

**NATIONAL FAMILY HEALTH SURVEY 2015-2016  
(NFHS-4)**

**CLINICAL ANTHROPOMETRIC BIOCHEMICAL  
(CAB) MANUAL**

**DECEMBER 2014**



(स्थापना / Established in 1956)  
बेहतर भविष्य के लिए क्षमता निर्माण  
Capacity Building for a Better Future

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## PREFACE

In combination with classroom instruction and practical experience, this manual will be used to teach you how to collect blood samples and conduct basic tests to measure biomarkers for the Fourth National Family Health Survey (NFHS-4). Before each training session, you should carefully study this manual and the biomarker section of the household questionnaire. You are encouraged to ask questions during training and to discuss problems encountered in order to avoid making mistakes during fieldwork. The training is organized into 4 phases:

- During the first phase, we will review with you the chapters of this manual. You will learn how to identify eligible respondents for biomarker measurement, how to record information relating to the biomarker being measured in the biomarker questionnaire or on special field forms, how to handle the technical procedures involved in height/length and weight measurement and, in blood sample collection, testing, and transportation, and other related instructions.
- In the second phase, you will practice the procedures you've been taught by role playing with other trainees. This practice will include height, weight and blood pressure measurements, finger pricks for haemoglobin, blood glucose testing and for the preparation of dried blood spots (DBS) for HIV testing in a central laboratory.
- In the third phase, you will visit a health facility for additional practice. After obtaining the consent from respondents or their guardian, you will practice taking height and weight measurements among children and adults, collect blood samples from eligible respondents and practice measuring biomarkers.
- In the final phase, known as field practice, you will be assigned to a NFHS-4 trainee team. During field practice, you will collect blood samples from eligible children and adults and measure biomarkers exactly as you will during the survey fieldwork. Households that you visit will be in clusters that are not part of the India NFHS-4 sample.

Throughout the training, you may be given homework assignments and tests. At the end of the training, your overall performance will be assessed and those who have performed the best will be selected to work in the survey. Your training does not end at the start of fieldwork. Rather, it is a continuous process. Your team supervisor and the NFHS-4 health and survey coordinators will play important roles in continuing your training and in ensuring the quality of data you collect throughout the survey. They will:

- Periodically observe your fieldwork activities to ensure that you are conducting yourself professionally, obtaining informed consent from respondents, and following the sample collection and biomarker measurement protocol correctly;
- Spot check that you 1) visited the correct households, and 2) collected blood samples and measured biomarkers only from eligible respondents;
- Collect blood specimens for transport to the laboratory and consolidate the field record forms;
- Regularly meet with you to discuss your performance and give out future work assignments.

Any field staff member who is not performing at the level necessary to produce the high quality data required to make NFHS-4 a success may be released from service.

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## CHAPTER 1: OVERVIEW OF SURVEY

### INTRODUCTION

The Fourth National Family Health Survey (NFHS-4) is a nationally representative household survey that measures a wide range of indicators relating to fertility, family planning, and maternal and child health, as well as knowledge, attitudes and behaviour around HIV/AIDS and the prevalence of HIV infection among Indian adults. This is the fourth NFHS conducted in India and will include for the first time blood glucose and hypertension measurements. NFHS-4 will produce population-based estimates of anaemia, HIV prevalence, blood glucose, blood pressure, and height and weight measurements among women age 15-49 and men age 15-54. Among young children age 6-71 months, anaemia, height and weight will be measured.

#### ***Anaemia Testing***

For anaemia testing, fingerstick blood samples be collected from all women age 15-49 living in all households and men age 15-54 in a sub-set of households, sampled for NFHS-4 who voluntarily consent to the testing. In addition, anaemia testing will also be performed for children under age six<sup>1</sup> found in the households. For the children, consent will have to be obtained from their parents or guardian. The anaemia testing will be conducted in the field using the HemoCue system (photometer and microcuvette), which requires a very small volume of blood to measure the level of haemoglobin. The results of the anaemia test will be recorded in the NFHS-4 Biomarker Questionnaire and provided immediately to each individual tested both verbally and in writing. The health investigator will describe to the respondent the meaning of the results and will advise the participant if medical treatment is necessary. Individuals whose haemoglobin levels fall below the designated cut-off points (9g/dl for men and pregnant women and 7g/dl for women who are not pregnant or don't know if they are pregnant) and children under age six will be referred to local health facilities for assessment and treatment (see Appendix 3). All households, whether eligible individuals found in the household participate in the anaemia testing or not, will be provided an informational brochure on anaemia (see Appendix 6 and 7). Brochures will also be made available to other community residents who request them.

#### ***HIV Testing***

HIV sero-prevalence in India is routinely estimated using sentinel surveillance among pregnant women attending selected health facilities. This methodology, however, has significant limitations because men are excluded and because pregnant women are not representative of all women due to biases in their distribution by age as well as fecundity. One of the objectives of NFHS-4 is to provide updated estimates of HIV sero-prevalence among adults in India. Consequently, a subsample of women age 15-49 and all men age 15-54 in the selected households will be requested to provide a finger stick blood sample for subsequent testing in a laboratory. Three to five pre-printed circles on Whatman 903 filter paper cards will be filled with blood obtained from a finger prick for subsequent

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<sup>1</sup>Children born in January 2010 or later and older than 6 months are eligible for anaemia testing; for the sake of simplicity, this group of children is referred to as "children under age six" throughout.

HIV testing in a laboratory. As part of the informed consent process, individuals who are eligible for the blood sample collection will be advised of the purposes for which the blood will be used and will be assured of the anonymity of the HIV test results. They will be told that it will not be possible to provide them with the results of the test, but if they are interested in determining their HIV status, they will be given a voucher for free HIV testing at one of the Integrated Counselling and Testing Centres (ICTC) nearest to their locale. They will also be given a referral about HIV/AIDS and counselling and testing (see Appendix 5). Parents or guardians of adolescents age 15-17 will be asked for permission to test the adolescent before assent of the adolescent is sought. Brochures will also be made available to other community residents who request them.

### ***Blood Glucose Testing***

Finger stick blood will also be collected from women age 15-49 and men age 15-54 for glucose testing using the same procedures as those used for anaemia testing. NFHS-4 will use the Free Style Optium H Glucometer to conduct blood glucose testing. The readings are considered equivalent to blood glucose levels in laboratory estimations using the GlucoseOxidase Method for glucose levels in the range of 10-600 mg/dL. The results will be available in five seconds on an LCD digital display and they will be given to respondents on a health card immediately after the test is completed. The health investigator will describe to the respondent the meaning of the results and will advise the respondent if a referral (see Appendix 4) to a medical centre is necessary.

### ***Blood Pressure Measurements***

Elevated blood pressure (high blood pressure) is a known risk factor for a number of chronic and non-communicable diseases. NFHS-4 will measure the blood pressure of eligible respondents using an OMRON Blood Pressure Monitor to determine the prevalence of hypertension. Blood pressure measurements for each respondent will be taken on three separate occasions and the readings recorded in the biomarker questionnaire with interval of 5 minutes between readings. The first result will be discarded and the average of the last two measurements will be calculated. The results will be given to respondents on a health card immediately after the test is completed. The health investigator will describe to the respondent the meaning of the results and will advise the respondent if a referral to a medical centre is necessary. Respondents whose systolic blood pressure (SBP) is >130 mm Hg or diastolic blood pressure (DBP) > 85 mm Hg are considered to have elevated blood pressure readings and will be encouraged to see their doctors as soon as possible for a full evaluation (see Appendix 2).

### ***Survey Design***

The NHFS-4 design calls for anonymous, linked HIV data analysis. The blood samples collected during fieldwork will be stored in the laboratory and will not be tested for HIV until the fieldwork is completed and the sample point and household numbers have been modified so as to ensure respondents' anonymity. NFHS-4 will provide all the basic HIV/AIDS indicators of interest both nationally and internationally. Data on HIV sero-prevalence will be useful in comparing with data from other sources such as HIV sentinel surveillance. The linked information will be invaluable in providing answers to many questions about the characteristics of HIV infected persons.

The fieldwork will be carried out by a number of interviewing teams. Field teams will consist of one field supervisor, three female interviewers, one male interviewer, and two health investigators. Female interviewers will interview women only and male interviewers will interview men only. Either female or male interviewers may conduct the household interview. Height and weight measurements of male and female respondents and children under the age of six years will be done by the health investigators on each team with assistance from interviewers, as needed. However, blood pressure measurement, blood collection for haemoglobin and blood glucose testing, and preparation of dried blood spots for HIV testing will be done only by the specially trained health investigators.

## OVERVIEW OF BIOMARKER COLLECTION

A biomarker may be thought of as a characteristic that can be independently measured and evaluated as an indicator of normal biologic processes, pathogenic processes, or pharmacologic response to a therapeutic intervention<sup>2</sup>. Biomarker measurements can serve as diagnostic tools to identify diseases in their early stages and can be used as surveillance tools to track changes in disease patterns or to evaluate intervention programs. In population-based surveys, biomarkers help assess the prevalence or occurrence of diseases or health conditions and can also be used at a macro level to measure the long-term effect of policies and programs. In NFHS, biomarkers are measured in order to report levels of specific diseases and conditions at the population level. Specific to NFHS-4, the following biomarkers will be measured: Haemoglobin, random blood glucose, blood pressure, height & weight, and HIV. This training manual will discuss the proper collection techniques and the appropriate recording and result reporting of these biomarkers.

