

**Baseline/Randomization/Follow-Up Phlebotomy Form: Processing and Inventory  
Instructions  
PHP Version C, 02/18/2005  
QxQ Date: 03/02/2005**

## **GENERAL INSTRUCTIONS**

The Baseline/Randomization/Follow-up Phlebotomy Form: Processing and Inventory (PHP) is completed during the screening/baseline combination visit or the randomization clinic visit and at follow-up visits. It is used to document data during the phlebotomy process.

If specimen processing is done by a laboratory, they must be certified and familiar with and understand chapter 6: Specimen Collection and Processing, in the FAVORIT Manual of Operations (MOP). The Study Coordinator should insure an updated copy of this chapter is available in the laboratory as reference. The appropriate sections of the paper form must be completed by the Phlebotomist/Laboratory (processor) during specimen processing and then recorded in the data management system (DMS) by the Study Coordinator at the completion of the day's activities, and when shipping occurs, respectively. If processing is done by the Study Coordinator, s/he must be certified and familiar with and understand MOP chapter 6: Specimen Collection and Processing, and chapter 14: Administrative Procedures, prior to completing this form.

The form header information (ID, Contact Occasion, Sequence Number, Name and Initials) is completed as described in the MOP. Data related to storage and shipping must be appropriately recorded by the Study Coordinator. If a laboratory is processing the specimens, they should receive the form with **only the patient's ID Number**, Contact Occasion and Sequence Number. The Study Coordinator should complete the patient's Name after the laboratory has completed processing.

Apply pre-printed specimen ID labels to the vacutainer tubes and aliquot vials. If a label is missing or unusable, prepare a hand-printed label using a fine-point cryomarker, writing on the "white area" of a blank FAVORIT specimen label. Record ID, Contact Occasion and tube type on the label. Verify the correct ID is used for the participant whose specimen is being drawn.

All specimens collected during the baseline, randomization and follow-up visits are for long-term storage; none will be shipped immediately to the Central Lab. You should have a separate storage box for each aliquot type. Each storage box should contain the type of aliquot, contact occasion, beginning and ending date of the specimens and a box number.

Two lavender, one marble, one light blue tube and one urine void will be collected.

To avoid elevated plasma homocysteine values and to ensure optimal retrieval of the buffy coat, sample should be centrifuged and processed within 45 minutes of collection. All processing **must** be completed within 4 hours of specimen collection.

### **Aliquotting Information:**

Upon completion of aliquotting, all vials should be tightly capped and frozen; remaining cells should be discarded. Samples should be placed **immediately** into the freezer.

Two 10mL lavender top tubes (LT label) with EDTA additive are collected. One tube is used to create seven vials LT1-LT7; the other is used to create four vials LT8-LT11. The 10mL marble top tube (MT label) for serum is used to create five vials MT1-MT5. The 4mL light blue top tube (BT label) for sodium citrate plasma is used to create two vials BT1 and BT2. The 5oz urine void container (UR label) is used to create three vials UR1-UR3.

## **SPECIFIC INSTRUCTIONS**

1. Time of centrifugation:
  - a. Record the time of centrifugation using a 12-hour clock format, filling in the fields using leading zeroes where necessary.
  - b. Record the time of day of centrifugation indicating AM or PM.
2. Placement of vials in the freezer:
  - a. Record the time vials were placed in the freezer using a 12-hour clock format, filling in the fields using leading zeroes where necessary.
  - b. Record the time of day that vials were placed in the freezer indicating AM or PM.
3. Record date of processing using the US order (month/day/year).
4. Record whether the data was collected directly into the date entry system on the computer or whether it was recorded on a paper form.
5. Record processor's three initials. If s/he only has two initials, then record the 1<sup>st</sup> name initial in the first box, the last name initial in the 2<sup>nd</sup> box and leave the third box blank.

Items 6-26 record data on collection, hemolysis, and storage of aliquots. Each item has three data entry parts: "a", "b", and "c". General instructions for these three parts are as follows:

- a. If the specimen was collected, record "Yes", otherwise record "No".
- b. If the specimen was hemolyzed, record "Yes", otherwise record "No".
- c. Using leading zeroes, record the three-digit number of the yellow storage box used for long-term storage of the vial. The vial must be placed in the box allotted for its aliquot type and contact occasion in an upright position and stored at -80°C. The label on the box should have the aliquot code (i.e., LT1, LT2, LT3, etc.), contact occasion, beginning and end dates, and box number for the aliquot code.

Note: Items 10-12 and 24-26 do not have a part "b".

Following is an item-by-item description of each aliquot:

Items 6-12 refer to vials from the first lavender top tube.

