if reactivation is suspected.

<sup>2</sup> Chemistry includes: Sodium, albumin, magnesium, chloride, potassium, alk phosphatase, total bilirubin, CO<sub>2</sub>, creatinine, ALT (SGPT), BUN, gamma GT, glucose, AST (SGOT), calcium, phosphorus

<sup>3</sup> Confirm negative result prior to administering Belatacept.

<sup>4</sup> Also collect as necessary to perform graft failure. Do not collect after graft failure has been confirmed.

<sup>5</sup> MMTT should include 90 minute c-peptide and glucose measurements, add 60 minute as necessary when determining graft failure.

<sup>6</sup> Atherogenic profile consisting of fasting lipid panel (TG, TC, HDL, LDL, non-HDL), C reactive protein, serum amyloid A, apolipoprotein A1 and apolipoprotein B. If blood is drawn locally, sample should be sent from local lab to study site and then shipped to the central laboratory (Univ of Washington).

<sup>7</sup> Blood sugar Record (BSR) eCRF is completed using information gathered from subject diary logs, glucometer download data, and insulin requirements.

<sup>8</sup> Complete test prior to Belatacept infusion. For pregnancy test, confirm negative result prior to administering Belatacept.

<sup>9</sup> For each transplant, complete alloantibody assessment every 6 months and again on Day -2, regardless of the most recent draw.

<sup>10</sup> Autoantibody testing includes GAD, IA-2, and IAA.

<sup>11</sup> Collect immunogenicity samples prior to belatacept infusion. For subjects that discontinue belatacept, collect additional immunogenicity samples at 4 weeks and 8 weeks post last dose.

## Appendix 4: Subsequent Transplant Schedule of Events

### Appendix 3: Subsequent Transplant Schedule of Events

The following immediate post-transplant and endpoint assessments should be completed for subjects who receive a subsequent islet transplant. If any of the visits below fall within an acceptable window of a follow-up visit on the Year One Schedule of Events (Appendix 1) or Year Two Schedule of Events (Appendix 2), the assessments may be added to that follow-up visit so that a separate visit does not need to occur.

| Time point (in days relative to most recent infusion) | 0 <sup>1</sup> | 3   | 4   | 7     | 75    | 365    |
|---|----------------|-----|-----|-------|-------|--------|
| Visit Number  | TBD            | TBD | TBD | TBD   | TBD   | TBD    |
| Visit Window (in days)                                | N/A            | N/A | N/A | +/- 3 | +/- 5 | +/- 14 |
| Equivalent Week (post most recent infusion)           | N/A            | N/A | N/A | W1    | N/A   | W52    |
| <b>GENERAL ASSESSMENTS</b>                            |                |     |     |       |       |        |
| Physical Exam   | X              |     |     | X     | X     | X      |
| QOL   |                |     |     |       | X     | X      |
| Chest X-Ray   |                |     |     |       |       | X      |
| Abdominal US (Pelvis/Liver)                           |                |     |     | X     |       | X      |
| ECG   |                |     |     |       |       | X      |
| PPD   |                |     |     |       |       | X      |
| AE/Hypo Event/Toxicity Assess                         | X              |     | X   | X     | X     | X      |
| <b>LOCAL LABORATORY ASSESSMENTS</b>                   |                |     |     |       |       |        |
| CBC (WBC + Diff & Plat)                               | X              |     |     | X     | X     | X      |
| Chemistry <sup>2</sup>                                | X              |     |     | X     | X     | X      |
| Lipids  |                |     |     |       | X     | X      |
| Coagulation (PT, PTT, INR)                            | X              |     |     |       |       |        |
| Blood Type & HLA                                      | X <sup>3</sup> |     |     |       |       |        |
| Crossmatch  | X <sup>4</sup> |     |     |       |       |        |
| PRA by flow cytometry                                 | X <sup>5</sup> |     |     |       |       |        |
| Fasting & post-prandial c-peptide <sup>6</sup>        |                | X   |     | X     |       |        |
| Glucose (immediately post-transplant) <sup>7</sup>    | X              |     |     |       |       |        |
| Pregnancy test (WOCBP) <sup>8</sup>                   | X              |     |     |       |       | X      |
| CMV by PCR <sup>9</sup>                               |                |     |     |       | X     |        |
| <b>CENTRAL LABORATORY ASSESSMENTS</b>                 |                |     |     |       |       |        |
| First morning spot urine <sup>10</sup>                |                |     |     |       | X     | X      |
| GFR   |                |     |     |       | X     | X      |
| HbA1c   |                |     |     |       | X     | X      |
| Fasting serum glucose/c-peptide & creat <sup>11</sup> |                |     |     |       | X     | X      |
| Insulin modified P51GT <sup>11</sup>                  |                |     |     |       | X     | X      |
| 90 min c-pep/glucose (MMIT) <sup>11</sup>             |                |     |     |       | X     | X      |
| Artherogenic Profile <sup>12</sup>                    |                |     |     |       |       | X      |
| <b>LOCAL METABOLIC ASSESSMENTS</b>                    |                |     |     |       |       |        |
| Glycemic Stability (CGMS) <sup>11</sup>               |                |     |     |       | X     | X      |
| BSR eCRFs <sup>11,13</sup>                            |                |     |     |       | X     | X      |
| <b>CALCULATED METABOLIC ASSESSMENTS</b>               |                |     |     |       |       |        |
| MAGE  |                |     |     |       | X     | X      |
| LI  |                |     |     |       | X     | X      |
| Clarke Score  |                |     |     |       |       | X      |
| HYPO  |                |     |     |       | X     | X      |
| Beta Score  |                |     |     |       | X     | X      |