Biomarker measurement or testing should take place after the completion of the household and individual questionnaires. However, prior to measurement or testing, certain tasks must be completed. This chapter reviews these tasks, which include:

- Determining eligibility
- Obtaining informed consent.

## ELIGIBILITY

Not all household members are eligible for biomarker measurements. Thus, the first step in the collection process is to identify members of the household who are eligible: women age 15-49, men age 15-54 in a subsample of households, and children under age 6 years old who are usual household residents or are visitors that have stayed in the house the night before the household interview took place. Based on age and residency, eligible respondents would qualify for one or more biomarker measurements or tests.

**It is the responsibility of the interviewer** to identify all of the household members who are eligible for biomarker measurements. Individuals eligible for biomarker measurements will be identified by Biomarker Summary menu in the CAPI programme.

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<sup>2</sup> Biomarker Definitions Working Group, National Institutes of Health, 2001

The table below summarizes which householdmembers are eligible for which measurements and tests

Groups eligible for biomarker measurements*		Weight	Height	Haemoglobintesting	Blood pressure	Blood glucose	DBS Collection for HIV testing
Children	0–5 months	√	√				
Children	6–71 months	√	√	√			
Women	15-49 years	√	√	√	√	√	
Women**	15-49 years	√	√	√	√	√	√
Men**	15-54 years	√	√	√	√	√	√

\*Must be usual residents or have slept in the household the night before the household interview took place.

\*\* Women and men in households selected for men’s interview.

When the interviewers have completed the household and individual interviews, they will fill in information for all adults and children in the household eligible for biomarker measurements from their CAPI device in the appropriate sections of the Biomarker Questionnaire.

In the Biomarker Questionnaire, the adult’s name, line number, age, and marital status must be accurately recorded. It is the responsibility of the interviewer to transfer all the above information from his/her CAPI device in to the Biomarker Questionnaire. The health investigator should check with the interviewer to make sure that the information entered in the Biomarker Questionnaire is correct and accurately recorded prior to collecting any biomarker-related information from the respondent. For children under age 6, the day and month of the child’s birth should be obtained directly from the mother or other knowledgeable adult.

**The following are important points to keep in mind when completing the Biomarker Questionnaire:**

1. **For all eligible adults who consent to biomarker testing and, who are also eligible for the individual interview, the biomarker data should only be collected *after* their individual interview has been completed.**
2. **Measure biomarkers from one individual at a time.** All biomarker testing should be completed on one individual at a time before moving on to the next eligible, consenting person. For example, if there is more than one eligible respondent in a household who has consented to biomarker testing, complete all the biomarker measurements on her/him *before* proceeding to the next respondent. Likewise, complete the testing of all biomarkers from one child before proceeding to the next child. Failure to do so will lead to results being recorded in the wrong columns of the biomarker questionnaire.

3. **Never alter any responses in the Biomarker Questionnaire without consulting the interviewer.** Even in cases where there are concerns about an individual's eligibility for testing, proceed with height and weight measurement and testing. Record in the comments section of the Biomarker Questionnaire a description of the problem. Provide as many details as possible. The field organization/central office will decide later what will be done about the test results for the respondent in question.

## INFORMED CONSENT

One of the most important tasks that must be done before collecting any biomarkers is for you to explain the purpose of the testing to eligible respondents, or in the case of children, to the parent or adult responsible for the child and, to obtain their consent before collecting any blood samples. In order to ensure that these individuals can make an "informed" decision about whether or not they want to be tested, the NFHS-4 Biomarker Questionnaire contains consent statements for each biomarker to be measured that must be read to the respondent-- if an adult, or in the case of a child, to the parent or adult responsible for the child, before you do the biomarker testing (see Appendix 1). For height/length and weight measurements among children or adults, you need to explain the procedures and ask for a verbal permission from the respondent or responsible adult to take the anthropometric measurements.

### **These consent statements include the following basic elements:**

- a description of the objectives of the test
- basic information on how the test will be conducted
- assurances about the confidentiality of the results
- a specific request for permission to collect the sample

You must read the informed consent statements to each eligible respondent age 18 and over and obtain the respondent's consent before you can begin any testing or measurement. The approach for obtaining consent differs slightly when the eligible individual is a child under age 6 or an adolescent age 15-17. If the respondent is a child or adolescent, you must first obtain the consent of one of the respondent's parents, or in the absence of a parent, the consent of an adult who is at least 18 years of age and is responsible for the care of the child. For adolescents, you must also directly seek their assent. If the parent/responsible adult or the adolescent does not consent to the test, the test must not be performed. There are two exceptions to this rule of obtaining consent from a parent or responsible adult to test adolescents who are 15-17 years old: 1) if the adolescent is married or 2) if the adolescent lives alone or in a household in which there are no adults. In either instance, the adolescent is considered an emancipated minor, and is to be treated like an adult. Under these conditions, consent of the adolescent is sufficient.

Prior to performing the blood pressure measurement, haemoglobin test, the test for random blood glucose, or collecting blood samples for dried blood spot (DBS) preparation, you must record the outcome of the consent request in the applicable sections of the Biomarker Questionnaire. This is discussed in more detail within the upcoming chapters. You must also sign your name to indicate that you read the consent statement to the respondent, or in the case of children, to

the parent/responsible adult and have recorded their response accurately. Signing your name does not indicate that the respondent consented to be tested!

**Key points to remember include:**

1. **Read the applicable consent statements to each eligible respondent exactly as they appear in the questionnaire.** When you arrive at the household and begin talking about the blood tests and blood pressure measurements with the respondent, you may informally discuss many of the items included in the informed consent statement. However, before beginning the testing procedures or taking blood pressure readings, you must still read the informed consent statements exactly as they are worded in the biomarker questionnaire. If you feel that the respondent may find the statements repetitive, tell him/her that you are required to read the statement to ensure that respondents are given all the appropriate information.
2. **Read the informed consent statements clearly.** Practice reading the consent statements out loud so that you become comfortable delivering them in a clear, natural voice and manner. Avoid speaking rapidly or in a monotone. Look at the respondent as you read the statement.
3. **For adults and adolescents, always request consent for the different tests separately.** Be sure the respondent knows that it is possible to consent to one test and not to the other. Since the outcome of the consent process may differ for the haemoglobin and blood glucose test, the blood pressure measurement and dried blood spot sample (DBS) collection for HIV testing, it is important that you accurately record the results for individual tests at the appropriate places.
4. **Never collect blood from an adolescent before obtaining the consent of the parent or the adult responsible for the adolescent** unless the adolescent is married, was formerly married, lives alone, or lives in a household where there are no adult members.
5. **Never attempt to force or coerce consent.** Some respondents may be suspicious or fearful of having their blood collected for testing. Others may have questions or want to discuss the procedures before giving consent. Take time to patiently respond to all questions.
6. Some respondents may be reluctant to allow testing without consulting someone not present at the time of your visit (e.g., a woman may want to consult her husband before giving permission). **In such cases, make an appointment to return to the household later at an agreed upon time.** If you believe it will help, ask the team supervisor to visit a household where eligible respondents express fear or reluctance to be tested.

Once the individual interviews have been completed, eligible respondents have been identified, and consent has been obtained, biomarker measurement can take place.

## Summary of Steps in Identifying Eligible Respondents and Obtaining Consent

- Interviewer completes the household and individual questionnaires.
- Interviewer checks the list of individuals eligible for biomarkers to confirm individuals who are eligible for biomarker measurements:
  - Adults: women age 15 - 49, men age 15 – 54 in a subsample of households, who are usual residents or who stayed in the household the night before the interview, are eligible for biomarker measurement.
  - Children under age 6, who are usual residents or who stayed in the household the night before the interview, are eligible for biomarker measurement.
    - Children 6 -71 months: eligible for anthropometric measurements and anaemia testing
    - Children below 6 months: eligible for anthropometric measurements only
- For adults, age 18 and older: Obtain consent for biomarker measurement and testing as follows
  - Read consent exactly as written;
  - Record the outcome of the consent request and sign in the space provided;
  - If consent is granted, proceed with biomarker measurement.
- For never-married adolescents, age 15-17:
  - Obtain consent for biomarker measurement from parent or responsible adult
    - Record whether the parent/responsible adult consented or refused and sign in the space provided;
  - If the parent/responsible adult consented, obtain assent for biomarker measurement from the adolescent
    - Read consent exactly as written;
    - Record the outcome of the consent request and sign in the space provided;
    - If consent was granted, proceed with biomarker measurement and testing.
- For children 6 -71 months (for anaemia testing):
  - Obtain consent for biomarker measurement and testing from parent or responsible adult
    - Read consent exactly as written;
    - Record the outcome of the consent request and sign in the space provided;
    - If consent was granted, proceed with biomarker measurement and testing.
- For height/length and weight measurement, you must obtain a verbal permission from the respondent.