6. LT1 (tHcy), i.e., lavender top vial #1, is used to measure total homocysteine (tHcy). 500  $\mu$ L is required for the sample aliquot.
7. LT2 (B12/folate), i.e., lavender top vial #2, contains specimen for measuring B12/folate. 1000  $\mu$ L is required for the sample aliquot.
8. LT3 (PLP), i.e., lavender top vial #3, contains specimen for measuring pyridoxal phosphate. 500  $\mu$ L is required for the sample aliquot.
9. LT4 (plasma archive 1), i.e., lavender top vial #4, contains the first plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.
10. LT5 (buffy coat), i.e., lavender top vial #5, contains specimen of white blood cells (buffy coat). Approximately 500  $\mu$ L is required for the sample aliquot.
11. LT6 (RBCs), i.e., lavender top vial #6, contains the first specimen of red blood cells for the archive. 1000  $\mu$ L is required for the sample aliquot.
12. LT7 (RBCs), i.e., lavender top vial #7, contains the second specimen of red blood cells for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 13-16 refer to vials from the second lavender top tube.

13. LT8 (chol/trig), i.e., lavender top vial #8, is used to measure cholesterol triglyceride. 500  $\mu$ L is required for the sample aliquot.
14. LT9 (HDL), i.e., lavender top vial #9, is used to measure high-density lipoprotein. 500  $\mu$ L is required for the sample aliquot.
15. LT10 (LDL), i.e., lavender top vial #10, is used to measure low-density lipoprotein. 500  $\mu$ L is required for the sample aliquot.
16. LT11 (plasma archive 2), i.e., lavender top vial #11, contains the second plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 17-21 refer to vials from the marble top tube.

17. MT1 (creatinine), i.e., marble top vial #1, contains specimen for measuring creatinine. 500  $\mu$ L is required for the sample aliquot.

18. MT2 (glucose), i.e., marble top vial # 2, contains specimen for measuring glucose. 500  $\mu$ L is required for the sample aliquot.
19. MT3 (fructosamine), i.e., marble top vial #3, contains specimen for measuring fructosamine. 500  $\mu$ L is required for the sample aliquot.
20. MT4 (serum archive 1), i.e., marble top vial #4, contains the first serum specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.
21. MT5 (serum archive 2), i.e., marble top vial #5, contains the second serum specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 22-23 refer to vials from the light blue top tube.

22. BT1 (BT archive 1), i.e., light blue top vial #1, is used to collect the first citrate plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.
23. BT2 (BT archive 2), i.e., light blue top vial #2, is used to collect the second citrate plasma specimen for the archive. The remainder is required for the sample aliquot.

Items 24-26 refer to urine void vials from the urine void container.

24. UR1 (creatinine), i.e., urine void vial #1, is used for measuring creatinine. 1500  $\mu$ L is required for the sample aliquot.
25. UR2 (microalbumin), i.e., urine void vial #2, is used for measuring microalbumin. 1500  $\mu$ L is required for the sample aliquot.
26. UR3 (archive), i.e., urine void vial #3, contains urine for the archive. 1500  $\mu$ L is required for the sample aliquot.

### **Blind Replicate Matching (BRM) Instructions**

Items 27 through 49 refer to Blind Replicate Matching (BRM) specimens used for quality control. These are specimens processed from extra tubes drawn from participants and labeled with a BRM ID assigned by the DCC. This extra tube must be the last tube drawn from the participant. Never perform a second stick to obtain a BRM; instead obtain the BRM tube from the next appropriate participant. BRM specimens are stored and shipped along with participant's specimens. See MOP chapter 11, for more information.

When it is time for a site to start a BRM collection for a randomization or follow-up visit, the DCC will email a blind replicate matching notification request and a BRM ID sheet which contains the BRM ID. When you have been notified to do a blind replicate, a complete set of duplicate vials must be created and labeled using the BRM ID. Only one extra tube of blood may be drawn from a participant, therefore multiple participants will need to donate an

extra tube to complete the BRM. Each of the participants will have the items 27 through 49 entered in their respective PHP forms; all will show the same BRM ID in item 28. For example, at the randomization or follow-up visit at least four participants will be used to complete the BRM, one will have data for the lavender top tube#1, one will have data for the marble to tube, one for the light blue top tube, and one will have data for the lavender top tube#2. Any one of the four participants can donate an extra urine sample or a fifth participant can donate the urine sample and all will have the same BRM ID recorded in item 28.

**The following is an example of a BRM request and collection scenario:**

- The clinic is sent an email containing a BRM notification request and a BRM ID sheet. The notification sheet and ID sheet contain the BRM ID (e.g., WU12343) that should be used for the BRM samples. The notification sheet indicates that a randomization and follow-up (05) BRM specimens are requested
- Randomization and contact occasion 05 Follow-up ID labels are mailed to clinic for BRM WU12343
- The coordinator sees on his/her schedule that Susan P., a randomization participant is due in today
- The coordinator takes one extra lavender top tube#1 of blood from Susan P. and puts the BRM label WU12343 for contact occasion 01 on the extra lavender top tube (note the extra tube must be the last tube drawn)
- The coordinator processes the extra BRM lavender top tube#1 along with the study participant's lavender top tube (aliquot each into 7 vials)
- Susan P.'s ID label (e.g., WU24680) is placed on the BRM ID sheet under lavender top tube #1 for the randomization visit to keep track of which participant donated which specimen
- The coordinator puts a check mark on the notification sheet under lavender top tube #1 for the randomization contact
- When Susan P.'s PHP is completed, the section on BRM (PHP items 27-49) is completed to reflect the processing of the BRM tube
  
