Sample processing and storage manual - BSTOP Study
Version 1.0, 10 January 2017
<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Overview and contact details for sample management</td>
<td>3</td>
</tr>
<tr>
<td>Site Equipment requirements</td>
<td>4</td>
</tr>
<tr>
<td>Sample collection and processing supplies</td>
<td>5</td>
</tr>
<tr>
<td>Sample tubes</td>
<td>6</td>
</tr>
<tr>
<td>Sample schedule</td>
<td>7</td>
</tr>
<tr>
<td>Blood tube and Eppendorf labelling</td>
<td>9</td>
</tr>
<tr>
<td>Completing the sample request form</td>
<td>10</td>
</tr>
<tr>
<td>Collecting and processing samples</td>
<td>11</td>
</tr>
<tr>
<td>Sample Shipments</td>
<td>13</td>
</tr>
<tr>
<td>Sample Shipments - Manifest/Sample Log</td>
<td>14</td>
</tr>
<tr>
<td>Packaging samples for shipping on dry ice</td>
<td>15</td>
</tr>
<tr>
<td>Packaging samples for shipping at ambient temperature (DNA and Mtx only)</td>
<td>17</td>
</tr>
</tbody>
</table>
Overview and contact details for sample management

This manual

The details for specimen handling, processing, storage and shipment are provided in this manual. This is meant to provide a practical guide for BSTOP sample management and answer the most common questions that we receive regarding BSTOP samples.

If you have any queries regarding supplies, sample management or arranging a shipment then please refer to the contact details provided below:

Contact details

Study Administrator
Maria Troy
Maria.Troy@gstt.nhs.uk
Contact for:
Sending CRFs by email/post
Receipt of documents

Lab Technician
David Baudry
David.Baudry@gstt.nhs.uk
Contact for:
Ordering/Requesting supplies
Arranging and receipt of sample shipments
Receipt of posted Methotrexate samples

Data and Sample Manager
Michael Duckworth
Michael.Duckworth@gstt.nhs.uk
Contact for:
Queries regarding sample collection, processing and storage.

Study Manager
Alice Russell
Alice.Russell1@gstt.nhs.uk
Contact for:
Study related queries
Site Equipment requirements

Sample storage
- Each site is expected to have at minimum a -20°C freezer to store samples.

Fridge
- Sites will need to have access to a fridge (4 or 5°C) for the temporary storage of samples between collection, processing, freezing and/or transport.

Centrifuge - Serum
- Sites collecting serum samples will also need a centrifuge capable of centrifuging serum at 1000g.

Discuss any equipment or storage problems that arise as soon as possible with the Study Manager. It is best that these issues are tackled early rather than left too late where samples may be compromised.
Sample collection and processing supplies

The following is a general list of BSTOP supplies used to collect, process, store and ship samples. The blood collection tubes used in BSTOP and the sample request forms are detailed separately in this guide.

Requests for supplies, including blood collection tubes, should be directed to the contact listed on the overview and contact details page.

DO NOT use expired kits or tubes. The expiration date should be clearly marked on the collection kits and blood tubes. For all tubes and kits the expiry date is the last day of the month given. If supplies have expired, please request more supplies from the BSTOP team.

**Permanent Marker pen - Staedtler Lumocolor Black (All)**
To be used to label all blood collection tubes and Eppendorf’s for serum before storage.

**Eppendorf tubes (Serum)**
For storage of all serum samples.

**Blood collection kits (All)** - these can be sent out to any site who do not regularly use Vacuette tubes, or whose trust collection kits are not compatible with our tubes.

**Plastic sample bags (All)** - for sample collection and storage

**Pipettes (Serum)** - to transfer serum from Vacuette tubes to Eppendorf’s for storage.

**Royal Mail Safe Box (Methotrexate)** -
All Methotrexate samples, and the DNA samples from Methotrexate patients, should be posted to the St John’s Institute of Dermatology for processing, in the Royal Mail Safe Boxes. Postage is pre-paid so doesn’t require a freepost label.
The sample tubes required for BSTOP are listed below. Not all sites will use all of these tubes for BSTOP. If you are unsure, please do contact the BSTOP team to clarify.

