Parkinson’s Progression Markers Initiative (PPMI)
Biospecimen Processing Maps Manual
### Title
Parkinson’s Progression Markers Initiative (PPMI) Biospecimen Processing Maps Manual

### Description
Study processing maps outlining the path each sample will take through the acquisition, aliquoting, storage and shipping processes from the clinical sites to the study biorepository and, eventually, to the researcher.

### Created By
Indiana University PPMI Biorepository

### Date Created
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### Maintained By
Indiana University PPMI Biorepository

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1.0 | IU | • Initial Version | October 20, 2021 | Active
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PPMI Skin Tissue Processing
PPMI DNA (Whole Blood and Buffy Coat) Processing
Biospecimen Collection and Processing
Background

The PPMI biospecimen collection ranges in many samples types. As the PPMI study has continued and specimen analysis expanded, so has the need to update these collections. Most biospecimen collections have continued from the initial PPMI collection into the expanded phase (RNA, plasma, serum, buffy coat, urine and CSF), while other specimens would have only been collected during the initial phase of the study (cell lines and PBMCs). For further information on cell lines, please refer to the Cell Lines Manual of Procedures.

The newest addition to biospecimen collection and processing is skin biopsy tissue. PPMI scientists continue to explore the addition of other samples types that may be instrumental in biomarker research.

Furthermore, the biorepository at Indiana University oversees all DNA fingerprinting and analysis (previously, DNA was sent to LGC, formally KBioscience). The updated process has been implemented for whole blood DNA samples received at the PPMI biorepositories in Tel Aviv Sourasky Hospital in Israel, BioRep in Milan, Italy, and the Indiana University Genetics Biobank.

The following processing maps are a guide to how samples are processed once they are received by the biorepository labs.
PBMC samples collected and stored at room temperature

PBMC samples shipped ambient temperature

Clinical Sites notified of problems

Yes

No

Samples missing, damaged or mislabeled

Sample info entered into database

PBMCs isolated same day

Aliquots are created 2.0-3.0 x 10^6 cells per vial

Samples stored at a controlled rate and frozen at -80°C

Issues resolved

PPMI PBMC Processing

versions 1.0

PPMI Biospecimen Processing Maps

Modified on September 29, 2016
PPMI Fibroblast and iPSCs Processing from CDI

<table>
<thead>
<tr>
<th>Accessioning</th>
<th>Aliquoting</th>
<th>Storage</th>
<th>Shipping</th>
<th>Quality Control</th>
<th>Distribution</th>
</tr>
</thead>
<tbody>
<tr>
<td>CDI</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Repository Coordinator</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
</tbody>
</table>

- CDI: Received blood and created fibroblast lines and iPSC lines.
- Batch ship lines on dry ice to IUGB.
- Frozen lines received at repository.
- Visual inspection of samples, tube label matches submission form.
- Samples missing, damaged or mislabeled.
- Sample info entered into database.
- CDI notified of problems.

Flow:
- Yes: CDI notified of problems.
- No: Issue resolved.

- Samples stored in LN2 Vapor Phase.
- Samples and data distributed upon PPMI approval to researcher.
PPMI Fibroblast and iPSCs Processing from NYSCF

<table>
<thead>
<tr>
<th>Accessioning</th>
<th>Aliquoting</th>
<th>Storage</th>
<th>Shipping</th>
<th>Quality Control</th>
<th>Distribution</th>
</tr>
</thead>
<tbody>
<tr>
<td>Received blood and created fibroblast lines and iPSC lines</td>
<td>Batch ship lines on dry ice to IUGB</td>
<td>Visual inspection of samples, tube label matches submission form</td>
<td>Samples missing, damaged or mislabeled</td>
<td>Sample info entered into database</td>
<td>Samples stored in LN2 Vapor Phase</td>
</tr>
<tr>
<td>NYSCF</td>
<td>NYSCF notified of problems</td>
<td>Issue resolved</td>
<td>No</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Yes

Issued resolved

Repository Coordinator

NYSCF

Samples and data distributed upon PPMI approval to researcher

Modified on September 29, 2016

Version 1.0
PPMI DNA (Whole Blood and Buffy Coat) Processing

**Accessioning**
- Clinical Site
  - Whole Blood EDTA tube collected or Buffy Coat isolated from EDTA tube
  - Whole Blood EDTA tube shipped ambient and Buffy Coat shipped frozen
  - Visual inspection of samples, tube label matches submission form