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| Time point (in days relative to most recent infusion) | 0 <sup>1</sup>  | 3   | 4   | 7     | 75    | 365    |
|---|-----------------|-----|-----|-------|-------|--------|
| Visit Number  | TBD             | TBD | TBD | TBD   | TBD   | TBD    |
| Visit Window (in days)                                | N/A             | N/A | N/A | +/- 3 | +/- 5 | +/- 14 |
| Equivalent Week (post most recent infusion)           | N/A             | N/A | N/A | W1    | N/A   | W52    |
| <b>CALCULATED METABOLIC ASSESSMENTS (continued)</b>   |                 |     |     |       |       |        |
| C-peptide glucose creatinine ratio                    |                 |     |     |       | X     | X      |
| <b>IMMUNOSUPPRESSION LEVELS</b>                       |                 |     |     |       |       |        |
| Belatacept trough levels <sup>14</sup>                |                 |     | X   | X     | X     | X      |
| <b>MECHANISTIC ASSAYS</b>                             |                 |     |     |       |       |        |
| Alloantibody <sup>15</sup>                            | X               |     |     |       | X     | X      |
| Autoantibody <sup>16</sup>                            |                 |     |     |       | X     | X      |
| TAT, C3a, & c-peptide                                 | X <sup>17</sup> |     |     |       |       |        |
| Immunogenicity samples <sup>18</sup>                  |                 |     |     |       |       | X      |
| <b>ARCHIVED SAMPLES</b>                               |                 |     |     |       |       |        |
| Serum   |                 |     |     |       | X     | X      |
| PBMC & Plasma   |                 |     |     |       | X     | X      |
| RNA   |                 |     |     |       | X     | X      |

<sup>1</sup> Day 0 = the day of transplant.

<sup>2</sup> Chemistry includes: Sodium, albumin, magnesium, chloride, potassium, alk phosphatase, total bilirubin, CO<sub>2</sub>, creatinine, ALT (SGPT), BUN, gamma GT, glucose, AST (SGOT), calcium, phosphorus

<sup>3</sup> Repeat for subsequent transplant(s).

<sup>4</sup> Sample used for crossmatch may be obtained up to 60 days prior to islet infusion, as long as there is no evidence of infections or transfusions since the time the sample was drawn.

<sup>5</sup> PRA by flow cytometry should be performed locally prior to any subsequent transplant. Local result used to determine eligibility for subsequent transplants only.

<sup>6</sup> C-peptide should be done locally and drawn fasting, and twice between 1-3 hrs post-prandial on Day 3 and Day 7 post-transplant.

<sup>7</sup> Finger stick glucose should be done locally and drawn every hour for the first 6 hours immediately post-transplant.

<sup>8</sup> Perform pregnancy test and confirm negative result prior to each belatacept infusion.

<sup>9</sup> EBV by PCR should only be done post-randomization if reactivation is suspected.

<sup>10</sup> First morning spot urine includes: albumin, protein, and creatinine.

<sup>11</sup> Also collect as necessary to confirm graft failure. Do not collect after graft failure has been confirmed.