## CHAPTER 2: ANTHROPOMETRY

Anthropometry refers to the measurement of humans. In NFHS-4, anthropometry refers solely to the measurement of a person's height (length) and weight. This information can be used to assess the nutritional status of a population. For children, standard indices of physical growth related to nutritional status are height-for-age, weight-for-height, and weight-for-age. A child who is below minus two standard deviations (-2 SD) from the median of a reference population in terms of height-for-age is considered short for his/her age or stunted. Stunting reflects the cumulative effect of chronic malnutrition. A child who is below minus two standard deviations (-2 SD) from the median of a reference population in terms of weight-for-height is considered too thin for his/her height, or wasted. Wasting is a condition reflecting acute or recent nutritional deficit. Weight-for-age is a composite index of stunting and wasting and is a good indicator to monitor nutritional status over time.

Among adults, height and weight measurement are used to calculate a person's body mass index (BMI) and to assess a woman's risk of having difficulty in delivering due to her short stature (height <145 cm). BMI is calculated by dividing the weight in kilograms by the height in meters squared ( $\text{kg}/\text{m}^2$ ). BMI values are used to determine the percentage of the adult population that is normal, thin, overweight and obese.

### MATERIALS AND EQUIPMENT FOR ANTHROPOMETRY

- **SECA 874 U digital scale<sup>3</sup>**: for weighing children and adults. The scale has a maximum capacity of 200 kg and weighs in 0.01 kg increments. The scale is powered by six type AA 1.5 V batteries and has an "ON-OFF" switch located at the front side of the scale.
- **SECA 213 Stadiometer**: for measuring the height of adults.
- **SECA 417 Infantometer**: for measuring the length of children under 2 years or less than 85 cm.
- **Biomarker Questionnaire**

### PROCEDURES AND PRECAUTIONS BEFORE MEASURING

1. **Layout of the Procedures**: Each step of the measurement procedures is directed at specific participants, who are named in bold letters at the beginning of each step: "**Measurer**" and "**Assistant**".
2. **Two Trained People Required**: Two trained people are required to measure a child's height or length. The measurer holds the child and takes the measurements. The assistant helps hold the child and records the measurements on the questionnaire. If there is an untrained assistant such as the mother, then the trained measurer should also record the measurements on the questionnaire. One person alone can take the weight of a child and record the results if an assistant is not available.
3. **Measuring Board and Scale Placement**: Be selective about where you place the measuring board and scale. It is best to measure outdoors during daylight hours. If it is cold, raining or if

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<sup>3</sup> The Seca 874 digital floor scale is manufactured by Seca Corporation, Munich, Germany. These instructions were adapted from instructions that accompany the Uniscale and revised by Irwin J. Shorr, MPH, MPS.

too many people congregate and interfere with the measurements, it may be more comfortable to weigh and measure indoors. Make sure there is adequate light.

4. **Age Assessment:** Before you measure, determine the child's age. If the child is less than two years, measure length (that is, with the child lying). If the child is two years of age or older, measure height (that is, with the child standing). If accurate age is not possible to obtain, measure length if the child is less than 85 cm. Measure height if the child is equal to or greater than 85 cm.
5. **Weigh and Measure One Child at a Time:** If there is more than one eligible child in a household, complete the weighing and measuring of one child at a time. Then proceed with the next eligible child. DO NOT weigh and measure all the children together. If there is more than one eligible woman in a household, weigh and measure her and all her eligible children before proceeding with the next woman, otherwise measurements may get recorded in the wrong columns of the questionnaire. Return measuring equipment to the storage bags immediately after you complete the measurements for each household.
6. **Control the Child:** When you weigh and measure, you must control the child. The strength and mobility of even very young children should not be underestimated. Be firm yet gentle with children. Your own sense of calm and self-confidence will be felt by the parent and the child.

When a child has contact with any measuring equipment, i.e., on an infantometer, you must hold and control the child so the child will not trip or fall. Never leave a child alone with a piece of equipment.

7. **Coping with stress:** Since weighing and measuring requires touching and handling children, normal stress levels for this type of survey work are higher than for surveys where only verbal information is collected.

Explain the weighing and measuring procedures to the mother, father, or other responsible adult and to a limited extent, the child, to help minimize possible resistance, fears or discomfort they may feel. You must determine if the child or the parent is under so much stress that the weighing and measuring must stop. Remember, young children are often uncooperative; they tend to cry, scream, kick and sometimes bite. If a child is under severe stress and is crying excessively, try to calm the child or return the child to the parent before proceeding with the measuring.

**Do not weigh or measure a child if:**

- The parent/responsible adult refuses,
- The child is too sick or distressed,
- The child is physically deformed which will interfere with or give an incorrect measurement. To be kind, you may want to measure such a child and make a note of the deformity on the questionnaire.

8. **Recording Measurements and Being Careful:** Keep objects out of your hands and pens out of your mouth, hair or breast pocket when you weigh and measure so that neither the child nor you will get hurt due to carelessness. When you are not using a pen, place it in your equipment pack or on the questionnaire. Make sure you do not have long fingernails. Remove interfering rings and watches before you weigh and measure.
9. **Strive for Improvement:** You can be an expert measurer if you strive for improvement and follow every step of every procedure the same way every time. The quality and speed of your measurements will improve with practice. You will be required to measure women, men, and children. **Do not take these procedures for granted even though they may seem simple and repetitious. It is easy to make errors when you are not careful.** Do not omit any steps. Concentrate on what you are doing.

#### STEPS FOR MEASURING WEIGHT AND HEIGHT/LENGTH:

1. Before the health investigator (HI) starts any biomarker measurements, the interviewer should carefully check that all adults and children in the household who are eligible for weight and height/length measurement have been recorded on the paper Biomarker Questionnaire. Children born in 2010 or later who are usual residents of the household or who are visitors who spent the previous night in the household are eligible for anthropometry. This information should be entered in the appropriate columns of the Biomarker Questionnaire by the interviewer.
2. In **Question 203**, ask for the day, month and year of birth of the child from the mother or other knowledgeable adult.
3. In **Question 204** confirm that the child was born in **JANUARY 2010** or later. If the child was born before **JANUARY 2010**, go to **Question 203** for the next child.
4. Perform the child weight and height/length measurements according to instructions below.
5. Check that the **Name, Line Number, Age, and Marital Status** all eligible women and men has been recorded in **Question 302/402**, respectively, of the Biomarker Questionnaire.
6. Perform the adult weight and height measurements according to the instructions below.

#### TAKING WEIGHT

##### **Preparing the Adult and Children to Take Their Weight**

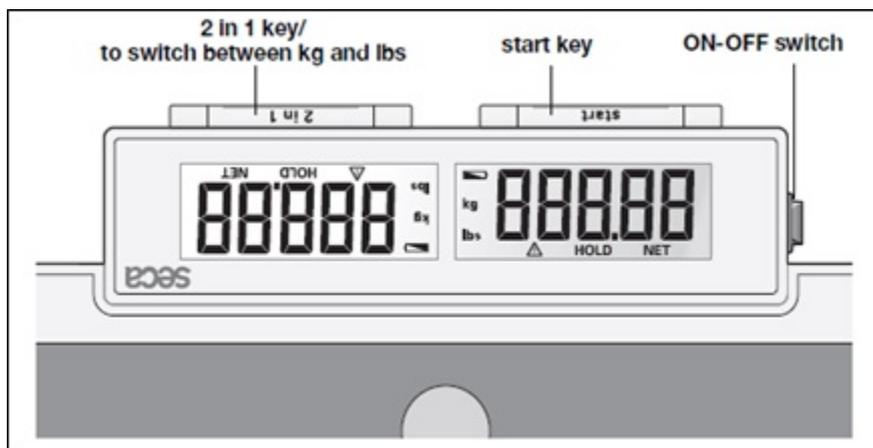
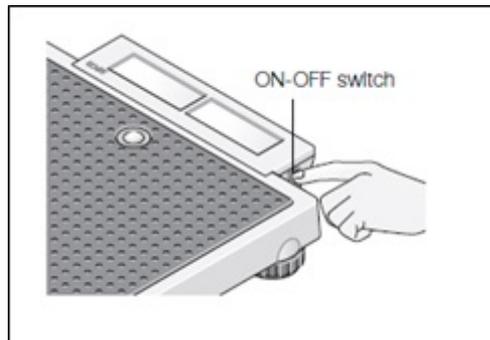
Show the scale to the adult and explain that you will weigh her/him and their children on the scale. Tell her/him that infants and any other children who will not stand on the scale alone can be held by the adult to obtain the child's weight. Ask the adult to wear light clothing while being weighed and to remove shoes/sandals and any heavy clothing, etc. Ask the adult to undress the child just before taking his/her weight. Leave underpants on the child.



Seca 874 model

### Preparing the Scale

When you are about to weigh children or adults in the household, take the scale out of the storage bag and place the scale on a hard, level surface. Uneven surfaces or vibration may cause the scale to malfunction. Turn on the power to the scale by pushing the switch located at the side of the display window of the scale to position “ON”.



The scale will not function correctly if it is bumped, knocked or moved during the weighing. It is best to use the scale in the shade or indoors. Handle the scale carefully:

- Turn on the scale for weighing by pressing the “**START**” key.
- Do not drop or bump the scale.
- Do not weigh a total weight of more than 200 kg.
- Do not store the scale in direct sunlight or other hot places.