- The next day John B., comes in for his randomization visit
- The coordinator takes one extra marble top tube from John B. (last tube drawn) and attaches the BRM ID label WU12343 for contact occasion 01
- The coordinator processes the extra BRM marble top tube along with the participant's marble top tube.
- John B.'s ID label (e.g., WU45670) is placed on the BRM ID sheet under marble top tube for the randomization visit
- The coordinator puts a check mark on the notification sheet under marble top tube for the randomization contact
- When John B.'s PHP is completed, the section on BRM (PHP items 27-49) is completed to reflect the processing of the BRM tube
  
- Later that day, Fred Z. comes in for his contact occasion 05 follow-up visit

- The coordinator takes one extra marble top tube from Fred Z. (last tube drawn) and puts the BRM ID label WU12343 for contact occasion 05 on the extra marble top tube
- The coordinator processes the extra BRM marble top tube along with the participant's study marble top tube
- Fred Z.'s ID label (e.g., WU13579) is placed on the BRM ID sheet under marble top tube for the follow-up (05) visit
- The coordinator puts a check mark on the notification sheet under marble top tube for the follow-up (05) contact
- When Fred Z.'s PHP is entered, the section on BRM (PHP items 27-49) is completed to reflect the processing of the BRM tube
- The clinic continues to collect the remainder of the BRM samples for the randomization visit which includes the lavender top tube#2, a light blue top tube, and a urine void from various participants. Likewise, the clinic continues to collect the remainder of the BRM sample for the follow-up (05) visit which includes the lavender top tube #1, lavender top tube#2, a light blue top tube, and a urine void from various participants.

### **PHP ITEMS 27-49 BRM**

27. Record whether a BRM tube was collected and processed from the participant. If "No", data entry for the form is complete.
28. Record the BRM ID used to label the tubes and set of BRM aliquot vials being prepared.

### **BRM Aliquotting**

Items 29-49 record data on collection, hemolysis, and storage of BRM aliquots. Each item has three data entry parts: "a", "b", and "c" (except items 33-35 and 46-48 which do not have a part "b"). General instructions for these three parts are as follows:

- a. If the specimen was collected, record "Yes", otherwise record "No".
- b. If the specimen was hemolyzed, record "Yes", otherwise record "No".
- c. Using leading zeroes, record the three-digit number of the yellow storage box used for long-term storage of the vial. The vial must be placed in the box allotted for its aliquot type and contact occasion in an upright position and stored at -80°C. The label on the box should have the aliquot code (i.e., LT1, LT2, LT3, etc.), contact occasion, beginning and end dates, and box number for the aliquot code.

Following is an item-by-item description of each aliquot. Items 29-35 refer to BRM vials from the first lavender top BRM tube.

29. LT1 (tHcy), i.e., lavender top vial #1, is used to measure total homocysteine (tHcy). 500 µL is required for the sample aliquot.

30. LT2 (B12/folate), i.e., lavender top vial #2, contains specimen for measuring B12/folate. 1000  $\mu$ L is required for the sample aliquot.

31. LT3 (PLP), i.e., lavender top vial #3, contains specimen for measuring pyridoxal phosphate. 500  $\mu$ L is required for the sample aliquot.

32. LT4 (plasma archive 1), i.e., lavender top vial #4, contains the first plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

33. LT5 (buffy coat), i.e., lavender top vial #5, contains specimen of white blood cells (buffy coat). Approximately 500  $\mu$ L is required for the sample aliquot.

34. LT6 (RBCs), i.e., lavender top vial #6, contains the first specimen of red blood cells for the archive. 1000  $\mu$ L is required for the sample aliquot.

35. LT7 (RBCs), i.e., lavender top vial #7, contains the second specimen of red blood cells for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 36-39 refer to BRM vials from the second lavender top BRM tube.

36. LT8 (chol/trig), i.e., lavender top vial #8, is used to measure cholesterol triglyceride. 500  $\mu$ L is required for the sample aliquot.

37. LT9 (HDL), i.e., lavender top vial #9, is used to measure high-density lipoprotein. 500  $\mu$ L is required for the sample aliquot.

38. LT10 (LDL), i.e., lavender top vial #10, is used to measure low-density lipoprotein. 500  $\mu$ L is required for the sample aliquot.

39. LT11 (plasma archive 2), i.e., lavender top vial #11, contains the second plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 40-44 refer to BRM vials from the marble top BRM tube.

40. MT1 (creatinine), i.e., marble top vial #1, contains specimen for measuring creatinine. 500  $\mu$ L is required for the sample aliquot.

