COMPASS®
SAMPLE PREPARATION HANDBOOK
A GUIDE TO BONE MARROW AND PERIPHERAL BLOOD SAMPLE PREPARATION FOR NEOGENOMICS
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I. SAMPLE REQUIREMENTS

COMPASS® for Blood Evaluation Optimal Specimen Requirements

<table>
<thead>
<tr>
<th>TEST/TECHNOLOGY</th>
<th>PERIPHERAL BLOOD (WBC≤5,000 INCLUDE 3 TUBES)</th>
<th>PERIPHERAL BLOOD SMEAR</th>
</tr>
</thead>
<tbody>
<tr>
<td>COMPASS® for Blood Evaluation</td>
<td>5-6 ml in <strong>two (2)</strong> green top (sodium heparin) tubes AND 2-3 ml in purple top (EDTA) tube</td>
<td>2 bedside smears</td>
</tr>
</tbody>
</table>

(Includes COMPASS® Consultation Report, clinical pathology evaluation, blood morphology, flow cytometry and genomic testing as medically necessary; a consultative review and correlation with prior findings is performed by a NeoGenomics hematopathologist.)


COMPASS® Bone Marrow Evaluation Optimal Specimen Requirements

<table>
<thead>
<tr>
<th>TEST/TECHNOLOGY</th>
<th>PERIPHERAL BLOOD</th>
<th>PERIPHERAL BLOOD SMEAR</th>
<th>BONE MARROW ASPIRATE</th>
<th>BONE MARROW SMEAR</th>
<th>BONE MARROW</th>
</tr>
</thead>
<tbody>
<tr>
<td>COMPASS® Bone Marrow Evaluation</td>
<td>Fill the 6 ml EDTA (large purple top) tube with peripheral blood</td>
<td>2-3 bedside smears (OR 1 ml blood in purple top [EDTA] tube)*</td>
<td>Three 1-2 ml aspirate in each of the 4 ml sodium heparin (small green top) tubes. One 1-2 ml aspirate in the 4 ml EDTA (small purple top) tube. Please Note: Do not mix aspirate and peripheral blood in the same tube.</td>
<td>4-6 bedside smears</td>
<td>Form aspirate clot, place in formalin jar #2, and label appropriately.</td>
</tr>
</tbody>
</table>

(Includes COMPASS® Consultation Report, clinical pathology evaluation, bone marrow and/or blood morphology, flow cytometry, genomic testing as medically necessary; a consultative review and correlation with prior findings is performed by a NeoGenomics hematopathologist.)

The samples for a complete bone marrow evaluation originate from multiple sources:
1. Aspirate (liquid marrow sample)
2. Core (tissue sample obtained from the needle’s penetration of the bone marrow)
3. Clot (congealed aspirate)
4. Peripheral Blood

* Peripheral blood and bone marrow aspirate smears prepared immediately after collection have been shown to provide significantly better morphology than smears made at a later time or made from an anticoagulated specimen.
PERIPHERAL BLOOD SAMPLES

• For COMPASS® for Blood Evaluation, at least 2 bedside smears along with a minimum of 5-6 ml of peripheral blood should be submitted in a total of two (2) green top (sodium heparin) tubes AND 2-3 ml of peripheral blood in a purple top (EDTA) tube.

• For COMPASS® for Blood or Bone Marrow Evaluation, at least 2 bedside smears should be submitted.

• If submitting bone marrow samples, please include the purple top (EDTA) tube used for the CBC, collected the same day as the bone marrow procedure when available.

DRY TAP

• For COMPASS® for Bone Marrow Evaluation, create touch preparation slides of core on 2 slides. Place core biopsy in formalin jar and label appropriately.

• Place one or more additional cores in RPMI. It is recommended RPMI sits at room temperature for 30 minutes prior to use. (RPMI is not included in kit content. Limited quantities may be requested by calling Client Services at 866.776.5907).

PLEASE REVIEW THESE RESOURCES FOR HELPFUL TIPS:

Better Bone Marrow Biopsies in the On-demand Webinar series
BONE MARROW ASPIRATE SAMPLES

Marrow aspirate should be collected in two syringes, one for each pull.