### Sample tubes

<table>
<thead>
<tr>
<th>Tube Top</th>
<th>Collection type</th>
<th>Container</th>
<th>Storage</th>
<th>Sites</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pink</td>
<td>DNA</td>
<td>6ml</td>
<td>≤ -20°C</td>
<td>All</td>
</tr>
</tbody>
</table>

**Vacuette - K3E Crossmatch K3EDTA**

<table>
<thead>
<tr>
<th>Tube Top</th>
<th>Collection type</th>
<th>Container</th>
<th>Storage</th>
<th>Sites</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gold</td>
<td>Serum</td>
<td>6ml</td>
<td>≤ -20°C</td>
<td>Longitudinal and Supersites</td>
</tr>
</tbody>
</table>

**Vacuette - Z Serum Sep Clot Activator**

<table>
<thead>
<tr>
<th>Tube Top</th>
<th>Collection type</th>
<th>Container</th>
<th>Storage</th>
<th>Sites</th>
</tr>
</thead>
<tbody>
<tr>
<td>Purple</td>
<td>Methotrexate</td>
<td>4ml</td>
<td>Ambient</td>
<td>Methotrexate collection only</td>
</tr>
</tbody>
</table>

**Vacuette - K2 EDTA2**

<table>
<thead>
<tr>
<th>Tube Top</th>
<th>Collection type</th>
<th>Container</th>
<th>Storage</th>
<th>Sites</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blue</td>
<td>RNA</td>
<td>6ml</td>
<td>≤ -20°C</td>
<td>Supersites only</td>
</tr>
</tbody>
</table>

**Vacuette Tempus - RNA Stabilisation Solution**
Sample schedule

- The samples required at each of the BSTOP sites is summarised below and on the next page
- If you are unsure about which tubes to take, or the type of site you are at then please contact the BSTOP team to clarify.

Single sample sites

- This also includes patients who are recruited to the DNA only cohort at other sites

<table>
<thead>
<tr>
<th>Sample type</th>
<th>Baseline</th>
</tr>
</thead>
<tbody>
<tr>
<td>DNA</td>
<td>Pink top x 2</td>
</tr>
</tbody>
</table>

Longitudinal sites

<table>
<thead>
<tr>
<th>Sample type</th>
<th>Baseline</th>
<th>Week 4</th>
<th>Week 12</th>
<th>6 Monthly</th>
</tr>
</thead>
<tbody>
<tr>
<td>DNA</td>
<td>Pink top x 2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Serum</td>
<td>Gold top x 1</td>
<td>Gold top x 1</td>
<td>Gold top x 1</td>
<td>Gold top x 1</td>
</tr>
</tbody>
</table>

Required for all patients at the designated visits:

| DNA         | Pink top x 2 |        |         |           |

Additional samples required, depending on the patient’s therapy:

- If on a biologic therapy an additional Serum
- If patient is on Methotrexate

<table>
<thead>
<tr>
<th>If on a biologic therapy an additional Serum</th>
<th>Gold top x 1</th>
<th>Gold top x 1</th>
<th>Gold top x 1</th>
<th>Gold top x 1</th>
</tr>
</thead>
<tbody>
<tr>
<td>If patient is on Methotrexate</td>
<td>Purple top x 1</td>
<td>Purple top x 1</td>
<td>Purple top x 1</td>
<td>Purple top x 1</td>
</tr>
</tbody>
</table>

1. The extra visits at Week 4 and Week 12 are specifically for patients who have switched or are switching to a new psoriasis treatment.
2. Align with the patient’s BADBIR visits
3. For patients on biologic therapy AND methotrexate - take Gold top x 2 and the Purple top x 1.
4. For patients on Methotrexate only - take Gold top x 1 and the Purple top x 1
Sample schedule cont...