41. MT2 (glucose), i.e., marble top vial # 2, contains specimen for measuring glucose. 500  $\mu$ L is required for the sample aliquot.

42. MT3 (fructosamine), i.e., marble top vial #3, contains specimen for measuring fructosamine. 500  $\mu$ L is required for the sample aliquot.

43. MT4 (serum archive 1), i.e., marble top vial #4, contains the first serum specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

44. MT5 (serum archive 2), i.e., marble top vial #5, contains the second serum specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 45-46 refer to BRM vials from the light blue top BRM tube.

45. BT1 (BT archive 1), i.e., light blue top vial #1, is used to collect the first citrate plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

46. BT2 (BT archive 2), i.e., light blue top vial #2, is used to collect the second citrate plasma specimen for the archive. The remainder is required for the sample aliquot.

Items 47-49 refer to BRM urine void vials from the BRM urine void container.

47. UR1 (creatinine), i.e., urine void vial #1, is used for measuring creatinine. 1500  $\mu$ L is required for the sample aliquot.

48. UR2 (microalbumin), i.e., urine void vial #2, is used for measuring microalbumin. 1500  $\mu$ L is required for the sample aliquot.

49. UR3 (archive), i.e., urine void vial #3, contains urine for the archive. 1500  $\mu$ L is required for the sample aliquot.



**Randomization/Follow-Up Phlebotomy Form: Processing and Inventory Instructions**  
**PHP Version B, 6/9/2003**  
**QxQ Date: 4/5/2004**

## **GENERAL INSTRUCTIONS**

The Randomization/Follow-up Phlebotomy Form: Processing and Inventory (PHP) is completed during the randomization clinic visit and at follow-up visits. It is used to document data during the phlebotomy process.

If specimen processing is done by a laboratory, they must be certified and familiar with and understand chapter 6: Specimen Collection and Processing, in the FAVORIT Manual of Operations (MOP). The Study Coordinator should insure an updated copy of this chapter is available in the laboratory as reference. The appropriate sections of the paper form must be completed by the Phlebotomist/Laboratory (processor) during specimen processing and then recorded in the data management system (DMS) by the Study Coordinator at the completion of the day's activities, and when shipping occurs, respectively. If processing is done by the Study Coordinator, s/he must be certified and familiar with and understand MOP chapter 6: Specimen Collection and Processing, and chapter 14: Administrative Procedures, prior to completing this form.

The form header information (ID, Contact Occasion, Sequence Number, Name and Initials) is completed as described in the MOP. Data related to storage and shipping must be appropriately recorded by the Study Coordinator. If a laboratory is processing the specimens, they should receive the form with **only the patient's ID Number**, Contact Occasion and Sequence Number. The Study Coordinator should complete the patient's Name after the laboratory has completed processing.

Label vials with the provided pre-printed specimen ID labels. If a label is missing or unusable, prepare a hand-printed label using a fine-point cryomarker, writing on the "white area" of a blank FAVORIT specimen label. Record ID, Contact Occasion and tube type on the label. Verify the correct ID is used for the participant whose specimen is being drawn.

All specimens collected during the randomization and follow-up visits are for long-term storage; none will be shipped immediately to the Central Lab.

Two lavender, one marble, one light blue tube and one urine void will be collected.

To avoid elevated plasma homocysteine values and to ensure optimal retrieval of the buffy coat, sample should be centrifuged and processed within 45 minutes of collection. All processing **must** be completed within 4 hours of specimen collection.

### **Aliquotting Information:**

Upon completion of aliquotting, all vials should be tightly capped and frozen; remaining cells should be discarded. Samples should be placed **immediately** into the freezer.

Two 10mL lavender top tubes (LT label) with EDTA additive are collected. One tube is used to create seven vials LT1-LT7; the other is used to create four vials LT8-LT11. The 10mL marble top tube (MT label) for serum is used to create five vials MT1-MT5. The 4mL light blue top tube (BT label) for sodium citrate plasma is used to create two vials BT1 and BT2. The 5oz urine void container (UR label) is used to create three vials UR1-UR3.

## **SPECIFIC INSTRUCTIONS**

1. Time of centrifugation:
  - a. Record the time of centrifugation using a 12-hour clock format, filling in the fields using leading zeroes where necessary.
  - b. Record the time of day of centrifugation indicating AM or PM.
2. Placement of vials in the freezer:
  - a. Record the time vials were placed in the freezer using a 12-hour clock format, filling in the fields using leading zeroes where necessary.
  - b. Record the time of day that vials were placed in the freezer indicating AM or PM.
3. Record date of processing using the US order (month/day/year).
4. Record processor's three initials. If s/he only has two initials, then record the 1<sup>st</sup> name initial in the first box, the last name initial in the 2<sup>nd</sup> box and leave the third box blank.

