CORRELATIVE SCIENCE PROCEDURE MANUAL

1. Purpose

This document describes the procedures required for the collection, shipping, and processing of biospecimens from all patients enrolled or registered on A062102. This document also describes the procedures that will be followed subsequent to the receipt of biospecimens by the Alliance Hematologic Malignancy Biorepository (HEME) prior to their use for protocol-specified and future, unspecified correlative science research studies. This document should be used by staff involved with any aspect of the A062102 biospecimen collection, processing, and submission, including staff at satellite institutions.

2. Scope

This document applies to all biospecimens collected specifically for A062102 only. Please refer to the trial protocol-specific language for additional details regarding eligibility, participant enrollment, data submission, and specific procurement procedures. **Please ensure that you are reading the most updated version of this document. This document may experience minor updates, revisions, and clarifications independent of a formal protocol amendment. The most recent version of this document may be found on the Alliance website and CTSU.**

3. Definitions

Term	Definition
HEME	Alliance Hematologic Malignancy Biorepository

4. Contact Information

Protocol-related questions may be directed as follows:	
Questions	**Contact (via email)**
Questions regarding patient eligibility, treatment, and dose modification:	Study Chair: Sascha Tuchman, MD, MHS Sascha_tuchman@med.unc.edu Nursing Contact: Ilene Galinsky, RN ilene_galinsky@dfci.harvard.edu Protocol Coordinator: Destin Carlisle dcarlisle@bsd.uchicago.edu Data Manager: Semir Begic begic.semir@mayo.edu
Questions related to data submission, RAVE or patient follow-up:	Data Manager: Semir Begic begic.semir@mayo.edu
Questions regarding the protocol document and model informed consent:	Protocol Coordinator: Destin Carlisle dcarlisle@bsd.uchicago.edu
Questions related to IRB review	Alliance Regulatory Inbox regulatory@allianceNCTN.org
Questions regarding CTEP-AERS reporting:	Alliance Pharmacovigilance Inbox pharmacovigilance@alliancenctn.org
Questions regarding specimens/specimen submissions:	Alliance Hematologic Malignancy Biorepository (HEME) AllianceLTB@osumc.edu
Questions regarding drug supply	NCI PMB: PMBAfterHours@mail.nih.gov
Questions regarding drug administration	Pharmacy Contact: Maria Andrea Monckeberg mamonckeberg@lifespan.org

4.1 For information on using the BioMS system, please refer to the 'Help' links on the BioMS webpage to access the online user manual, FAQs, and training videos. To report technical problems, such as login issues or application errors, please contact or for assistance in using the application or questions or problems related to specific specimen logging, please contact: 1-855-55-BIOMS or bioms@alliancenctn.org.

4.2 For specific questions regarding sample collection and submission to HEME, please contact the HEME Laboratory at AllianceLTB@osumc.edu or 1-**614-366-6295.**

4.3 For all other questions regarding biospecimen procurement and shipping procedures, please contact the Alliance Biorepository Program Manager: 1-314-747-4402 or alliance@email.wustl.edu.

5. Site Preparation

5.1 Please refer to the A062102 protocol document for any specific requirements related to patient enrollment, registration, and regulatory compliance.

5.2 Please ensure that you have appropriate log on credentials and can successfully access the BioMS application. The BioMS application is used for logging the collection and shipment of biospecimens to HEME. For training and assistance in using the application or questions or problems related to specific specimen logging, please contact: 1-855-55-BIOMS or bioms@alliancenctn.org**.**

5.3 Identify a reliable source of dry ice for freezing and shipping biospecimens and a -70 to -90 degree Celsius freezer ("ultralow") in which frozen biospecimens may be stored prior to shipment.

6. Collection Schema

The following biospecimens are to be collected at each of the time points. Please refer to biospecimen collection and processing methods and specific shipping procedures below.

Please be aware that when MRD analysis is performed, Adaptive Biotechnologies may reach out directly to the site to obtain archival tissue for the Clonoseq assay. This will be sent to Adaptive following their instructions and standard procedures.