1. First pull/syringe of aspirate: 1-2 ml
   a. Smears made on glass slides. Bone marrow smears prepared immediately after collection have been shown to provide significantly better morphology than smears made at a later time or made from an anticoagulated specimen. Aspirate smears should be made quickly to avoid clotting. Heparin may be used to prevent clotting of the aspirate; however, heparin causes cytologic artifacts and should be avoided whenever possible. If needed, aspirate several ccs of sodium (Na) heparin into the syringe and then expel it. This coats the inner surface of the syringe to prevent clotting, but introduces a minimum amount of heparin into the specimen. Na heparin of at least 1,000 units/mL, preferably 2,000 units/mL, should be used.
   b. A clot formed from letting the remaining aspirate congeal. Place the formed or unformed aspirate clot in formalin.

2. Second pull/syringe of aspirate: 4-6 ml
   a. Place approximately 2 ml of the aspirate into each green top (sodium heparin) tube. This material may be used for flow cytometry, cytogenetics and FISH studies.
   b. Any remaining aspirate should be put into a purple top (EDTA) tube. This material may be used for additional ancillary studies, such as molecular analyses.

BONE MARROW CORE SAMPLE

After touch preparation slides are made, the entire core sample should be placed in one of the formalin vials and labeled appropriately.
II. SMEAR AND CLOT PREPARATION

PERIPHERAL BLOOD SMEAR PREPARATION

Required Items:
• 4-6 glass slides labeled with the patient’s name, date and the sample type (PB for peripheral blood)
• Gloves
• Blood draw kit

PREPARATION:
1. Smears should be prepared immediately after the patient draw from a purple top (EDTA) tube or from the syringe in case of a syringe draw. Two well prepared peripheral blood smears should be submitted.

2. Upon completion of the draw, deliver one (1) small drop of blood to a slide on the end closest to the frosted edge. (Figure 1)

3. After placing the blood on the slide, place the slide on a flat surface.

4. With one hand, hold the edges of the frosted end of the slide between thumb and forefinger on the flat surface.

5. Pick up a second slide with the free hand. This slide will be used to spread the blood across the resting slide surface.

6. Position the spreader slide at a 15-30° angle to the slide on the flat surface. This angle should be maintained throughout the procedure and when properly established, the raised edge of the spreader slide will be about one (1) inch above the resting slide surface. (Figure 2)

7. Place the end of the spreader slide to make contact with the surface of the resting slide near the drop of blood. Pull it towards the drop of blood and allow it to make contact with it. The drop of blood should spread to the edges of the spreader slide. (Figure 3)

8. Push the spreader slide smoothly and rapidly over the length of the resting slide while maintaining the 15-30° angle and firm pressure. Two well prepared peripheral blood smears should be submitted.
   a. When properly prepared, the blood smear should make a gradual transition from thick to thin.
   b. It is acceptable for only half of the slide to be covered by the thin area with the maintenance of a gradual transition. NOTE: The feathered edge is the most important part of the smear. (Figure 4)
   c. If the feathered edge is not entirely visible on the glass slide, repeat the smear using a smaller drop of blood.

9. Allow the slides to air-dry completely. NOTE: Wet smears may stick to each other and the slide cassette, and therefore be unusable.

10. Please make sure that all slides are labeled with the patient’s name, date, and sample type (PB for peripheral blood).

11. Place air-dried slides into plastic slide cassette.
BONE MARROW ASPIRATE SMEAR PREPARATION

Required Items:
- 6-8 clean, glass slides labeled with the patient’s name, date and the sample type (BM for bone marrow)
- Sterile petri dish (disposable ones are available)
- Bone marrow biopsy/aspiration tray (from NeoGenomics or other vendor)
- NeoGenomics collection kit
- Optional: Pasteur pipettes

SLIDE PREPARATION:

NOTE: The marrow aspirate is a mixture of blood and marrow particles (spicules). It is critical to select marrow particles for making the aspirate smears so they will be representative of the bone marrow, and not diluted by blood.