Items 5-25 record data on collection, hemolysis, and storage of aliquots. Each item has three data entry parts: "a", "b", and "c". General instructions for these three parts are as follows:

- a. If the specimen was collected, record "Yes", otherwise record "No".
- b. If the specimen was hemolyzed, record "Yes", otherwise record "No".
- c. Using leading zeroes, record the three-digit number of the yellow storage box used for long-term storage of the vial. The vial must be placed in the box allotted for its aliquot type and contact occasion in an upright position and stored at -80°C. The label on the box should have the aliquot code (i.e., LT1, LT2, LT3, etc.), contact occasion, beginning and end dates, and box number for the aliquot code.

Note: Items 9-11 and 23-25 do not have a part "b".

Following is an item-by-item description of each aliquot:

Items 5-11 refer to vials from the first lavender top tube.

5. LT1 (tHcy), i.e., lavender top vial #1, is used to measure total homocysteine (tHcy). 500  $\mu$ L is required for the sample aliquot.
6. LT2 (B12/folate), i.e., lavender top vial #2, contains specimen for measuring B12/folate. 1000  $\mu$ L is required for the sample aliquot.
7. LT3 (PLP), i.e., lavender top vial #3, contains specimen for measuring pyridoxal phosphate. 500  $\mu$ L is required for the sample aliquot.
8. LT4 (plasma archive 1), i.e., lavender top vial #4, contains the first plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.
9. LT5 (buffy coat), i.e., lavender top vial #5, contains specimen of white blood cells (buffy coat). Approximately 500  $\mu$ L is required for the sample aliquot.
10. LT6 (RBCs), i.e., lavender top vial #6, contains the first specimen of red blood cells for the archive. 1000  $\mu$ L is required for the sample aliquot.
11. LT7 (RBCs), i.e., lavender top vial #7, contains the second specimen of red blood cells for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 12-15 refer to vials from the second lavender top tube.

12. LT8 (chol/trig), i.e., lavender top vial #8, is used to measure cholesterol triglyceride. 500  $\mu$ L is required for the sample aliquot.
13. LT9 (HDL), i.e., lavender top vial #9, is used to measure high-density lipoprotein. 500  $\mu$ L is required for the sample aliquot.
14. LT10 (LDL), i.e., lavender top vial #10, is used to measure low-density lipoprotein. 500  $\mu$ L is required for the sample aliquot.
15. LT11 (plasma archive 2), i.e., lavender top vial #11, contains the second plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 16-20 refer to vials from the marble top tube.

16. MT1 (creatinine), i.e., marble top vial #1, contains specimen for measuring creatinine. 500  $\mu$ L is required for the sample aliquot.
17. MT2 (glucose), i.e., marble top vial # 2, contains specimen for measuring glucose. 500  $\mu$ L is required for the sample aliquot.

18. MT3 (fructosamine), i.e., marble top vial #3, contains specimen for measuring fructosamine. 500  $\mu$ L is required for the sample aliquot.
19. MT4 (serum archive 1), i.e., marble top vial #4, contains the first serum specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.
20. MT5 (serum archive 2), i.e., marble top vial #5, contains the second serum specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 21-22 refer to vials from the light blue top tube.

21. BT1 (BT archive 1), i.e., light blue top vial #1, is used to collect the first citrate plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.
22. BT2 (BT archive 2), i.e., light blue top vial #2, is used to collect the second citrate plasma specimen for the archive. The remainder is required for the sample aliquot.

Items 23-25 refer to urine void vials from the urine void container.

23. UR1 (creatinine), i.e., urine void vial #1, is used for measuring creatinine. 1500  $\mu$ L is required for the sample aliquot.
24. UR2 (microalbumin), i.e., urine void vial #2, is used for measuring microalbumin. 1500  $\mu$ L is required for the sample aliquot.
25. UR3 (archive), i.e., urine void vial #3, contains urine for the archive. 1500  $\mu$ L is required for the sample aliquot.

### **Blind Replicate Matching (BRM) Instructions**

Items 26 through 48 refer to Blind Replicate Matching (BRM) specimens used for quality control. These are specimens processed from extra tubes drawn from participants and labeled with a BRM ID assigned by the DCC. This extra tube must be the last tube drawn from the participant. Never perform a second stick to obtain a BRM; instead obtain the BRM tube from the next appropriate participant. They are stored and shipped along with regular specimens. See MOP chapter 11, for more information.

When it is time for a site to start a BRM collection for a randomization or follow-up visit, the DCC will email a blind replicate matching notification and an BRM ID sheet which contains the BRM ID to be used as well as the contact occasion for which samples are required. When you have been notified to do a blind replicate, a complete set of duplicate vials must be created and labeled using the BRM ID. Only one extra tube of blood may be drawn from a participant, therefore multiple participants will need to donate an extra tube each to complete the BRM. Each of the participants will have the items 26 through 48 entered in their

respective PHP forms; all will show the same BRM ID in item #27. For example, at the randomization or follow-up visit at least four participants will be used to complete the BRM, one will have data for the lavender top tube#1, one will have data for the marble top tube, one for the light blue top tube, and one will have data for the lavender top tube#2. Any one of the four participants can donate an extra urine sample or a fifth participant can donate the urine sample.