- Protect the scale against excess humidity or moisture.
- To clean the scale, wipe surfaces with a damp cloth and dry immediately.
- Never put the scale in water.
- After using the scale, turn off the scale by pressing the “**START**” key.
- The scale switches off on its own after a certain time:
  - After 3 minutes in Normal mode
  - After 2 minutes in the “**2 in 1**” mode

### **Weighing Adults and Children Who Can Stand on the Scale by Themselves**

1. If the power supply is not activated, push the power switch to position “ON”. The scale now has power. To prepare the scale for weighing, press the “**START**” key when no one is on the scale. The display should show “SECA, 8.8.8.8.8 and “0.00.” The scale automatically sets to zero and is now ready for use. Wait for the scale to display the numbers “0.00” before asking the adult or child to step on the scale.
2. Ask the adult or child to step onto the centre of the scale and stand still. Wait until the numbers on the display no longer change and stay fixed in the display.
3. “**HOLD**” and a triangle [Δ] with an exclamation mark appear in the display window and the weight remains frozen until the next weighing operation. Record the weight to 0.01 kg on the biomarker questionnaire.
  - For children, record the child’s **weight** measurement in **Questions 205**. If the child’s weight was not measured, record the appropriate code in **Question 205**.
  - Record an adult’s **weight** measurements in **Questions 303/403**. If the adult’s weight and was not measured, record the appropriate code in **Questions 303/403**.

### **Weighing Infants or Children Who Must be Held by an Adult While on the Scale**

*NOTE!!: If it is cold and the adult wants the child to be covered during the weighing, give her/him a blanket or cloth for covering the child **after** you have recorded the adult’s weight in the biomarker questionnaire, **however, you must carefully follow the instructions below.***

#### **If You Do NOT Give the Adult a Blanket or Cloth to Cover the Child:**

1. While the adult is on the scale, press (“**SHORT PRESS**”) the “**2 in 1 Function**” button. The scale stores the weight of the mother and the display returns to “**0.00**” and the “**NET**” appears in the window display.

**PLEASE NOTE!!!** In the above statement, only a “**SHORT PRESS**” on the “2 in 1 function” is required in order to weigh children who need to be held. A “**LONG PRESS**” on the “2 in 1 function” will switch the units of measurements from **KILOGRAMS to POUNDS** instead.

2. Give the child to the adult. The scale will determine the weight of the baby even though the adult is on the scale. Once the value for the child’s weight becomes stable for about 3 seconds, the value is retained and “**HOLD**”, [Δ] with an exclamation mark and “**NET**”

appear in the display window. The number in the display is the weight of the child only, even though the adult is also standing on the scale.

3. Record the weight of the child to 0.01 kg on the biomarker questionnaire.



4. If there are other children to be weighed who must be held by the adult, ask the adult to remain on the scale. It is very important that the same adult holds all the other children who are to be weighed. Take the previous child from her/him and repeat steps 2-4. The “**2 in 1**” remains on until it is pressed (“**SHORT PRESS**”) again (the total weight of all individuals weighed is displayed) or the scale switches of automatically

#### **If You Give the Adult a Blanket or Cloth to Cover the Child:**

1. Ask the adult to step off the scale after you have recorded her/his weight.
2. Wait for the scale to display the numbers “0.00” and is ready for use.
3. Give her/him the blanket or cloth and ask her/him to step back on the scale.
4. Press (“**SHORT PRESS**”) the “**2 in 1**” key.
5. The scale displays “**0.00**” and “**NET**”.
6. Give the child to the adult. When the value (weight) becomes stable, “**HOLD**”, [Δ] with an exclamation mark and “**NET**” are displayed in the display window.
7. Record the weight of the child to 0.01 kg on the biomarker questionnaire.

If there is another child to be weighed, ask the adult to remain on the scale. Take the previous child from her/him and repeat steps 6-7above.

#### **Additional Notes on the SECA scale:**

- The SECA scale switches off automatically 3 minutes after the last weighing in the “Normal Mode” or two minutes, if the “**2 in 1**” function is activated.
- The “**2 in 1**” key can be used to change the units of weight—from kilograms (Kg) to pounds (lb) or vice versa, if the key is held in for 3 seconds or longer. **DO NOT HOLD IN THE ‘2 IN 1’ KEY! PRESS THE KEY ONCE TO ACTIVATE THE FUNCTION FOR WEIGHING CHILDREN WHO MUST BE HELD BY AN ADULT.**
- **Check that you have not changed the units of weight from Kg to lb. NFHS does not record weights in lb.**
- Do not weigh loads with a total weight of more than 200 kg.

- Possible reasons for the scale not taring (returning to zero (“0.00”) after pressing the “2 in 1” key when the adult stands on the scale):
  - There was no weight on the scale to tare (i.e., the adult was not on the scale).
  - The “2 in 1” function was not activated.
  - The load weighs more than 200 kg; “STOP” appears in the display. Use a lighter load.

**What to do if the Scale Display Shows the Following Errors:**

No weight is displayed when there is a load on the scale?

- Check to see if the scale is switched on. Ask the adult to step off the scale and step gently on the weighing platform.
- Check to see if the power switch at the side of the scale is in the “ON” position
- Press the “START” key to prepare the scale for weighing
- Check the batteries

The scale keeps switching on, while being transported? The “START” key has been activated. Turn off the scale by pushing the “ON-OFF” switch to the “OFF” position.

The scale displays a weight after being transported or after new batteries have been put in? Press the “START” key; the scale will work normally again.

“0.00” does not appear before weighing? Start the scale again by pressing the “START” key. There should not be any load on the scale.

“----” appears instead of “0.00” before weighing? Start the scale again after it switches off automatically; there should not be any load on the scale.

One segment of the display is illuminated constantly or not at all? There is a problem with that segment of the scale. Inform your service dealer.

The display shows a battery with split shading? The battery voltage is running low. The batteries should be changed in a few days.

“..batt” appears in the display? The batteries are empty. Replace the batteries.

“STOP” appears in the display? The maximum load capacity of the scale has been exceeded.

The display flashes? Take the load off the scale and start again. Wait until 0.00 appears and weight again.

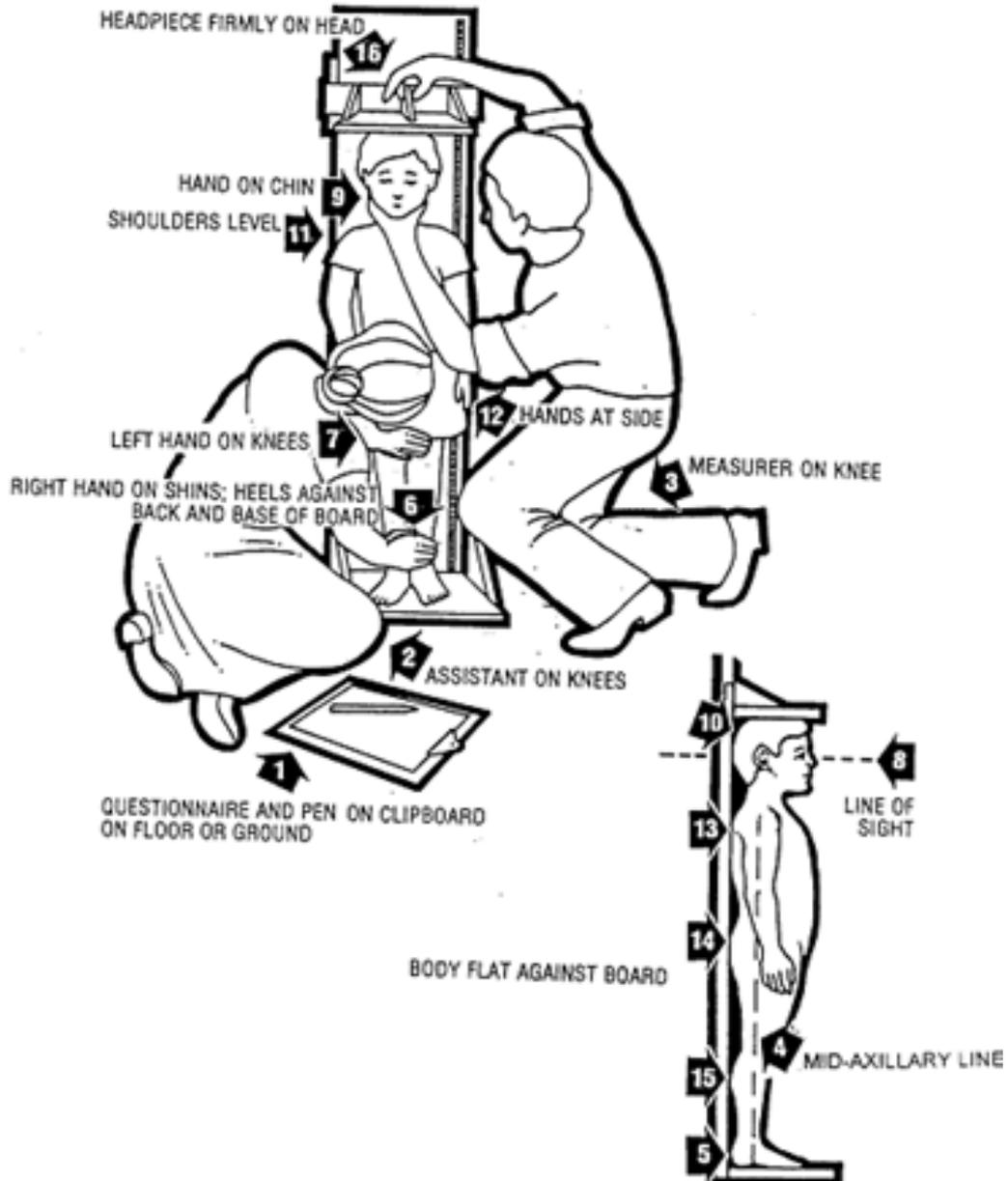
The display *Er* and a number appear in the display window? Start the scale again after it switches off automatically. The scale will then work normally again. If this does not happen, turn off the power to the scale. If the scale still does not work properly, inform the Service dealer.