<sup>12</sup> Atherogenic profile consisting of fasting lipid panel (TG, TC, HDL, LDL, non-HDL), C reactive protein, serum amyloid A, apolipoprotein A1 and apolipoprotein B. If blood is drawn locally, sample should be sent from local lab to study site and then shipped to the central laboratory (Univ of Washington).

<sup>13</sup> Blood Sugar Record (BSR) eCRF is completed using information gathered from subject diary logs, glucometer download data, and insulin requirements.

<sup>14</sup> Complete test prior to belatacept infusion. For pregnancy test, confirm negative result prior to administering belatacept.

<sup>15</sup> For each transplant, complete alloantibody assessment every 6 months and again on Day -2, regardless of the most recent draw.

<sup>16</sup> Autoantibody testing includes GAD, IA-2, and IAA.

<sup>17</sup> TAT & c-peptide: pre-infusion, 15, 60, and 180 min post-infusion.

<sup>18</sup> Collect immunogenicity samples prior to belatacept infusion. For subjects that discontinue belatacept, collect additional immunogenicity samples at 4 weeks and 8 weeks post last dose.

## Appendix 5: Schedule of Events for 1-Year Additional F/U

### Appendix 4: Schedule of Events for 1-Year Additional Follow-Up

| Time point (Equivalent weeks after 'Day 365 post final transplant' visit) | W4 (AY) | W8 (AY) | W12 (AY)       | W16 (AY) | W20 (AY) | W24 (AY) | W28 (AY) | W32 (AY) | W36 (AY) | W40 (AY)       | W44 (AY) | W48 (AY) | W52 (AY) | Y2 <sup>1</sup> (AY) |
|---|---------|---------|----------------|----------|----------|----------|----------|----------|----------|----------------|----------|----------|----------|----------------------|
| Visit Number (relative to final islet transplant)                         | 51      | 52      | 53             | 54       | 55       | 56       | 57       | 58       | 59       | 60             | 61       | 62       | 63       | 64                   |
| Visit Window (specified in days)  | ±5      | ±5      | ±5             | ±5       | ±5       | ±5       | ±5       | ±5       | ±5       | ±5             | ±5       | ±5       | ±5       | ±30                  |
| <b>GENERAL ASSESSMENTS</b>  |         |         |                |          |          |          |          |          |          |                |          |          |          |                      |
| Physical Exam   |         |         |                |          |          | X        |          |          |          |                |          |          | X        | X                    |
| Telephone Consult   |         |         | X              |          |          |          |          |          |          | X              |          |          |          |                      |
| QOL   |         |         |                |          |          |          |          |          |          |                |          |          | X        | X                    |
| AE /Hypoglycemic Events/Toxicity Assessment                               |         |         | X              |          |          | X        |          |          |          | X              |          |          | X        | X                    |
| <b>LOCAL LABORATORY ASSESSMENTS</b>                                       |         |         |                |          |          |          |          |          |          |                |          |          |          |                      |
| CBC (WBC + Diff & Plat)   |         |         | X              |          |          | X        |          |          |          | X              |          |          | X        |                      |
| Chemistry <sup>2</sup>  |         |         | X              |          |          | X        |          |          |          | X              |          |          | X        |                      |
| Lipids  |         |         |                |          |          | X        |          |          |          |                |          |          | X        |                      |
| Pregnancy test (WOCBP) <sup>3</sup>                                       | X       | X       | X              | X        | X        | X        | X        | X        | X        | X              | X        | X        | X        |                      |
| <b>CENTRAL LABORATORY ASSESSMENTS</b>                                     |         |         |                |          |          |          |          |          |          |                |          |          |          |                      |
| First morning spot urine <sup>4</sup>                                     |         |         |                |          |          | X        |          |          |          |                |          |          | X        |                      |
| GFR   |         |         |                |          |          |          |          |          |          |                |          |          | X        |                      |
| HbA1c   |         |         | X <sup>5</sup> |          |          | X        |          |          |          | X <sup>5</sup> |          |          | X        | X                    |
| 90-min c-pep/glucose (MMIT) <sup>2</sup>                                  |         |         |                |          |          | X        |          |          |          |                |          |          | X        | X                    |
| Atherogenic Profile   |         |         |                |          |          |          |          |          |          |                |          |          | X        |                      |
| <b>LOCAL METABOLIC ASSESSMENTS</b>  |         |         |                |          |          |          |          |          |          |                |          |          |          |                      |
| Glycemic Stability (CGMS) <sup>2</sup>                                    |         |         |                |          |          |          |          |          |          |                |          |          | X        | X                    |

<sup>1</sup> Two years post initial transplant.