**The following is an example of a BRM request and collection scenario:**

- The clinic is sent an email containing a BRM notification request and a BRM ID sheet. The notification sheet and ID sheet contain the BRM ID (e.g., WU12343) that should be used for these BRM samples. The notification sheet indicates that a randomization and follow-up (05) BRM specimens are requested
- Randomization and contact occasion 05 Follow-up ID labels are mailed to clinic for BRM WU12343
- The coordinator sees on his/her schedule that Susan P., a randomization participant is due in today
- The coordinator takes one extra lavender top tube#1 of blood from Susan P. and puts the BRM label (WU12343) on the extra lavender top tube (note the extra tube must be the last tube drawn)
- The coordinator processes the extra BRM lavender top tube#1 along with the study participant's lavender top tube (aliquots it into 7 vials)
- Susan P.'s ID label (e.g., WU24680) is placed on the BRM ID sheet under lavender top tube #1 for the randomization visit to keep track of which participant donated which specimen
- The coordinator puts a check mark on the notification sheet under lavender top tube #1 for the randomization contact
- When Susan P.'s PHP is completed, the section on BRM (PHP items 26-48) is completed to reflect the processing of the BRM tube
  
- The next day John B., comes in for his randomization visit
- The coordinator takes one extra marble top tube from John B. (last tube drawn) and attaches the same BRM ID (WU12343) label
- The coordinator processes the extra BRM marble top tube along with the participant's marble top tube.
- John B.'s ID label (e.g., WU45670) is placed on the BRM ID sheet under marble top tube for the randomization visit
- The coordinator puts a check mark on the notification sheet under marble top tube for the randomization contact
- When John B.'s PHP is completed, the section on BRM (PHP items 26-48) is completed to reflect the processing of the BRM tube
  
- Later that day, Fred Z. comes in for his contact occasion 05 follow-up visit
- The coordinator takes one extra marble top tube from Fred Z. (last tube drawn) and puts the same BRM ID (WU12343) label on the extra marble top tube

- The coordinator processes the extra BRM marble top tube along with the participant's study marble top tube
- Fred Z.'s ID label (e.g., WU13579) is placed on the BRM ID sheet under marble top tube for the follow-up (05) visit
- The coordinator puts a check mark on the notification sheet under marble top tube for the follow-up (05) contact
- When Fred Z.'s PHP is entered, the section on BRM (PHP items 26-48) is completed to reflect the processing of the BRM tube
- The clinic continues to collect the rest of the BRM samples for the randomization visit which includes the lavender top tube#2, a light blue top tube, and a urine void. Likewise, the clinic continues to collect the rest of the BRM sample for the follow-up (05) visit which includes the lavender top tube #1, lavender top tube#2, a light blue top tube, and a urine void.

### **PHP ITEMS 26-48 BRM**

26. Record whether a BRM tube was collected and processed from the participant. If "No", data entry for the form is complete.
27. Record the BRM ID used to label the tubes and set of BRM aliquot vials being prepared.

### **BRM Aliquotting**

Items 28-48 record data on collection, hemolysis, and storage of BRM aliquots. Each item has three data entry parts: "a", "b", and "c" (except items 32-34 and 45-47 which do not have a part "b"). General instructions for these three parts are as follows:

- a. If the specimen was collected, record "Yes", otherwise record "No".
- b. If the specimen was hemolyzed, record "Yes", otherwise record "No".
- c. Using leading zeroes, record the three-digit number of the yellow storage box used for long-term storage of the vial. The vial must be placed in the box allotted for its aliquot type and contact occasion in an upright position and stored at -80°C. The label on the box should have the aliquot code (i.e., LT1, LT2, LT3, etc.), contact occasion, beginning and end dates, and box number for the aliquot code.

Following is an item-by-item description of each aliquot. Items 28-34 refer to BRM vials from the first lavender top BRM tube.

28. LT1 (tHcy), i.e., lavender top vial #1, is used to measure total homocysteine (tHcy). 500 µL is required for the sample aliquot.
29. LT2 (B12/folate), i.e., lavender top vial #2, contains specimen for measuring B12/folate. 1000 µL is required for the sample aliquot.

30. LT3 (PLP), i.e., lavender top vial #3, contains specimen for measuring pyridoxal phosphate. 500  $\mu$ L is required for the sample aliquot.
31. LT4 (plasma archive 1), i.e., lavender top vial #4, contains the first plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.
32. LT5 (buffy coat), i.e., lavender top vial #5, contains specimen of white blood cells (buffy coat). Approximately 500  $\mu$ L is required for the sample aliquot.
33. LT6 (RBCs), i.e., lavender top vial #6, contains the first specimen of red blood cells for the archive. 1000  $\mu$ L is required for the sample aliquot.
34. LT7 (RBCs), i.e., lavender top vial #7, contains the second specimen of red blood cells for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 35-38 refer to BRM vials from the second lavender top BRM tube.