## MEASURING A CHILD'S HEIGHT: STANDING UP (ILLUSTRATION 1):



1. **Measurer or Assistant:** Place the Seca stadiometer on a hard, flat surface against a wall, table, tree or staircase. Make sure the stadiometer is stable. Many walls and floors are not at perfect right angles; if necessary, place small rocks underneath the height board to stabilize it during the measurement.
2. **Measurer or Assistant:** Ask the parent to take off the child's shoes and to unbraided or push aside any hair that would interfere with the height measurement. Ask the parent to bring the child to the stadiometer and to kneel in front of the child so that the child will look forward at the parent.
3. **Assistant:** Place the biomarker questionnaire and pen on the ground (Arrow 1) and kneel on the right side of the child (Arrow 2).
4. **Measurer:** Kneel on the left of the child (Arrow 3).

Illustration 1  
Child Height Measurement



**Assistant:** Place the child's knees and feet in the correct position, with knees and feet either together or apart. There are three possible positions for the knees and feet:

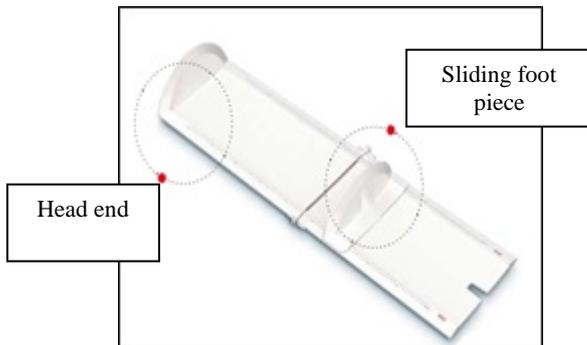
- Knees together and feet together
  - Knees together and feet apart
  - Knees apart and feet together
- } whichever touches first!

5. **Measurer:** Determine if the child's feet should be against or away from the back of the stadiometer by observing the imaginary line drawn from the tip of the shoulder to the heel, which is called the "mid-axillary line" (Arrow 4). This line should be perpendicular (i.e., 90°) to the base of the stadiometer where the child is standing

6. (You may have to move the child's feet away from the back of the stadiometer to put them in the proper position). Note that with most preschool-age children who are not heavy or obese, the heels will probably touch the back of the height board (Arrow 5).
7. **Assistant:** With your thumbs against the index finger of each hand, place your right hand on the child's shins (Arrow 6) and your left hand on the child's knees (Arrow 7). Do not wrap your hands around the knees or feet (ankles) or squeeze them together. Make sure the child's legs are straight.
8. **Measurer:** Ask the child to look straight ahead at the parent if she is kneeling in front of the child. Make sure the child's line of sight is parallel to the ground (Arrow 8). Place the thumb and index finger of your left hand, one finger on each side of the child's chin, and gradually close your hand (Arrow 9). Note that with most preschool-age children who are not heavy or obese, the back of the head will touch the back of the stadiometer (Arrow 10); however, if the child is heavy or obese, there will be a space between the back of the child's head and the back of the stadiometer. Make sure the child's shoulders are level (Arrow 11), the hands are at the child's side (Arrow 12), and at least the child's buttocks touch the back of the stadiometer. Note that with most preschool-age children who are not heavy or obese, the back of the head, the shoulder blades, the buttocks, the calves and heels will touch the back of the stadiometer (Arrows 10, 13, 14, 15 & 5).
9. **Measurer and Assistant:** Check the position of the child (Arrows 1-15). Repeat any steps as necessary.
10. **Measurer:** When the child's position is correct, lower the headpiece on top of the child's head (Arrow 16) making sure to push through the child's hair. Read and call out the measurement to the nearest 0.1 cm. Remove the headpiece from the child's head, your left hand from the child's chin, and allow the child to return to the parent.
11. **Assistant:** Immediately record the **height** measurements in **Questions 206** on the questionnaire and show it to the measurer. Record that the child was measured standing up in **Question 207**.
12. **Measurer:** Check the recorded measurement on the questionnaire for accuracy and legibility. Instruct the assistant to correct any errors.

## MEASURING A CHILD'S LENGTH: LYING DOWN (ILLUSTRATION 2):

For children less than two years old; or, when age cannot be obtained, length is measured for children less than 85 centimetres:



**SECA 417 Infantometer**

### **SECA 417 Infantometer**

**Measurement range:** 10–100 cm / 4-40"

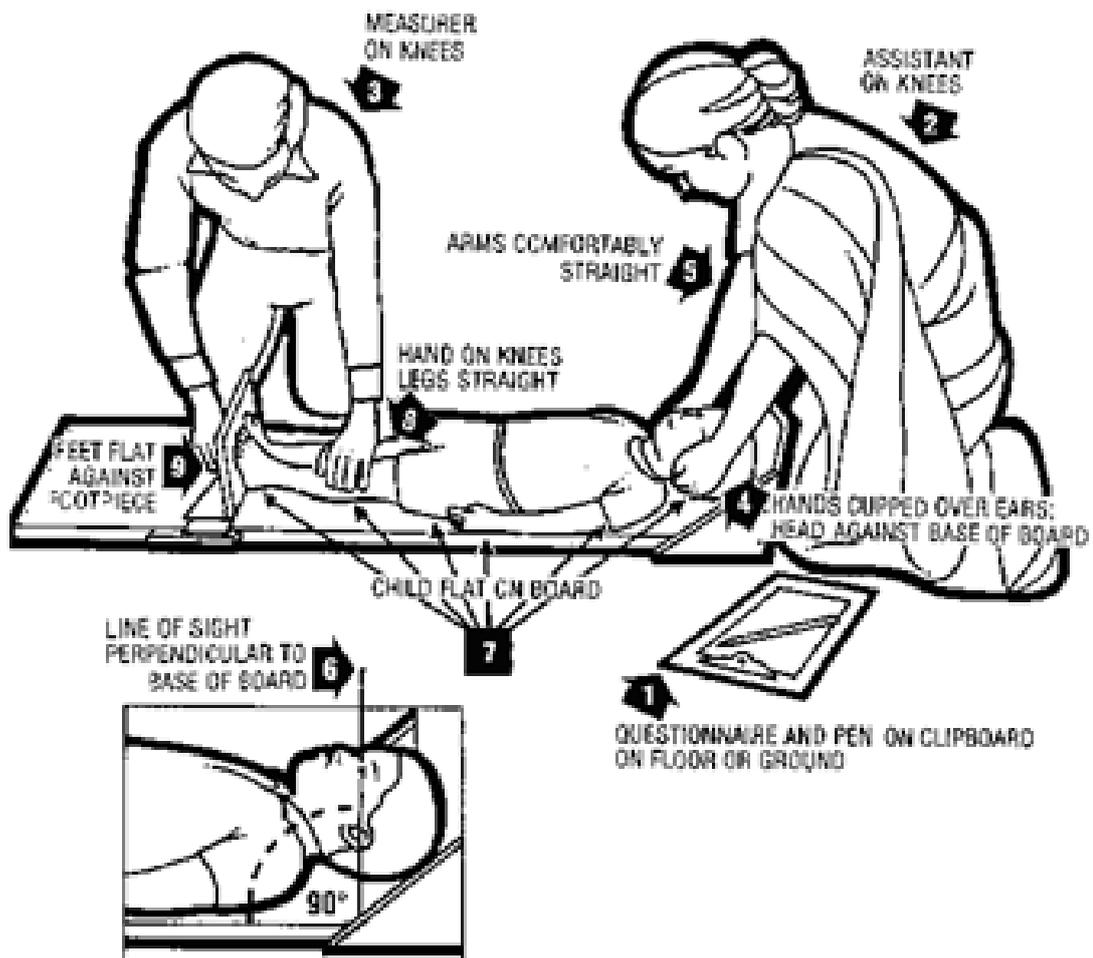
- Graduation: 2 mm / 1/8"
- Dimensions, measurement board (WxHxD): approx. 1.120 x 120 x 310 mm
- Dimensions, folded for transport (WxHxD): approx. 577 x 115 x 300 mm
- **Device weight:** approx. 1.4 kg

1. **Measurer or Assistant:** Place the Seca infantometer on a hard, flat surface, such as the ground, floor or a solid table. Make sure infantometer is stable.
2. **Assistant:** Place the biomarker questionnaire on the ground, floor or table (Arrow 1) and kneel behind the fixed head end of the infantometer if it is on the ground or floor (Arrow 2).
3. **Measurer:** Kneel at the right side of the child (at the child's feet) so that you can move the sliding foot piece with your right hand (Arrow 3).
4. **Measurer and Assistant:** With the help of the parent, gently lower the child onto the infantometer, making sure the measurer supports the child at the trunk of the body while the assistant supports the child's head.