**Aliquoting**
- Clinical Sites notified of problems
  - Yes
  - No
  - Samples missing, damaged or mislabeled

**Storage**
- Sample info entered into database

**Shipping**
- At the end of every quarter an aliquot is plated for KBio fingerprinting
- Plated to KBio for fingerprinting
- KBio runs fingerprint panel including gender SNPs
- Match script ran to compare Whole Blood and Buffy Coat extractions

**Quality Control**
- Concentration and 260/280 Ratio determined
- Samples stored at -80°C
- Primary and back up DNA aliquots are created

**Distribution**
- Results reported to repository and entered into database
- Feedback to clinical sites
- Samples and data distributed upon PPMI approval to researcher

PPMI Biospecimen Processing Maps

*Modified on April 15, 2015*

Version 1.0
PPMI RNA Processing

Clinical Site
- Blood collected in 2 PAXgene RNA tubes and frozen at -80°C
- Frozen PAXgene RNA tubes shipped on dry ice

Repository Coordinator
- Repository Coordinator

Clinical Sites
- Clinical Sites notified of problems
- Yes
- No
- Samples missing, damaged or mislabeled
- Issue resolved
- Results reported to repository and entered into database
- Feedback to clinical sites

Repository
- Samples info entered into database

Accessioning
- Samples and data distributed upon PPMI approval to researcher

Aliquoting
- 1 µg aliquots are created
- Samples stored at -80°C

Storage
- PAXgene RNA tubes are stored at -80°C

Shipping
- RIN calculated using the Agilent BioAnalyzer

Quality Control
- Concentration, 260/280 Ratio and 260/230 Ratio determined

Distribution
- Batches of 12 PAXgene RNA tubes, 1 tube per visit, are extracted to RNA & miRNA
- 1 PAXgene tube stays in long term storage, extracted later via request

PPMI Biospecimen Processing Maps

Modified on March 7, 2016
PPMI Serum Processing

**Accessioning**
- Serum samples collected and frozen at -80°C
- Serum samples shipped on dry ice

**Aliquoting**
- Clinical Sites notified of problems
  - Yes
    - Visual inspection of samples, tube label matches submission form
    - Samples missing, damaged, or mislabeled
      - No
        - Sample info entered into database
        - Samples stored at -80°C
      - Yes
        - Issues resolved

**Storage**
- one tube held in processing rack for batch processing
- one tube divided into 200µL aliquots and one 100µL

**Shipping**
- 100µL aliquot sent for QNT hemoglobin test
- Results reported to repository and entered into database
- QNT hemoglobin test performed

**Quality Control**
- Samples and data distributed upon PPMI approval to researcher
- Samples stored at -80°C

**Distribution**
- Feedback to clinical sites
**PPMI Plasma Processing**

**Clinical Sites**
- Buffy Coat and Plasma samples collected and frozen at -80°C
- Buffy Coat and Plasma samples shipped on dry ice

**PPMI Clinical Sites**
- Frozen Buffy Coat and Plasma samples received at repository
- Visual inspection of samples, tube label matches submission form
- Samples missing, damaged or mislabeled

**Repository Coordinator**
- Problem identified
- Issue resolved
- Sample info entered into database
- Samples stored at -80°C
- 100µL aliquot sent for QNT hemoglobin test
- QNT hemoglobin test performed
- 100µL aliquot sent for QNT hemoglobin test
- Results reported to repository and entered into database
- Feedback to clinical sites
- Samples and data distributed upon PPMI approval to researcher
- Frozen Buffy Coat extracted per DNA workflow
- One tube held in processing rack for batch processing
- One tube divided into 200µL aliquots and one 100µL
- Samples stored at -80°C

**Accessioning** | **Aliquoting** | **Storage** | **Shipping** | **Quality Control** | **Distribution**
---|---|---|---|---|---