Time Point	Biospecimen	Quantity	Collection / Processing Method	Shipping / Recipient Lab	Notes
For all patients consented to biobanking					
Pre-registration	Bone marrow aspirate in EDTA tubes for MRD	1 x 10 ml	Bone Marrow – EDTA (10.1)	Ambient / HEME	1, 2, 4, 5
Pre-registration	Whole Blood in EDTA tubes for Immunophenotyping	2 x 10 ml	Whole Blood – EDTA (11.1)	Ambient / HEME	1, 4
Pre-registration	Whole Blood in EDTA tubes for PBMC	2 x 10 ml	Whole Blood - EDTA (11.1)	Ambient / HEME	1, 4
Pre-registration	Whole Blood for serum	3 x 1ml aliquots	Whole Blood – Red top (11.2)	Frozen / HEME	1, 4
Pre-registration	Unstained slides from Bone Marrow Core Biopsy	10-15 x 4-6um	Bone Marrow Unstained Slides (9.2)	Ambient / HEME	1, 4, 6
Cycle 3 Day 15 (+/- 3 days)	Whole Blood in EDTA tubes for Immunophenotyping	2 x 10 ml	Whole Blood - EDTA (11.1)	Ambient / HEME	4
Cycle 3 Day 15 (+/- 3 days)	Whole Blood in EDTA tubes for PBMC	2 x 10 ml	Whole Blood - EDTA (11.1)	Ambient / HEME	4
Cycle 3 Day 15 (+/- 3 days)	Whole blood for serum	3 x 1ml aliquots	Whole Blood – Red top (11.2)	Frozen / HEME	4
Upon achievement of CR/sCR	Bone marrow aspirate in EDTA tubes for MRD	1 x 10 ml	Bone Marrow – EDTA (10.1)	Ambient / HEME	3, 4, 5
Cycle 12	Bone marrow aspirate in EDTA tubes for MRD	1 x 10 ml	Bone Marrow – EDTA (10.1)	Ambient / HEME	2, 4, 5
Cycle 12	Whole Blood in EDTA tubes for Immunophenotyping	2 x 10 ml	Whole Blood - EDTA (11.1)	Ambient / HEME	4
Cycle 12	Whole Blood in EDTA tubes for PBMC	2 x 10 ml	Whole Blood - EDTA (11.1)	Ambient / HEME	4
Cycle 12	Unstained slides from Bone Marrow Core Biopsy	10-15 x 4-6um	Bone Marrow Unstained Slides (9.2)	Ambient / HEME	4, 6
Cycle 12	Whole blood for serum	3 x 1ml aliquots	Whole Blood – Red top (11.2)	Frozen / HEME	4

Study Drug Discontinuation	Whole Blood in EDTA tubes for Immunophenotyping	2 x 10 ml	Whole Blood - EDTA (11.1)	Ambient / HEME	4
Study Drug Discontinuation	Whole blood for serum	3 x 1ml aliquots	Whole Blood – Red top (11.2)	Frozen / HEME	4
Study Drug Discontinuation	Unstained slides from Bone Marrow Core Biopsy	10-15 x 4-6um	Bone Marrow Unstained Slides (9.2)	Ambient / HEME	4, 6

1. To be submitted any time prior to C1D1.
2. Submitted only if patient is in CR/sCR.
3. Only if patient was not in CR/sCR at pre-registration and achieves CR/sCR while on study.
4. Collection is optional for patients but requires all sites offer to patients during consent. Please see protocol-specific consent documents.
5. There is no alternative for the bone marrow specimen.
6. When bone marrow biopsies are available as standard of care, send a minimum of 10 and up to 15 x 4-6um unstained slides from the bone marrow core biopsy tissue block (FFPE).

7. Biospecimen Collection Kits

7.1 Blood, Bone Marrow, and Tissue Specimens

7.1.1 There is no independent "kit" for submission of slides, frozen serum, peripheral blood or bone marrow aspirates. Sites are responsible for acquiring materials for collection and shipping of these specimens.

7.1.2 Please see **Section 12 – Biospecimen Shipping** for specific instructions on shipping to HEME.

8. Biospecimen Labeling and Tracking

8.1 All research biospecimens (vacutainer tubes) MUST be labeled with the protocol number (A062102), Alliance patient ID number, patient initials (Last, First, Middle), the date and time (if applicable) of collection and specimen type (e.g. bone marrow, peripheral blood).

8.2 Label all containers and vials with an indelible, solvent-resistant marker when they are at ambient temperature.

8.3 Do not affix any labels to vials or tubes. Label the collection containers directly with the marking pen.

8.4 All biospecimens that are collected and sent to HEME must be **logged and tracked in BioMS**. The BioMS system is a web-based application that tracks the collection and shipping of biospecimens. Once individual biospecimens are logged and 'shipped' in the BioMS system, a packing manifest will be created by the system. This manifest must be printed out and must accompany all biospecimen shipments. To become familiar with the BioMS system and for further information about training, access, and use, please contact the BioMS Help desk at: 1-855-55-BIOMS or bioms@alliancenctn.org.

8.5 In the event that BioMS cannot be accessed, please complete a BioMS Specimen Log and Shipping Manifest form which can be found here- **http://tinyurl.com/alliance-biomscontingency**.