5. **Assistant:** Cup your hands over the child's ears (Arrow 4). With your arms straight (Arrow 5), place the child's head against the base of the fixed head end. The child should be looking straight up (Arrow 6) so that the line of sight is perpendicular to the board. Your head should be directly over the child's head. Watch the child's head to make sure it is in the correct position against the base of the fixed head end of the infantometer.

Illustration 2  
Child Length Measurement



6. **Measurer:** Make sure the child is lying flat in the centre of the infantometer (Arrow 7).

Place the child's knees and feet in the correct position, with knees and feet either together or apart. There are three possible positions for the knees and feet:

- Knees together and feet together
  - Knees together and feet apart
  - Knees apart and feet together
- } whichever touches first!

With your thumb against your index finger, place your left hand on the child's knees (Arrow 8) and press them gently, but firmly against the board. Do not wrap your hand around the knees or squeeze them together. Make sure the child's legs are straight.

7. **Measurer:** Check the position of the child (Arrows 1-8). Repeat any steps as necessary.
8. **Measurer:** When the child's position is correct, move the sliding foot piece with your right hand until it is firmly against the child's heels (Arrow 9). Read the measurement to the nearest 0.1 cm and call out the measurement to the assistant. Return the child to the parent.
9. **Assistant:** Record the **height** measurements in **Questions 206** and that the child was measured lying down in **Question 207**. Show it to the assistant for confirmation.
10. **Measurer:** Check the recorded measurement on the questionnaire for accuracy and legibility. Instruct the assistant to correct any errors.

### MEASURING AN ADULT'S HEIGHT (ILLUSTRATION 3):

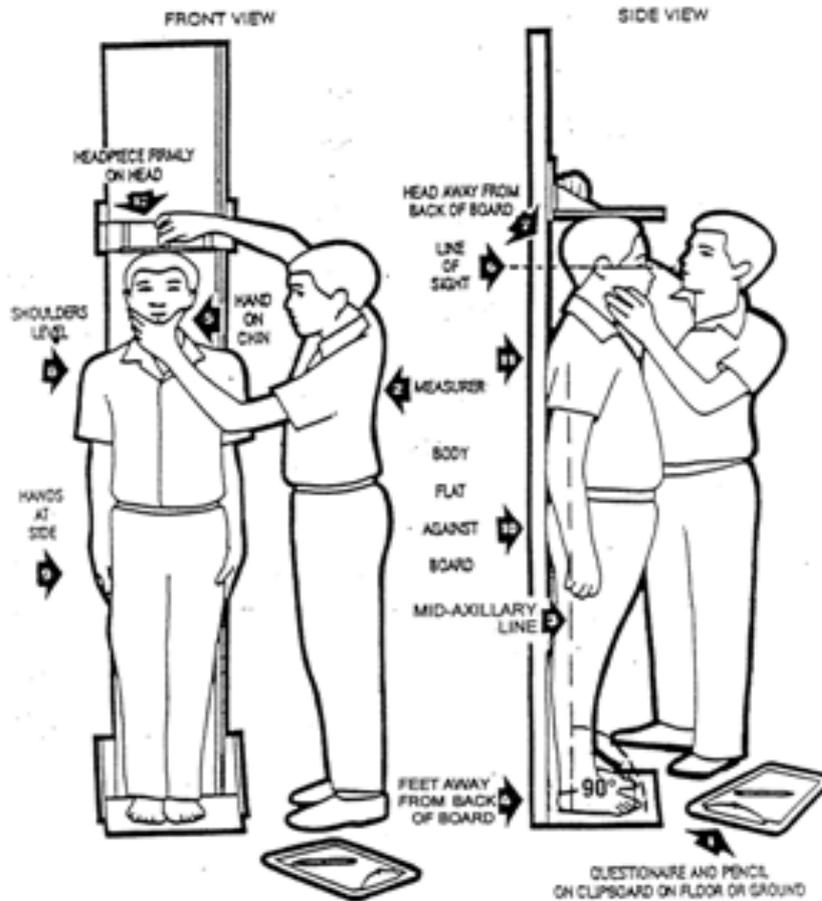
NOTE: The height of adults can be taken by one person alone, the Measurer.

1. **Measurer:** Place the stadiometer on a hard, flat surface against a wall, table, tree or staircase. Make sure the stadiometer is stable. Many walls and floors are not at perfect right angles; if necessary, place small rocks underneath the height board to stabilize it during the measurement.
2. **Measurer:** Ask the person to take off his/her shoes and ask him/her to unbraided or push aside any hair that would interfere with the height measurement. Ask the person to stand on the base of the height stadiometer and to face forward.
3. **Measurer:** Place the biomarker questionnaire and pen on the ground (Arrow 1) and stand on the left side of the person (Arrow 2).
4. **Measurer:** Determine if the person's feet should be against or away from the back of the height board by observing the imaginary line drawn from the tip of the shoulder to the heel, which is called the "mid-axillary line" (Arrow 3). This line should be perpendicular (i.e., 90°) to the base of the stadiometer where the person is standing. Note that with almost all adults you will have to move the person's feet away from the back of the height board to put them in the proper position; Arrow 4.
5. **Measurer:** Place the knees and feet in the correct position, with knees and feet either together or apart. There are three possible positions for the knees and feet:
  - Knees together and feet together
  - Knees together and feet apart
  - Knees apart and feet together

} Whichever touches first!

Illustration 3

## Standing Height of Adults\*



\* From: "Anthropometry as Part of Household Surveys", I.J. Shorr, The World Bank, Washington, D.C., (in press)

6. **Measurer:** Ask the person to look straight ahead. Cup the respondent's chin between the thumb and index finger of your left hand and gradually close your hand (Arrow 5). Position the person's head so that the line of sight is parallel to the ground (Arrow 6). Note that with most adults, the back of the head will not touch the back of the stadiometer—there will be a space between the back of the person's head and the back of the stadiometer (Arrow 7). After you have placed the person's head in the proper position, release your hand from the person's chin and ask him/her to hold his/her head in the position you have just placed it in.

Make sure the person's shoulders are level (Arrow 8), the hands are at the person's side (Arrow 9), and at least the buttocks touches the back of the stadiometer). Note that with most adults, only the buttocks and perhaps the shoulder blades, will touch the back of the stadiometer (Arrows 10 & 11).

7. **Measurer:** Check the position of the person (Arrows 1-11). Repeat any steps as necessary.
8. **Measurer:** When the person's position is correct, lower the headpiece on top of the head (Arrow 12) Making sure to push through the person's hair. **Read and call out the measurement to the nearest 0.1 cm.** Remove the headpiece from the person's head, and escort the person off the height board.  
**Measurer:** Immediately record the measurement on the questionnaire. Record an adult's **height** measurements in **Questions 304/404**. If the adult's height was not measured, record the appropriate code in **Questions 304/404**.
9. **Measurer:** Check the recorded measurement on the questionnaire for accuracy and legibility. Correct any errors.

### **Summary of Steps in Measuring Anthropometry**

- Children less than 2 years old or, in absence of age information, less than 85 cm
  - Ask the child's mother or other knowledgeable adult for the child's exact day, month, and year of birth
  - Confirm child was born in January 2010 or later
  - Measure weight
  - Measure length with child laying down on an infantometer
  - Record weight and height measurements in Q205 and Q206
  - Record that child was measured laying down in Q207
- Children 2 years or older or, in absence of age information, 85 cm or greater
  - Ask the child's mother or other knowledgeable adult for the child's exact day, month, and year of birth
  - Confirm child was born in January 2010 or later
  - Measure weight
  - Measure height standing up
  - Record weight and height measurements in Q205 and Q206
  - Record that child was measured while standing in Q207
- Adults
  - Verify the name with the respondent
  - Measure weight
  - Measure standing height
  - Record weight and height measurements in the appropriate sections

### CHAPTER 3: BLOOD PRESSURE MEASUREMENT

Elevated blood pressure (high blood pressure) is a known risk factor for death from stroke and coronary heart disease (CHD).

Arterial blood pressure is the force exerted by the blood on the wall of the artery as the heart pumps (contracts) and relaxes. Systolic Blood Pressure (SBP) is the degree of force when the heart is pumping (contracting) and Diastolic Blood Pressure (DBP) is the degree of force when the heart is relaxed. In this survey, you will measure systolic and diastolic blood pressure.

#### TECHNIQUE OF BLOOD PRESSURE MEASUREMENT

In NFHS-4, blood pressure measurements will be taken to assess the population prevalence of high blood pressure; the focus is not on the clinical diagnosis of high blood pressure. **Therefore, these measurements in a survey situation do not constitute a medical diagnosis of disease, but will be used only as a statistical description of the survey population.** The OMRON BP monitor will be used in NFHS-4.



**OMRON BLOOD PRESSURE MONITOR**

The person having his/her blood pressure measured will be referred to as the “respondent” and the person taking the measurements will be referred to as the “health investigator”. The Blood Pressure Measurement will be recorded in the blood pressure section of the **Biomarker Questionnaire**.