1. Lay out 6 to 8 clean labeled slides. Please label all slides with the patient’s name, date and sample type (BM for bone marrow).

2. Place the initial bone marrow aspirate in a sterile Petri dish. Tip the Petri dish slightly to expose the marrow particles which look like tiny grains of sand. (Figure 5) Notify the person collecting the specimen whether or not particles are present so they may determine if another aspirate specimen is needed. If no marrow particles can be obtained (“dry tap”), go to step 3. If present, select individual marrow particles to make the aspirate particle smear slides. This is done by lightly scraping the non-frosted end of one glass slide against the surface of the Petri dish to pick up a particle. (Figure 6a) Alternately, a Pasteur pipette may be used to pick up the marrow particle which is then dropped just below the middle of a glass slide. Whichever method is used, this slide is then turned over and gently placed on the surface of a labeled glass slide. (Figure 6b) As soon as the liquid spreads out between the two slides, pull the top slide across the lower slide with a steady motion and without pressure, spreading the specimen along the length of that slide. (Figure 7)
   a. When properly prepared, the specimen should be spread evenly along the bottom slide in an elongated, oval shape, and spicules are visible. (Figure 8)
   b. The smear should be of equal thickness across the length of the slide. A smear that is too thick cannot be read.

3. If no marrow particles can be obtained (“dry tap”), lightly scrape the end of one glass slide against the Petri dish to pick up a small amount of the liquid aspirate. This slide is then gently placed on the surface of a labeled glass slide. (Figure 6) As soon as the liquid spreads out between the two slides, pull the top slide across the lower slide with a steady motion and without pressure, spreading the specimen along the length of that slide. (Figure 7)
   a. When properly prepared, the specimen should be spread evenly along the bottom slide in an elongated, oval shape. (Figure 8)
   b. The smear should be of equal thickness across the length of the slide. A smear that is too thick cannot be read.
4. The remaining specimen in the Petri dish is set aside and allowed to clot. Refer to BONE MARROW ASPIRATE CLOT PREPARATION below.

5. Allow slides to air-dry completely. **NOTE: Wet smears may stick to each other and the slide cassette, and therefore be unusable.**

6. Place air-dried slides into plastic slide cassette. Please be certain that all slides are labeled with the patient’s name, date, and sample type (BM for bone marrow).

**BONE MARROW ASPIRATE CLOT PREPARATION:**

1. After making smears from the aspirate specimen (see above), the remaining specimen in the Petri dish is set aside to clot.

2. Once the marrow collection procedure is completed, this specimen is submitted in a formalin vial whether or not it has clotted.

**BONE MARROW BIOPSY TOUCH IMPRINT PREPARATION:**

When no particles are obtained (“dry tap”), the core biopsy may be the only source of bone marrow for microscopic examination; however, it is best to always make touch imprints in case the aspirate particle smears are suboptimal.

1. To prepare touch imprints, the bone marrow core is expelled from the biopsy needle onto a labeled glass slide. The fresh core biopsy is then gently rolled between the first and a second labeled glass slide for a distance of about an inch. Alternately, if the core biopsy will not roll, you can touch the second glass slide to the surface of the core biopsy multiple times along a distance of about an inch. Repeat until you have two well-made touch preparations. The core biopsy is then placed into a formalin vial for transport.

2. Allow slides to air-dry completely. **NOTE: Wet smears may stick to each other and the slide cassette, and therefore be unusable.**

3. Place air-dried slides into plastic slide cassette. Please be certain that all slides are labeled with the patient’s name, date, and sample type (TP for touch prep).
How to Fill Out a Test Requisition Form

1. **Client Information**
   - Requisition Completed by: Signature and Date.
   - Account Number: If you do not know or do not have an account number, NeoGenomics will create and/or enter it when we receive the order.
   - Ordering Physician: Name (Last, First), NPI #
   - Treating Physician: Name (Last, First), NPI #

2. **Test Authorization and Physician Signature**
   - Important information to support that the testing is medically necessary for the patient’s condition, which supports claim payment for both clients and NeoGenomics