9. Tissue Collection

9.1 Overview.

9.1.1 Please refer to protocol-specific instructions for procedures related to actual tissue procurement from individual participants. The method for research tissue procurement (needle core biopsy, sampling of surgically resected tumor) is dependent upon the disease site and the individual patient.

9.1.2 When procuring tissue biospecimens by any method, when possible, avoid tissue that is grossly necrotic, hemorrhagic, fatty, or fibrous. If in doubt, briefly (1 min or less) place the tissue segment in a sterile specimen cup containing physiologic (normal) saline to rinse the tissue. Necrotic, hemorrhagic, and fatty tissue will generally dissolve or float on the surface while tumor and parenchymal tissue will remain intact and sink to the bottom of the cup.

9.1.3 During warm weather months, paraffin blocks and slides should be shipped in an insulated container that contains a refrigerant pack, to avoid heat > 25 degrees C (77degrees F) that may melt paraffin and damage the tissue specimens.

9.2 Bone Marrow Unstained Slides

9.2.1 A set of 10-15 bone marrow unstained slides must be sent. A minimum of 10 slides up to 15 should be sent. If fewer than 10 unstained slides can be submitted, please submit as many unstained slides as possible. Please follow the procedures below for submitting unstained tissue slides.

# of slides	Section thickness	Slide type	Purpose
10-15	4-6 micron	Charged	IHC

9.2.2 Serial, tissue sections should be cut fresh from the appropriate formalin fixed, paraffin embedded tissue block.

9.2.3 Cut sections at 4-6 micron thickness as indicated onto charged slides.

9.2.4 Ensure that each slide is labeled with the Alliance patient ID number, the institutional surgical pathology number and block ID, and the slide serial section number (1, 2, 3, etc.).

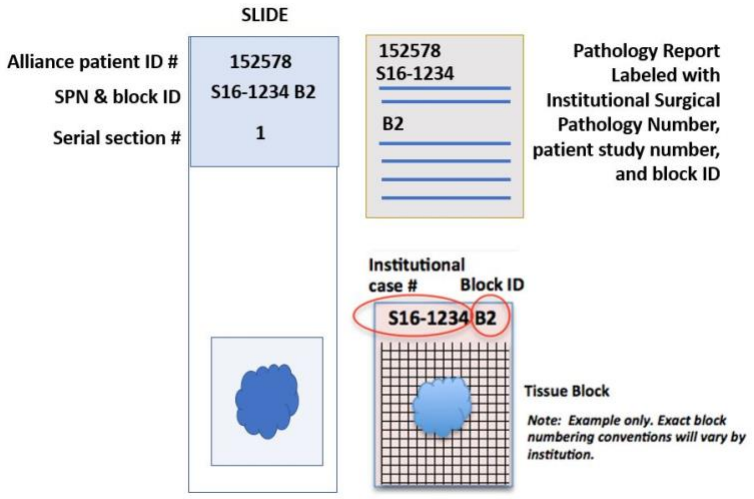
9.2.5 Do not label slides with adhesive labels. Write or print information on the textured surface of the slide with indelible, solvent-resistant ink.

9.2.6 No adhesives or other additives should be used in the water bath.

9.2.7 Mount only one tissue section per slide. Make certain sections are placed on the painted / textured side of the slide.

9.2.8 When placing the sections onto the slides, ensure that the tissue is placed on the bottom third of the slide. Ensure that each serial section from the block is placed in the same orientation on each slide.

9.2.9 See figure below for proper mounting and labeling.



9.2.10 Air dry slides for 12-24 hours prior to shipping. Do not oven dry slides.

9.2.11 Use slide mailers or a slide box to ship slides. Slides should not be touching each other. Ensure that slides from only one patient are placed in one slide mailer.

9.2.12 Include a copy of a **de-identified pathology report** with all slide submissions.

10. Bone Marrow Collection

10.1 Bone Marrow Aspirate- EDTA tubes (no processing)

10.1.1 Collect 10ml of bone marrow following standard institutional protocol into 10ml EDTA tube. Invert tube 10 times.

10.1.2 Store tube with bone marrow at ambient temperature. Do not freeze or refrigerate the tube. The tube must be shipped on the same day it is collected and must be received by the recipient lab within 24 hours of collection. **Please collect and ship specimens on Monday—Friday only. Specimens should not be collected or shipped on Saturday, Sunday, the day prior to a holiday, or on a holiday.** Ensure that the tube is shipped at ambient temperature with a refrigerated (not frozen) gel pack to avoid freezing.