FIRST BP MEASURE	
SYSTOLIC .....	<input type="text"/> <input type="text"/> <input type="text"/>
DIASTOLIC .....	<input type="text"/> <input type="text"/> <input type="text"/>
REFUSED .....	994
TECHNICAL PROBLEMS ...	995
OTHER .....	996
(IF NOT MEASURED, GO TO 338) ←	

To measure the respondent's blood pressure, three blood pressure readings will be obtained. For simplicity, all blood pressure measurements will be made on the respondent's left arm. Where this is not feasible, the right arm will be utilized.

### **PRELIMINARY STEPS IN TAKING BLOOD PRESSURE MEASUREMENTS FROM ADULTS**

1. Before starting the blood pressure measurements, the respondent should have been sitting quietly for at least 5 minutes.
2. Be sure that the respondent does not smoke or drink coffee or tea during the measurements since smoking or drinking coffee or tea can affect the blood pressure. If the respondent consumed any alcohol, coffee or tea, or smoked cigarettes before the examination, record this in Qs. 312/412.
3. If the respondent indicates any reason why the blood pressure procedure should not be done on the left arm, use the right. If there is a problem with both arms, do not take the blood pressure and note this in Qs.316/416. Observe the respondent's arm while talking to him/her. If you observe any rashes, small gauze/adhesive dressings, casts, withered arms, puffiness, tubes, open sores, haematomas or wounds on both arms, do not take the blood pressure.

### **Positioning the Respondent for Blood Pressure Measurement**

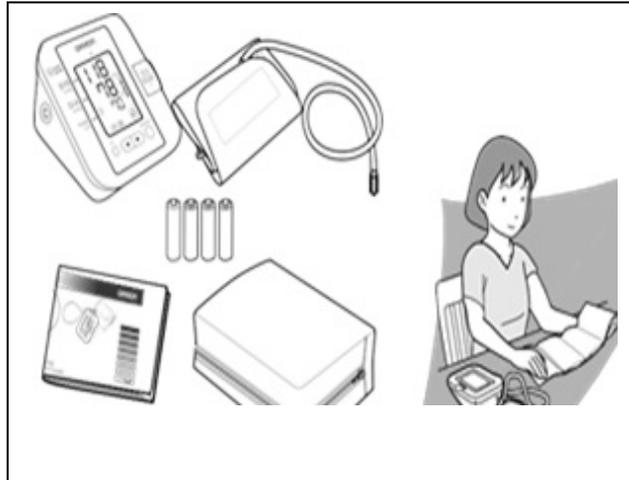
1. The respondent should be seated at a table in a relaxed, but not slouched, position with feet flat on the floor. The outer jacket or sweater should be removed and the sleeve should be rolled loosely up to the shoulder, ensuring that two fingers can be placed under the sleeve without difficulty. The respondent's left arm should be placed on the table, slightly flexed with palm upward.
2. The respondent's arm should be positioned so that it is resting on the table at heart level. Heart level is halfway between the shoulders and the waist. The respondent's elbow must be no lower than the lowest rib and must not be raised as high as the shoulder.
3. If the respondent is tall, it may be necessary to support the arm higher than a standard desk or table-top. Place the tall respondent's forearm on a pillow or a large book to raise the arm to heart level.

**For smaller or shorter adults, place a cushion or large book on the chair so that the arm is at heart level when the arm is resting on the desk or table-top. Place a box or large book under the respondent's feet if the feet do not rest flat on the floor.**

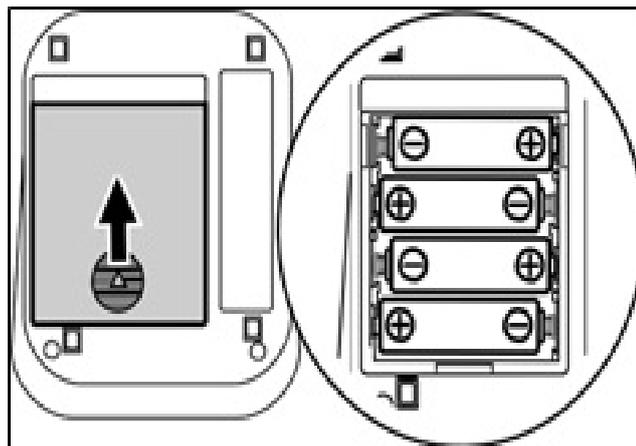
4. The health investigator should be seated facing and slightly to the left of the respondent, permitting easy access to the respondent's arm. The measuring equipment will be positioned so that the tube to the manometer is away from the respondent's body while the tube to the inflation bulb is closer to the body.

## BEFORE YOU START

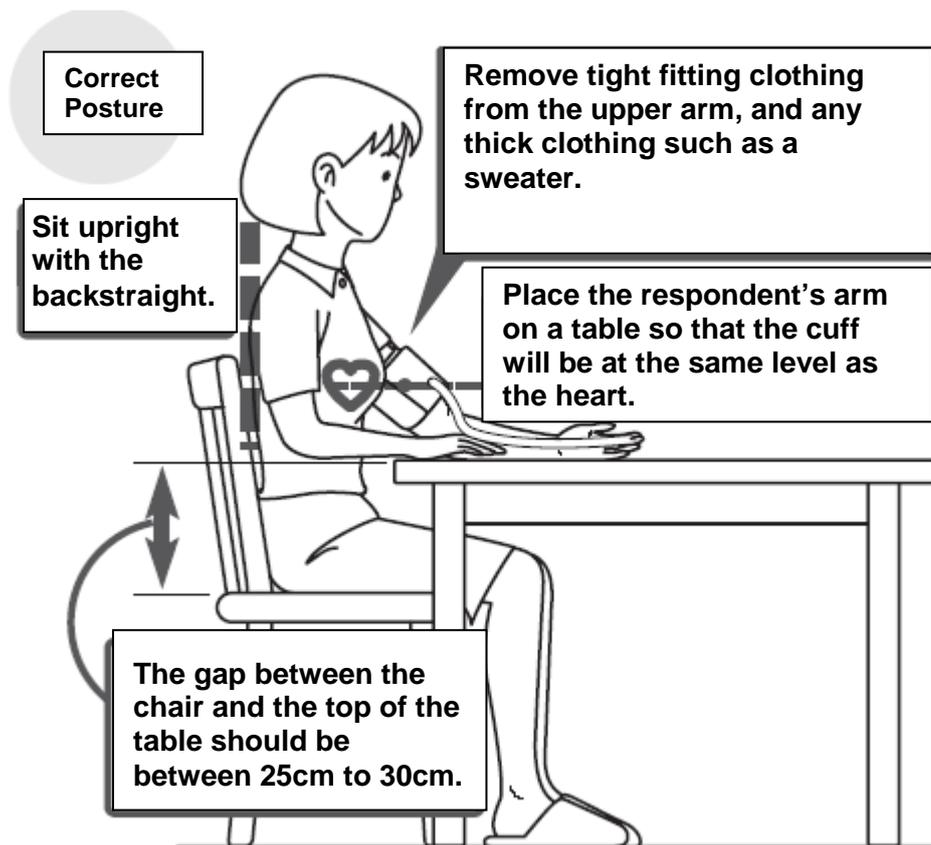
1. Make sure that all the components of the OMRON BP monitor are present.



2. Remove the battery cover and insert or replace 4 “AA” batteries as indicated in the battery compartment and replace the cover. If the battery is low, a symbol will appear on the display and requires that the batteries be replaced immediately.



## HOW TO SEAT THE RESPONDENT CORRECTLY WHEN TAKING A MEASUREMENT



### Notes:

- Measurements should be taken in a quiet place. The respondent should be in a relaxed, seated position.
- Make sure that the room is not too hot or cold.
- The respondent should avoid eating, smoking, or **exercising for at least 30 minutes before having a measurement taken.**
- The respondent should not move or talk during the measurement procedure.
- Do not place the cuff over thick clothes and do not roll up the sleeve if it is too tight.

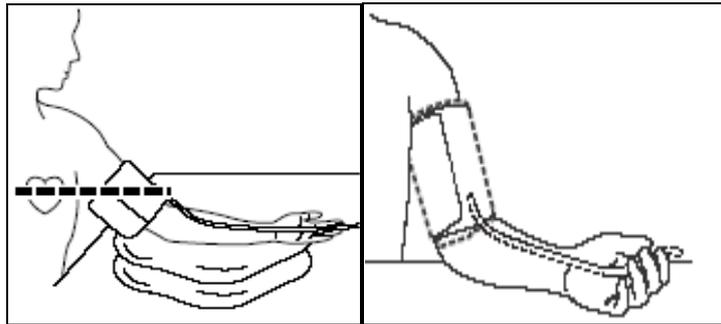
Correct posture during measurement is necessary to get accurate results. Examples of incorrect posture:

- Arched back (leaning forwards)
- Sitting cross-legged
- Seating the respondent on a sofa or at a low table so that the respondents tend to lean forward.

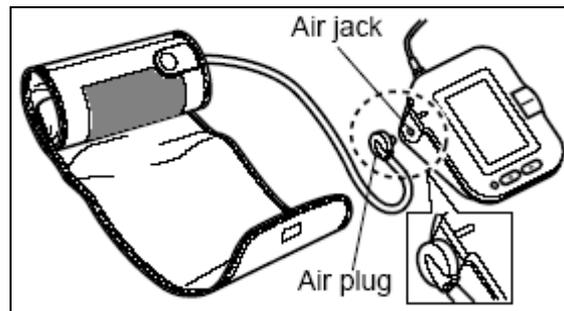


These situations could lead to higher blood pressure values due to strain or the arm cuff being lower than the heart.