<sup>2</sup> Also collect as necessary to confirm graft failure. Do not collect after graft failure is confirmed.

| Time point (Equivalent weeks after 'Day 365 post final transplant' visit) | W4 (AY) | W8 (AY) | W12 (AY) | W16 (AY) | W20 (AY) | W24 (AY) | W28 (AY) | W32 (AY) | W36 (AY) | W40 (AY) | W44 (AY) | W48 (AY) | W52 (AY) | Y2 <sup>1</sup> (AY) |
|---|---------|---------|----------|----------|----------|----------|----------|----------|----------|----------|----------|----------|----------|----------------------|
| Visit Number (relative to final islet transplant)                         | 51      | 52      | 53       | 54       | 55       | 56       | 57       | 58       | 59       | 60       | 61       | 62       | 63       | 64                   |
| Visit Window (specified in days)  | ±5      | ±5      | ±5       | ±5       | ±5       | ±5       | ±5       | ±5       | ±5       | ±5       | ±5       | ±5       | ±5       | ±30                  |
| CALCULATED METABOLIC ASSESSMENTS  |         |         |          |          |          |          |          |          |          |          |          |          |          |                      |
| Clarke Score  |         |         |          |          |          |          |          |          |          |          |          |          | X        |                      |
| IMMUNOSUPPRESSION LEVELS  |         |         |          |          |          |          |          |          |          |          |          |          |          |                      |
| Belatacept Trough Levels  | X       | X       | X        | X        | X        | X        | X        | X        | X        | X        | X        | X        | X        |                      |
| MECHANISTIC ASSAYS  |         |         |          |          |          |          |          |          |          |          |          |          |          |                      |
| Autoantibody  |         |         |          |          |          | X        |          |          |          |          |          |          | X        |                      |
| MECHANISTIC ASSAYS (Cont'd)   |         |         |          |          |          |          |          |          |          |          |          |          |          |                      |
| Alloantibody  |         |         |          |          |          | X        |          |          |          |          |          |          | X        |                      |
| Immunogenicity Samples  |         |         |          |          |          |          |          |          |          |          |          |          | X        | X                    |

<sup>1</sup> Since belatacept is infused every 28 days, an additional infusion visit will be required during the second year. Since the timing is dependent on final transplant date, this visit can be shifted as needed.

<sup>2</sup> Chemistry includes: Sodium, albumin, magnesium, chloride, potassium, alk phosphatase, total bilirubin, CO2, creatinine, ALT (SGPT), BUN, gamma GT, glucose, AST (SGOT), calcium, phosphorus.

<sup>3</sup> Perform pregnancy test and confirm negative result prior to each belatacept infusion.

<sup>4</sup> First morning spot urine includes: albumin, protein, and creatinine.

<sup>5</sup> Can be drawn locally.

### Appendix 6: Supply Order Form: Bulk Supply (shipping supplies)

Protocol CIT-99: Please complete form and fax to University of Iowa @+1-319-353-3960

|                   |  |                    |  |
|-------------------|--|--------------------|--|
| Site Name & #:    |  | CIT Protocol:      |  |
| Shipping Address: |  |                    |  |
| Order Date:       |  | Due Date @ Site:   |  |
| Requested By:     |  | Requestor's phone: |  |
| Requestor's FAX:  |  | Requestor's email: |  |

| SHIPPING SUPPLIES   |  |
|---|--|
| Shipping Container – ambient kit                            |  |
| Shipping Container – refrigerated kit                       |  |
| Shipping Container – frozen kit                             |  |
| STP-317 ambient gel packs                                   |  |
| FedEx Airbills to University of Washington—DRY ICE          |  |
| FedEx Airbills to University of Washington—COLD PACK        |  |
| FedEx Airbills to University of Pennsylvania                |  |
| FedEx Airbills to Barbara Davis Center                      |  |
| FedEx Airbills to Rudbeck Laboratory                        |  |
| FedEx Airbills to NIDDK Repository                          |  |
| FedEx Airbills to ITN Central Cell Processing Core Facility |  |
| FedEx Airbills to ITN RNA Isolation Core Facility           |  |
| FedEx Airbills to University of Minnesota                   |  |
| FedEx Airbills to Tandem Lab                                |  |
| FedEx Airbills to PPD                                       |  |