35. LT8 (chol/trig), i.e., lavender top vial #8, is used to measure cholesterol triglyceride. 500  $\mu$ L is required for the sample aliquot.
36. LT9 (HDL), i.e., lavender top vial #9, is used to measure high-density lipoprotein. 500  $\mu$ L is required for the sample aliquot.
37. LT10 (LDL), i.e., lavender top vial #10, is used to measure low-density lipoprotein. 500  $\mu$ L is required for the sample aliquot.
38. LT11 (plasma archive 2), i.e., lavender top vial #11, contains the second plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 39-43 refer to BRM vials from the marble top BRM tube.

39. MT1 (creatinine), i.e., marble top vial #1, contains specimen for measuring creatinine. 500  $\mu$ L is required for the sample aliquot.
40. MT2 (glucose), i.e., marble top vial # 2, contains specimen for measuring glucose. 500  $\mu$ L is required for the sample aliquot.
41. MT3 (fructosamine), i.e., marble top vial #3, contains specimen for measuring fructosamine. 500  $\mu$ L is required for the sample aliquot.
42. MT4 (serum archive 1), i.e., marble top vial #4, contains the first serum specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.
43. MT5 (serum archive 2), i.e., marble top vial #5, contains the second serum specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 44-45 refer to BRM vials from the light blue top BRM tube.

44. BT1 (BT archive 1), i.e., light blue top vial #1, is used to collect the first citrate plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

45. BT2 (BT archive 2), i.e., light blue top vial #2, is used to collect the second citrate plasma specimen for the archive. The remainder is required for the sample aliquot.

Items 46-48 refer to BRM urine void vials from the BRM urine void container.

46. UR1 (creatinine), i.e., urine void vial #1, is used for measuring creatinine. 1500  $\mu$ L is required for the sample aliquot.

47. UR2 (microalbumin), i.e., urine void vial #2, is used for measuring microalbumin. 1500  $\mu$ L is required for the sample aliquot.

48. UR3 (archive), i.e., urine void vial #3, contains urine for the archive. 1500  $\mu$ L is required for the sample aliquot.



**Randomization/Follow-Up Phlebotomy Form: Processing and Inventory Instructions**  
**PHP Version A, 02/18/2002**  
**QxQ Date: 8/9/2002**

## **I. GENERAL INSTRUCTIONS**

The Randomization/Follow-up Phlebotomy Form: Processing and Inventory (PHP) is completed during the randomization clinic visit and at follow-up visits. It is used to document data during the phlebotomy process.

If specimen processing is done by a laboratory, they must be certified and familiar with and understand chapter 6: Specimen Collection and Processing, in the FAVORIT Manual of Operations (MOP). The Study Coordinator should insure an updated copy of this chapter is available in the laboratory as reference. The appropriate sections of the paper form must be completed by the Phlebotomist/Laboratory (processor) during specimen processing and then recorded in the data management system (DMS) by the Study Coordinator at the completion of the day's activities, and when shipping occurs, respectively. If processing is done by the Study Coordinator, s/he must be certified and familiar with and understand MOP chapter 6: Specimen Collection and Processing, and chapter 14: Administrative Procedures, prior to completing this form.

The form header information (ID, Contact Occasion, Sequence Number, Name and Initials) is completed as described in the MOP. Data related to storage and shipping must be appropriately recorded by the Study Coordinator. If a laboratory is processing the specimens, they should receive the form with **only the patient's ID Number, Contact Occasion and Sequence Number**. The Study Coordinator should complete the patient's Name and ID after the laboratory has completed processing.

Label vials with the provided pre-printed specimen ID labels. If a label is missing or unusable, prepare a hand-printed label using a fine-point cryomarker, writing on the "white area" of a blank FAVORIT specimen label. Record ID, Contact Occasion and tube type on the label. Verify that the correct ID is being used for the participant whose specimen is being drawn.

All specimens collected during the randomization and follow-up visits are for long-term storage (i.e., none will be shipped immediately to the Central Lab).

Two lavender, one marble, one light blue tube and one urine void will be collected.

To avoid elevated plasma homocysteine values and to ensure optimal retrieval of the buffy coat, sample should be centrifuged and processed within 45 minutes of collection. All processing **must** be completed within 4 hours of specimen collection.

### **Aliquotting Information:**

Upon completion of aliquotting, all vials should be tightly capped and frozen; remaining cells should be discarded. Samples should be placed **immediately** into the freezer.

Two 10mL lavender top tubes (LT label) with EDTA additive are collected. One tube is used to create seven vials LT1-LT7; the other is used to create four vials LT8-LT11. The 10mL marble top tube (MT label) for serum is used to create five vials MT1-MT5. The 4mL light blue top tube (BT label) for sodium citrate plasma is used to create two vials BT1 and BT2. The 5oz urine void container (UR label) is used to create three vials UR1-UR3.