Supersites

<table>
<thead>
<tr>
<th>Sample type</th>
<th>Baseline</th>
<th>Week 4&lt;sup&gt;1&lt;/sup&gt;</th>
<th>Week 12&lt;sup&gt;1&lt;/sup&gt;</th>
<th>6 Monthly&lt;sup&gt;2&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>DNA</td>
<td>Pink top x 2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Serum</td>
<td>Gold top x 1</td>
<td>Gold top x 1</td>
<td>Gold top x 1</td>
<td>Gold top x 1</td>
</tr>
<tr>
<td>RNA&lt;sup&gt;1&lt;/sup&gt;</td>
<td>Tempus x 1</td>
<td>Tempus x 1</td>
<td>Tempus x 1</td>
<td></td>
</tr>
</tbody>
</table>

Required for all patients at the designated visits:

- **DNA**: Pink top x 2
- **Serum**: Gold top x 1, Gold top x 1, Gold top x 1, Gold top x 1
- **RNA<sup>1</sup>**: Tempus x 1, Tempus x 1, Tempus x 1

Additional samples required, depending on the patient’s therapy:

- **If on a biologic therapy an additional Serum<sup>3</sup>**:
  - Gold top x 1, Gold top x 1, Gold top x 1, Gold top x 1
- **If patient is on Methotrexate<sup>4</sup>**:
  - Purple top x 1, Purple top x 1, Purple top x 1, Purple top x 1

1. The extra visits at Week 4 and Week 12 and the RNA sample (Tempus x 1) are specifically for patients who have switched or are switching to a new psoriasis treatment.
2. Align with the patient’s BADBIR visits.
3. For patients on biologic therapy AND methotrexate - take Gold top x 2 and the Purple top x 1.
4. For patients on Methotrexate only - take Gold top x 1 and the Purple top x 1.
Blood tube and Eppendorf labelling

All blood collection tubes and Eppendorfs should be labelled appropriately so that they identify the patient and the date the sample was taken. The use of multiple identifiers, along with the sample request form, minimises the risk of not being able to identify the sample should details be smudged or rubbed off during processing, storage or transport. This is important as if there is any doubt as to the identity of the sample it will have to be discarded.

The identifiers needed to label samples are dependent on how BSTOP study data is collected:

If study data is collected directly on the CAPTURE database:

CAPTURE will automatically generate sample numbers for all samples taken at a patient visit. This is done through data entry, under the sample summary. Each sample tube will have a unique sample number, and these, along with the BSTOP ID and the date the sample was taken should be written on each tube or Eppendorf. Sites may, if it proves helpful for sample management, also write the patient initials on tubes, and the visit number on serum Eppendorf tubes.

If study data is collected on the paper CRF:

If your site is not yet using CAPTURE then you are using the paper based CRF and the sample numbers will be generated by the BSTOP team when the samples are received. The BSTOP ID number should be used rather than patient names or hospital numbers, to maintain patient confidentiality. Also written on the tubes should be the date the sample was taken and the patient initials. Sites may, if it proves helpful for sample management, also write the visit number on serum Eppendorf tubes.

Example - labelled blood collection and serum Eppendorf tubes for CAPTURE generated samples

Example - labelled blood collection and serum Eppendorf tubes for paper based CRF’s
Completing the sample request form

Sample request form

Every sample taken at a visit should be accompanied by a sample request form, depending whether your site is using CAPTURE and generating sample numbers will indicate which form to use.

Each line should be filled out as is appropriate for the types of samples taken. Examples of both completed sample request forms are given below:

Example - completed CAPTURE Sample request form

<table>
<thead>
<tr>
<th>BSTOP ID</th>
<th>Z Z B S T O 0 0 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient Initials</td>
<td>W - W</td>
</tr>
<tr>
<td>Date of Birth</td>
<td>0 3 0 5 7 8</td>
</tr>
<tr>
<td>Gender</td>
<td>M</td>
</tr>
<tr>
<td><strong>Samples taken</strong></td>
<td><strong>Blood drawn</strong></td>
</tr>
<tr>
<td>Pink top EDTA - DNA</td>
<td>✓</td>
</tr>
<tr>
<td>Yellow top - Serum</td>
<td>✓ ✓</td>
</tr>
<tr>
<td>Purple top - Methotrexate</td>
<td>✓</td>
</tr>
</tbody>
</table>

Was blood for serum refrigerated between draw and spin? Yes ✓ temp 4 °C No

Was serum refrigerated between spin and freeze? Yes ✓ temp 4 °C No

PLEASE NOTE: If this sample was exposed to any event that could potentially compromise its integrity, or otherwise deviates from standard protocol, please detail on the rear of this sheet. (Such as - unplanned thaw, mislabelling, left out overnight, etc.)