11. Blood Collection

11.1 Whole blood- EDTA Tubes for Immunophenotyping and PBMC (no processing)

11.1.1 Collect 10 ml of peripheral blood by standard venous phlebotomy technique into each of the EDTA tubes. Invert the tubes 10 times.

11.1.2 Store the EDTA tubes with peripheral blood at ambient temperature. Do not freeze or refrigerate the tubes. The whole blood must be shipped on the same day it is collected and must be received within 24 hours of collection. **Please collect and ship specimens on Monday—Friday only. Specimens should not be collected or shipped on Saturday, Sunday, the day prior to a holiday, or on a holiday.** Ensure that the EDTA tubes are shipped at ambient temperature with a refrigerated (not frozen) gel pack to avoid freezing.

11.2 Serum Processing- Red Top no additive

11.2.1 Collect 10 ml of whole blood by standard venous phlebotomy technique into the red top (plain glass with clot activator) tube. Do not collect whole blood into a "tiger top" / "SST" / "gel tube." Invert tube 10 times

11.2.2 Allow blood to clot for 30 minutes.

11.2.3 Label 3 cryovials as instructed in **section 8**. Make certain each vial is labeled completely and identically.

11.2.4 Spin blood in vacutainer tube at 4 degrees in a clinical centrifuge using standard programming for serum separation. Usually this is 1200 xG (actual speed will depend upon the centrifuge) for 10 minutes.

11.2.5 Carefully remove 3 ml of serum (without touching the clot layer) and divide into 3, 1 ml labeled cryovials.

11.2.6 Freeze serum containing cryovials on dry ice or a -70 to -90 degree Celsius ultralow freezer. Store at -70 to -90 degrees until ready for shipment on dry ice.

12. Biospecimen Shipping

12.1 Overview

12.1.1 Place the original, completed copy of the BioMS packing manifest in the shipment. Do not send specimens without a completed BioMS Packing Manifest or substitute "BioMS Downtime Form." Biospecimens cannot be accepted without this completed form.

12.1.2 Specimens should be sent according to IATA guidelines. All biospecimens should be shipped on the same day that they are collected (Monday – Friday). Whole Blood Biospecimens must be received by the recipient lab within 24 hours of collection. If collected biospecimens cannot be shipped on the same day that they are collected (e.g. Saturday, Sunday or holiday collections), please contact the Alliance Biorepository Program Manager: 1-314-747-4402 or alliance@email.wustl.edu for further instructions, **at least** 24 hours prior to anticipated collection.

12.1.3 Frozen serum aliquots should be placed in a biohazard bag inside of a Styrofoam cooler and covered with 3 to 4 lbs (2 kg) of commercially-prepared dry ice. Pellets or chunks are preferred. Make sure the box is filled with dry ice and the weight of the dry ice is noted on the dry ice label on the outside of the shipping container. It is the local sites' responsibility to obtain dry ice when shipping frozen specimens. **Frozen aliquots should be shipped to the Biorepository within 30 days of collection. Batch shipment of frozen aliquots is allowed.**

12.1.4 **Do not ship to HEME on Saturday, Sunday or on the day before a nationally recognized holiday.**

12.1.5 All specimens should be shipped via priority overnight shipping. **If shipping specimens on a Friday, please indicate Saturday delivery on the waybill.**

12.1.6 Shipping to HEME

Ship container for PRIORITY OVERNIGHT DELIVERY according to IATA guidelines and standard institutional policies and using the preferred vendor.

Ship to:
Alliance Hematologic Malignancy Biorepository (HEME)
The Arthur G. James Cancer Hospital and Solove Research Institute
300 West Tenth Avenue, Lobby
Columbus, OH 43210
Phone: **614-366-6295**
Fax: 614-688-4755

13. Biospecimen Receipt and Quality Assurance Measures

13.1 Upon receipt, all physical biospecimens received will be reconciled with what is recorded on the BioMS packing manifest. Any discrepancies noted will be communicated to the Program Manager who will contact the submitting site for reconciliation.

13.2 Upon receipt, any biospecimen received that is not in appropriate physical condition (broken vials, frozen samples that are thawed, ambient samples that are frozen) will be reported to the Program Manager, who will contact the submitting site for reconciliation.

13.3 Aliquoted biofluids will be stored under liquid nitrogen vapor.

13.4 Unstained slides from bone marrow core biopsy will be stored at -80C upon receipt and reconciliation by HEME.

13.5 All biospecimens will remain in storage until additional processing or review is requested in writing by the appropriate protocol PI.

14. Document History

Version	Description and Justification of Change	Author	Effective Date
1.0	New	KAL	03/29/2024