**Modified on June 2, 2015**

Version 1.0
Blood collected in 6ml EDTA tube and frozen at -80°C

Blood tube shipped on dry ice

Frozen Whole Blood samples received at repository

Visual inspection of samples, tube label matches submission form

Samples missing, damaged or mislabeled

Sample info entered into database

Samples stored at -80°C

Aliquots made on demand

Clinical Sites notified of problems

Yes

Issue resolved

Samples and data distributed upon PPMI approval to researcher
Urine collected and frozen at -80°C

Urine tube shipped on dry ice

Frozen urine sample received at repository

Visual inspection of samples, tube label matches submission form

Sample info entered into database

Clinical Sites notified of problems

Yes

No

Samples missing, damaged or mislabeled

Issue resolved

Samples stored at -80°C

Aliquots made on demand

Samples and data distributed upon PPMI approval to researcher

Clinical Sites notifed of problems

Repository Coordinator

Repository

PPMI Clinical Sites

Accessioning

Aliquoting

Storage

Shipping

Quality Control

Distribution
PPMI CSF Processing

Accessioning | Aliquoting | Storage | Shipping | Quality Control | Distribution
---|---|---|---|---|---
**Clinical Site**
- CSF collected and frozen at -80°C

**Frozen CSF samples shipped on dry ice**

**Clinical Sites notified of problems**

**Yes**

**No**

**Visual inspection of samples, tube label matches submission form**

**Samples missing, damaged or mislabeled**

**Issue resolved**

**Sample info entered into database**

**Samples stored at -80°C**

**One tube held in processing rack for batch processing**

**One tube divided into 250µL aliquots and one residual aliquot**

**Samples stored at -80°C**

**Samples and data distributed upon PPMI approval to researcher**

**Repository Coordinator**

**Repository**

**PPMI Biospecimen Processing Maps**

Modified on April 30, 2015

Version 1.0

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PPMI Tissue Processing

<table>
<thead>
<tr>
<th>Accessioning</th>
<th>Processing</th>
<th>Storage</th>
<th>Shipping</th>
<th>Quality Control</th>
<th>Distribution</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tissue samples collected and stored at 4°C</td>
<td>Tissue samples shipped on frozen ice packs</td>
<td>Clinical Sites notified of problems</td>
<td>Yes</td>
<td>No</td>
<td>Sample info entered into database</td>
</tr>
<tr>
<td>Tissue samples received at repository</td>
<td>Visual inspection of samples, label matches submission form</td>
<td>Samples missing, damaged or mislabeled</td>
<td>Issue resolved</td>
<td>Sample info entered into database</td>
<td>Tissue transferred to pathology same day for processing</td>
</tr>
<tr>
<td>Sample is paraffin embedded and has 18 slide cut</td>
<td>1 slide stained for H&amp;E</td>
<td>Samples stored at a room temperature</td>
<td>Samples and data distributed upon PPMI approval to researcher</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Clinical Sites

Repository Coordinator

Repository

PPMI Clinical Sites

PPMI Tissue Processing

Modified on August 12, 2021

Version 1.0

15 of 16
PPMI DNA (Whole Blood and Buffy Coat) Processing

**PPMI**

**Biospecimen Processing Maps**

**PPMI**

**DNA** *(Whole Blood and Buffy Coat)*

**Accessioning**

Clinical Sites

- Whole Blood EDTA tube collected or Buffy Coat isolated from EDTA tube
- Whole Blood EDTA tube shipped ambient and Buffy Coat shipped frozen

**Repository Coordinator**

- Visual Inspection of samples, tube label matches submission form
- Samples missing, damaged or mislabeled

**Sample info entered into database**

Yes

- Clinical Sites notified of problems

No

- Sample info entered into database

**Repository**

- Whole Blood EDTA tube or Frozen Buffy Coat extracted
- Whole Blood EDTA tube or Frozen Buffy Coat shipped frozen

**Aliquoting**

- Aliquots are plated for fingerprinting
- Plates ship to IU for fingerprinting

**Storage**

- IU runs fingerprint panel including gender SNPs
- Match script ran to compare Whole Blood and Buffy Coat extractions

- Concentration and 260/280 Ratio determined

**Shipping**

- Results reported to repository and entered into database

**Quality Control**

- Samples and data distributed upon PPMI approval to researcher

- Feedback to clinical sites

**Distribution**

- Samples stored at -80°C
- Samples and data distributed upon PPMI approval to researcher

**Modified on August 12, 2021**

**Version 1.0**