Example - completed Sample request form

<table>
<thead>
<tr>
<th>B-STOP ID</th>
<th>A A B S T O 0 0 1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient Initials</td>
<td>A - S</td>
</tr>
<tr>
<td>Date of Birth</td>
<td>1 9 0 1 5 2</td>
</tr>
<tr>
<td>Gender</td>
<td>M ✓</td>
</tr>
<tr>
<td><strong>Samples taken</strong></td>
<td><strong>Blood drawn</strong></td>
</tr>
<tr>
<td>Pink top - DNA</td>
<td>✓ ✓</td>
</tr>
<tr>
<td>Yellow top - Serum</td>
<td>✓ ✓</td>
</tr>
<tr>
<td>Purple top - Methotrexate</td>
<td>✓</td>
</tr>
<tr>
<td>Blue Top TEMBUS - RNA</td>
<td>✓</td>
</tr>
</tbody>
</table>

Was blood for serum refrigerated between draw and spin? Y ✓ temp 5 °C N

Was serum refrigerated between spin and freeze? Y ✓ temp 5 °C N

NOTE: If this sample was exposed to any event that could potentially compromise its integrity, or otherwise deviates from standard protocol, please detail on the rear of this sheet. (Such as - unplanned thaw, mislabelling, left out overnight, etc.)
Collecting and processing samples

These are the step by step guides for collecting each sample type:

**Whole Blood for DNA**
- Collect DNA blood from every BSTOP patient at the baseline visit unless special allowance is made to collect saliva.
- Collect 12 ml of whole blood in 2x 6ml K$_3$EDTA Vacuette tubes (pink top).
- Label the blood tubes as per the labelling instructions detailed on page 9.
- Write the date and time the blood was collected on the Sample request form.
- Store whole blood at -20°C or colder (ideally -80°C) until transported to the St John’s Institute of Dermatology.
- Write the date and time the blood was frozen on the Sample request form.
- DNA from patients taking Methotrexate: If a patient has both Methotrexate and DNA samples collected on the same day, post the Methotrexate and the DNA sample together in the same Royal Mail Safe box. Samples should be posted at ambient temperature to the St John’s Institute of Dermatology. Include the corresponding sample slips.

**Whole blood for Serum**
- Longitudinal sites and supersites collect serum for biomarker analysis.
- These sites collect serum from all patients at each visit.
- Collect 5ml whole blood in 1 x 5ml serum separating clotting factor Vacuette tube (yellow top).
- For patients on biologic therapies: Collect an extra serum sample (i.e. 2 x 5ml yellow top serum tubes in total).
- Write the date and time the blood was collected on the sample request form.
- Invert 3x to mix blood with clotting factor and rest for at least 30 minutes, ideally in an upright position.
- If longer than 30 minutes, then store serum samples upright in a 5°C refrigerator prior to centrifugation.
- Centrifuge samples ideally within 2 hours of but a maximum of 4 hours after blood collection.
- Centrifuge at 1000g for 15 minutes at ambient temperature.
- Write the date and time of centrifugation on the sample request form.
- Immediately following spin, use a pipette to transfer the separated serum into 2ml Eppendorf tube.
- Use a fresh pipette for each serum tube.
- Use caution to avoid transferring any blood cells or separator gel into the Eppendorf tube.
- Use only one Eppendorf tube if all the serum can fit into one tube.
- Leave some space at the top of each Eppendorf tube as the serum will expand in the tube once frozen. Fill to the 1.5ml graduation line (approximately ¾ full).
- Label the Eppendorf tubes as per the labelling instructions detailed on page 9.
- Store at -20°C or colder (ideally -80°C) until transported to St John’s Institute of Dermatology.
Collecting and processing samples cont...