3. **Billing Information**
   - **READ CAREFULLY TO PREVENT A DELAY IN RECEIVING RESULTS**
   - **Required:** Mark Specimen Origin
   - Complete Specimen Origin, choose at least one Bill to option and attach Prior Authorization to avoid unnecessary billing charges.
   - **Client Bill:** All charges billed to client (listed in #1 above)
   - **Insurance/Medicare/Medicaid:** All charges billed to insurance except when payer follows CMS guidelines and patient status indicated as inpatient or outpatient; if so, TC charges billed to client (listed in #1 above), PC charges to insurance
   - **Patient/Self Pay:** All charges billed to patient
   - **Split Billing - Client (TC) and Insurance (PC):** All TC charges billed to client (listed in #1 above), all PC charges to insurance
   - **OP Molecular to MCR, all other testing to client:** Molecular testing billed to Medicare, all other testing to client (listed in #1 above)
   - **Bill charges to other Hospital/Facility:** If client other than listed in #1 above is to be billed, please indicate name and address here

4. **Clinical Information / Patient History**
   - Accurate diagnosis information helps inform health insurance coverage and supports faster turn-around time by preventing the need for requests for additional information from our Client Services, Billing and Pathology groups.
   - To prevent a delay in patient care and to avoid billing issues, include:
     - **Required:** Diagnosis Code/ICD Code, Reason for Referral, CBC, Stage or Disease status
     - **Attachments:** Supplementary test results may assist our pathologists in their assessment of the case. Scan and include with submission. Utilizing online ordering will make this process easier. For more information or to order online, visit [https://neogenomics.com/online-ordering](https://neogenomics.com/online-ordering).

5. **Patient Information**
   - Patient Legal Name: (Last, First, MI), Sex, Date of Birth, and Medical Record #

6. **Specimen Information**
   - Provide information only for the specimen type that is being submitted.
   - All orders submitted **Required:** Specimen ID, Date (collection, retrieved, discharge), specimen type and prep method
PACKING THE SHIPPER

In order to ensure optimal analysis of patient samples, specimens should be handled properly from the time of collection to the time of assay performance. Please ensure the specimens are submitted in the proper tubes/vials by referring to the Optimal Specimen Requirements, listed on the back of the requisition form, NeoGenomics Testing Directory, or at the beginning of this guide.

All samples should be submitted to NeoGenomics within 24 hours of collection. For specimens drawn or stored beyond 24 hours, please inquire with NeoGenomics Client Services at 866.776.5907 regarding test performance.

<table>
<thead>
<tr>
<th>NEOGENOMICS KIT COMPONENT (TOTAL NUMBER)</th>
<th>USAGE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plastic slide holders (2) with slides</td>
<td>2-5 smears each</td>
</tr>
<tr>
<td>Green top (sodium heparin) tubes (2)</td>
<td>Bone marrow aspirate</td>
</tr>
<tr>
<td>Purple top (EDTA) tubes (2)</td>
<td>Peripheral blood</td>
</tr>
<tr>
<td>Purple top (EDTA) tubes (2)</td>
<td>Peripheral blood or bone marrow aspirate for smear preparation by NeoGenomics.</td>
</tr>
<tr>
<td>Purple top (EDTA) tubes (2)</td>
<td>Peripheral blood or bone marrow aspirate for molecular testing</td>
</tr>
<tr>
<td>Formalin Vials (2)</td>
<td>Bone marrow core (1)</td>
</tr>
<tr>
<td>Formalin Fix Specimens Only bag</td>
<td>Bone marrow clot (1)</td>
</tr>
<tr>
<td>Test requisition form</td>
<td>For shipping the two Formalin vials</td>
</tr>
<tr>
<td>Test requisition form</td>
<td>To submit patient, billing, and history info, etc., as well as indicate tests to be performed</td>
</tr>
<tr>
<td>Cold pack</td>
<td>To help protect samples from excessive heat (90°F or higher)</td>
</tr>
<tr>
<td>Large biohazard bag</td>
<td></td>
</tr>
<tr>
<td>FedEx’ Clinical Pak’</td>
<td></td>
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</tbody>
</table>