## **II. SPECIFIC INSTRUCTIONS**

1. Time of centrifugation:
  - a. Record the time of centrifugation using a 12-hour clock format, filling in the fields using leading zeroes where necessary.
  - b. Record the time of day of centrifugation indicating AM or PM.
2. Placement of vials in the freezer:
  - a. Record the time vials were placed in the freezer using a 12-hour clock format, filling in the fields using leading zeroes where necessary.
  - b. Record the time of day that vials were placed in the freezer indicating AM or PM.
3. Record date of processing using the US order (month/day/year).
4. Record processor's three initials. If s/he only has two initials, then record the 1<sup>st</sup> name initial in the first box, the last name initial in the 2<sup>nd</sup> box and leave the third box blank.

Items 5-25 record data on collection, hemolysis, and storage of aliquots. Each item has three data entry parts: "a", "b", and "c". General instructions for these three parts are as follows:

- a. If the specimen was collected, record "Yes", otherwise record "No".
- b. If the specimen was hemolyzed, record "Yes", otherwise record "No".
- c. Using leading zeroes, record the three-digit number of the yellow storage box used for long-term storage of the vial. The vial must be placed in the box allotted for its aliquot type and contact occasion in an upright position and stored at -80°C. The label on the box should have the aliquot code (i.e., LT1, LT2, LT3, etc.), contact occasion, beginning and end dates, and box number for the aliquot code.

Note: Items 9-11 and 23-25 do not have a part "b".

Following is an item-by-item description of each aliquot:

Items 5-11 refer to vials from the first lavender top tube.

5. LT1 (tHcy), i.e., lavender top vial #1, is used to measure total homocysteine (tHcy). 500  $\mu$ L is required for the sample aliquot.
6. LT2 (B12/folate), i.e., lavender top vial #2, contains specimen for measuring B12/folate. 1000  $\mu$ L is required for the sample aliquot.
7. LT3 (PLP), i.e., lavender top vial #3, contains specimen for measuring pyridoxal phosphate. 500  $\mu$ L is required for the sample aliquot.
8. LT4 (plasma archive 1), i.e., lavender top vial #4, contains the first plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.
9. LT5 (buffy coat), i.e., lavender top vial #5, contains specimen of white blood cells (buffy coat). Approximately 500  $\mu$ L is required for the sample aliquot.
10. LT6 (RBCs), i.e., lavender top vial #6, contains the first specimen of red blood cells for the archive. 1000  $\mu$ L is required for the sample aliquot.
11. LT7 (RBCs), i.e., lavender top vial #7, contains the second specimen of red blood cells for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 12-15 refer to vials from the second lavender top tube.

12. LT8 (chol/trig), i.e., lavender top vial #8, is used to measure cholesterol triglyceride. 500  $\mu$ L is required for the sample aliquot.
13. LT9 (HDL), i.e., lavender top vial #9, is used to measure high-density lipoprotein. 500  $\mu$ L is required for the sample aliquot.
14. LT10 (LDL), i.e., lavender top vial #10, is used to measure low-density lipoprotein. 500  $\mu$ L is required for the sample aliquot.
15. LT11 (plasma archive 2), i.e., lavender top vial #11, contains the second plasma specimen for the archive. 1000  $\mu$ L is required for the sample aliquot.

Items 16-20 refer to vials from the marble top tube.

16. MT1 (creatinine), i.e., marble top vial #1, contains specimen for measuring creatinine. 500  $\mu$ L is required for the sample aliquot.
17. MT2 (glucose), i.e., marble top vial # 2, contains specimen for measuring glucose. 500  $\mu$ L is required for the sample aliquot.

18. MT3 (fructosamine), i.e., marble top vial #3, contains specimen for measuring fructosamine. 500  $\mu\text{L}$  is required for the sample aliquot.
19. MT4 (serum archive 1), i.e., marble top vial #4, contains the first serum specimen for the archive. 1000  $\mu\text{L}$  is required for the sample aliquot.
20. MT5 (serum archive 2), i.e., marble top vial #5, contains the second serum specimen for the archive. 1000  $\mu\text{L}$  is required for the sample aliquot.

Items 21-22 refer to vials from the light blue top tubes.

21. BT1 (BT archive 1), i.e., light blue top vial #1, is used to collect the first citrate plasma specimen for the archive. 1000  $\mu\text{L}$  is required for the sample aliquot.
22. BT2 (BT archive 2), i.e., light blue top vial #2, is used to collect the second citrate plasma specimen for the archive. The remainder is required for the sample aliquot.

Items 23-25 refer to urine void vials from the urine void container.

23. UR1 (creatinine), i.e., urine void vial #1, is used for measuring creatinine. 1500  $\mu\text{L}$  is required for the sample aliquot.
24. UR2 (microalbumin), i.e., urine void vial #2, is used for measuring microalbumin. 1500  $\mu\text{L}$  is required for the sample aliquot.
25. UR3 (archive), i.e., urine void vial #3, contains urine for the archive. 1500  $\mu\text{L}$  is required for the sample aliquot.