Whole Blood for Methotrexate (MTX) PK drug levels

- Longitudinal sites and supersites collect PK drug levels from patients taking Methotrexate at each visit.
- Collect 4ml whole blood in 1x K$_2$EDTA Vacuette tube (purple top).
- Invert 5x to mix thoroughly.
- Label the blood tubes as per the labelling instructions detailed on page 9.
- Write the date and time the blood was collected on sample request form.
- Post Methotrexate samples at ambient temperature in postal safe boxes to St John’s Institute of Dermatology.
- Post the corresponding sample slip with the Methotrexate sample.
- If the Methotrexate sample is collected on a Monday, Tuesday or Wednesday, post the sample to the St John’s Institute of Dermatology on the day of collection.
- If the Methotrexate sample is collected on a Thursday or Friday, store the Methotrexate sample at 5°C over the weekend, and post to the St John’s Institute of Dermatology on the following Monday.

Whole blood for RNA

- Only BSTOP supersites collect RNA samples.
- Collect RNA samples at baseline (before treatment start) and then at 1 week, 1 month and 3 months after a patient has started their treatment.
- Collect 3ml of whole blood in a Tempus RNA isolation tube (blue top).
- Label the blood tubes as per the labelling instructions detailed on page 9.
- Write the date and time the blood was collected on Sample request form.
- Vortex or shake vigorously for 10 seconds immediately after collection to ensure thorough mixing.
- Store at -20°C or colder (ideally -80°C) until transported to the St John’s Institute of Dermatology.
- Write the date and time the blood was frozen on sample request form.
Sample Shipments

- Frozen samples will be transferred on dry ice using our preferred carrier - City Sprint.
- All shipments are arranged directly with the BSTOP team - please see the contact details provided on page 2 of this manual.
- When requesting a shipment please provide the estimate number of samples to be collected. This is to ensure that a shipment is feasible, and that the correct sized box is delivered.

When to arrange a shipment?
- Shipments should be arranged at least on an annual basis for each site so that we are able to monitor and process the samples.
- However, a shipment may be requested sooner if storage space is limited, or in case of emergencies (freezer failure, etc.)

Days of the week for shipment collection
- Shipments can be arranged for collection on Tuesdays, Wednesdays, or with prior agreement, Thursdays.
- City Sprint require a day to ensure that dry ice and packaging is available locally for a shipment at each BSTOP site.
- This dry ice and packaging will be delivered to your site on the day of the shipment, with enough time to process and package the samples before the courier comes to pick them up.
- Shipments are couriered overnight, requiring a day to arrive at St Johns Institute of Dermatology. Therefore shipments cannot be arranged for a Friday, as the B-STOP labs are closed over weekends.
- However, if your site is located close enough to London it may be possible to arrange a same day delivery.

Dry ice, packaging and collection times
- You may request a time for the dry ice and packaging to be delivered, as well as your desired time of collection.
Sample Shipments - Manifest/Sample Log

- We’ve put together a Sample shipment manifest to help you manage and track your samples. This also helps us to process them when they arrive at St Johns Institute of Dermatology as we’ll know exactly what to expect in the shipment box. It’ll also mean we can get back to you should anything be missing.

- If you would like to, you can also use it as a sample log for all of your BSTOP samples - by filling it in as you take samples from patients throughout the year.

- There is a PDF and Excel version of the sample manifest - please use whichever is easiest for you.

- Please include a complete paper copy with the shipment you are sending. If you are using the Excel version of the manifest and have filled this in electronically then please email it to both the Lab Technician and the Data and Sample Manager when you send the shipment.

- If you have any questions about using the log then do get in touch with the Data and Sample Manager.

Notes for completing the manifest/Sample Log

Example - completed patient and visit details

<table>
<thead>
<tr>
<th>Study ID</th>
<th>Patient initials</th>
<th>Date of sample</th>
<th>Visit (BL/FU)</th>
</tr>
</thead>
<tbody>
<tr>
<td>X X BST 0 0 0 1</td>
<td>AA</td>
<td>01/01/2017</td>
<td>BL</td>
</tr>
<tr>
<td>X X BST 0 0 0 1</td>
<td>AA</td>
<td>01/06/2017</td>
<td>FU</td>
</tr>
<tr>
<td>X X BST 0 0 0 2</td>
<td>BB</td>
<td>01/03/2017</td>
<td>BL</td>
</tr>
<tr>
<td>X X BST 0 0 0 3</td>
<td>CC</td>
<td>01/04/2017</td>
<td>BL</td>
</tr>
</tbody>
</table>

- Please use one visit per patient per line

- When preparing a manifest for a shipment, if possible, list the samples in order of Study ID, and then by the visit date for each Study ID, e.g. AABST0001 BL, AABST0001 FU then AABST0002 BL etc.

Example - completed sample details

<table>
<thead>
<tr>
<th>Pink Top (DNA)</th>
<th>Yellow Top (Serum)</th>
<th>Blue Tempus (RNA)</th>
<th>Shipped to St Johns Institute of Dermatology</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes/No Number</td>
<td>Yes/No Number</td>
<td>Yes/No Number</td>
<td>Yes/No</td>
</tr>
<tr>
<td>Yes</td>
<td>2</td>
<td>Yes</td>
<td>1</td>
</tr>
<tr>
<td>No</td>
<td>Yes</td>
<td>1</td>
<td>No</td>
</tr>
<tr>
<td>Yes</td>
<td>2</td>
<td>Yes</td>
<td>2</td>
</tr>
<tr>
<td>Yes</td>
<td>2</td>
<td>Yes</td>
<td>1</td>
</tr>
</tbody>
</table>

- Tick or write Yes/Y for each sample type collected at this visit, on the Excel version you can use the drop down.

- The number refers to the number of tubes collected, whether whole tubes of blood or Eppendorf for serum samples.
Packaging samples for shipping on dry ice

The guide below is provided to help you package BSTOP samples for shipment, to ensure that the samples stay frozen, and that samples can be easily processed when they arrive at St John’s Institute of Dermatology.

Always ensure that when handling dry ice, and packaging the samples, that this is done in a well ventilated area.

**Bottom layer - Dry Ice**

1/3 of the dry ice should be placed underneath the samples. Most of the dry ice should be placed over the samples rather than underneath them. This ensures samples stay frozen for longer.

**Middle layer - Samples**

Samples should be placed on the bottom layer of dry ice. It is important that samples are clearly labelled and contained in plastic sample bags to ensure no samples get lost within the dry ice.

Ideally, samples from each patient visit should be packed in a single plastic bag.
Packaging samples for shipping on dry ice continued...

**Top layer - Dry Ice**
2/3 of the dry ice should be placed over the top of the samples. This ensures that the samples remain frozen throughout transport.

**Sample Slips**
Every sample should have a corresponding sample slip. It is best to keep sample slips separately in either an A4 envelope or plastic wallet and place them down the side between the thermal container and the shipping box. Alternatively they can be placed on top of the lid of the thermal container.

**Shipment packing complete**
The shipment is ready for collection by the courier, who will provide labels needed for transit.
Packaging samples for shipping at ambient temperature (DNA and Mtx only)

- The Royal Mail SafeBoxes meet the UN3373 requirements for sending human blood samples through the post and are the BSTOP method for sending samples ambiently in the post.
- All BSTOP Methotrexate samples (Purple Top) should be sent ambiently by this method, as they cannot be processed once frozen.
- If the patient on Methotrexate has a DNA sample (pink top) taken at the same time, the DNA samples can be sent with the Methotrexate sample in the same Royal Mail SafeBox.
- No other BSTOP sample types should be sent through the post, as this will affect the sample quality and stability. All other BSTOP samples need to be frozen and shipped on dry ice.
- The Royal Mail have a very good guide on how to package samples in the SafeBoxes, which is available here: http://www.royalmail.com/business/sites/default/files/docs part 4/docs part 4/Royal-Mail-Safebox-Terms-Conditions-November-2014.pdf
- Please do not use the freepost address for these samples. The SafeBoxes have first class postage prepaid.
- Please send all SafeBoxes to the following address:
  
  Skin Therapy Research Unit  
  9th Floor Tower Wing  
  Guys Hospital  
  Great Maze Pond  
  LONDON  
  SE1 9RT

Please remember:

- Post the corresponding sample slip with the Methotrexate sample.
- If the Methotrexate sample is collected on a Monday, Tuesday or Wednesday, post the sample to the central coordinating centre on the day of collection.
- If the Methotrexate sample is collected on a Thursday or Friday, store the Methotrexate sample in a fridge over the weekend, and post to the central coordinating centre on the following Monday.