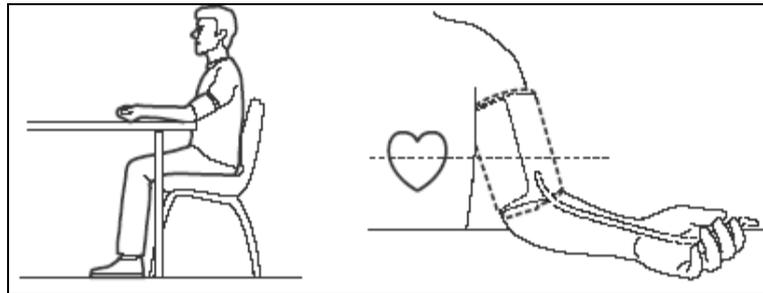
If the arm cuff is at a lower position than the respondent's heart, use cushions or pillow to adjust the height of the respondent's arm.



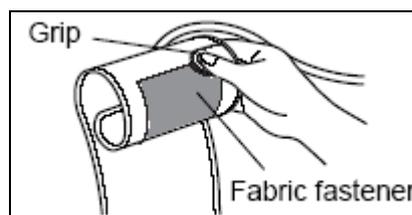
1. Insert the air plug into the air jack (on the left side of the device). The cuff must be fully deflated when it is inserted into the air jack.



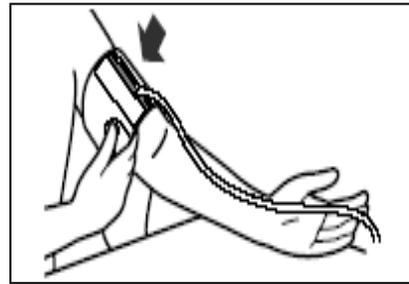
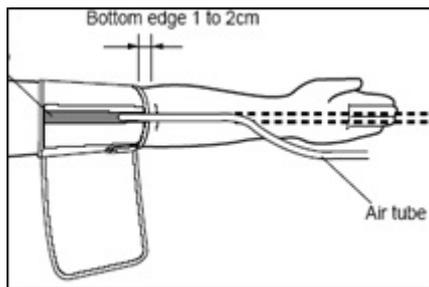
2. Have the respondent sit in a chair with her/his feet flat on the floor and place her/his arm on a table so that the cuff will be at the same level as her/his heart.



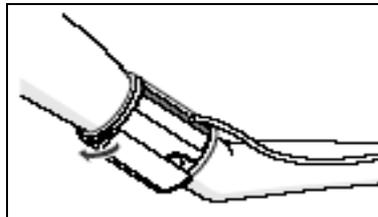
3. Hold the grip on the cuff securely with your hand.



- Turn the palm of the respondent's hand upward.

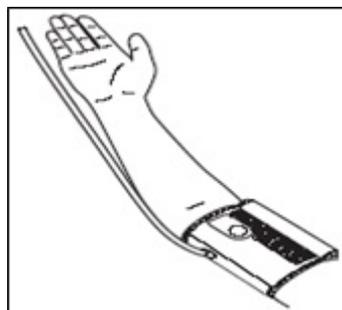


- Apply the cuff to the respondent's upper arm so that the air tube is centred on the middle of the respondent's inner arm and points down the inside of the arm. The air tube should run down the inside of the respondent's forearm and be in line with her/his middle finger. The bottom of the cuff should be approximately 1 to 2 cm above the elbow.
- When the cuff is positioned correctly, close the fabric fastener firmly.



- Make certain the cuff fits snugly around the arm.
- The cuff should make good contact with the respondent's skin. You should be able to fit your index finger between the cuff and the respondent's arm easily, so you can pull the cuff off and on.
- Make sure that there are no kinks in the air tubing.

#### Measurements on the right arm



**Note the following points when applying the cuff to your right arm. Apply the cuff so that the air tube is at the side of the respondent's elbow.**

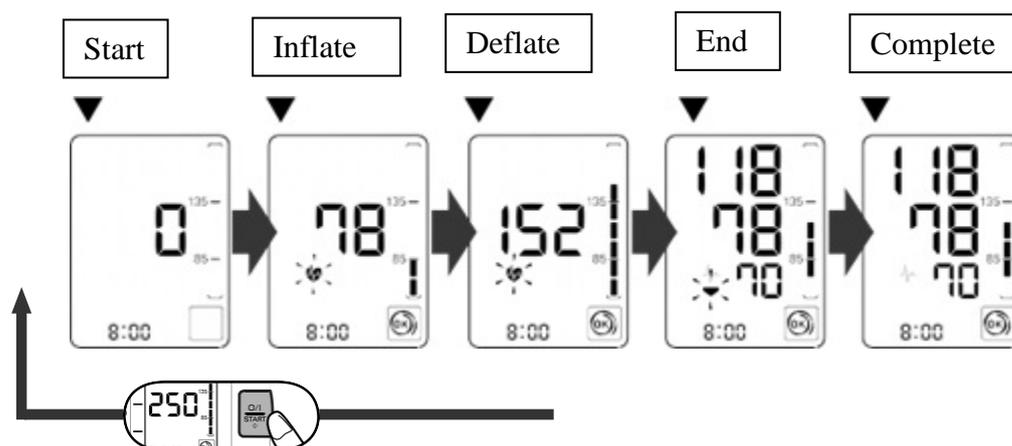
- Be careful not to rest your arm on the air tube, or otherwise restrict the flow of air to the cuff.
- Apply the arm cuff so that no part of the cuff is positioned over the elbow joint. The cuff should be 1 to 2 cm above the elbow.

## TAKING A BLOOD PRESSURE READING

For NFHS-4, the **OMRON BP Monitor** will be used for respondents with small, medium and large arm circumference. Measure the respondent's arm circumference to enable you select the correct cuff size for use. Press the **START** button and ask the respondent to remain still; the cuff will start to inflate automatically. As the cuff begins to inflate, the monitor automatically determines the ideal inflation level. The respondent should remain still and not talk until the measurement is completed.

**Note:** Do not inflate the cuff if it is not wrapped around the respondent's arm.

1. Inflation stops automatically and measurement is started. As the cuff slowly deflates, decreasing numbers appear on the display and the Heartbeat display  flashes at every heartbeat. In rare circumstances, the monitor might re-inflate the cuff to continue with the measurement.



2. When the measurement is complete, the arm cuff completely deflates and the blood pressure [and pulse rate] readings are displayed. Record the readings in the appropriate boxes.
3. Remove the arm cuff from the respondent's arm.
4. After the measurement is completed, you can either press the O/I start button to turn the monitor off or it will shut off automatically after 5 minutes.

### Safety and precaution:

- Do not use the instrument close to strong electromagnetic fields such as **mobile telephones or radio installations**.
- If the instrument is not going to be used for a prolonged period, the batteries should be removed.
- Do not open the instrument.

## WHAT DISPLAY SYMBOLS AND ERROR MESSAGES MEAN

Error Display	Cause	Remedy
	Irregular or weak pulses are detected.	Remove the arm cuff. Wait 2-3 minutes and then take another measurement. If this error continues to appear, contact your doctor.
	Movement during measurement.	Carefully repeat the steps measurement.
	Cuff is not applied correctly.	Apply the arm cuff correctly.
 Blink	The batteries are low.	You should replace them with new ones ahead of time.
 Lit	The batteries are exhausted.	You should replace them with new ones at once.

Error Display	Cause	Remedy
	Irregular or weak pulses are detected.	Remove the arm cuff. Wait 2-3 minutes and then take another measurement. If this error continues to appear, contact your doctor.
	Movement during measurement.	Carefully repeat the steps measurement.
	Cuff is not applied correctly.	Apply the arm cuff correctly.
 Blink	The batteries are low.	You should replace them with new ones ahead of time.
 Lit	The batteries are exhausted.	You should replace them with new ones at once.

Error Display	Cause	Remedy
EE	Cuff is under inflated.	Carefully read and repeat the steps listed under taking BP measurement
E	Movement during measurement.	Repeat measurement. Remain still and do not talk during measurement. Refer to section 3.3.
	Air plug disconnected.	Insert the air plug securely.
	Arm cuff not applied correctly.	Apply the arm cuff correctly.
	Clothing is interfering with the arm cuff.	Remove any clothing interfering with the arm cuff.
	Air is leaking from the arm cuff.	Replace cuff with new one.
EE	The arm cuff was inflated above 299 mmHg when inflating the cuff manually.	Do not inflate the arm cuff above 299 mmHg.
EE 25	Device error.	Contact your OMRON retail outlet or distributor.

**Note:**  
The irregular heartbeat symbol (  ) may also be displayed with error messages.

## Troubleshooting

Problem	C	Remedy
The reading is extremely low (or high).	Arm cuff not applied correctly.	Apply the arm cuff correctly.
	Movement or talking during measurement.	Remain still and do not talk during measurement.
	Clothing is interfering with the arm cuff.	Remove any clothing interfering with the arm cuff.
Arm cuff pressure does not rise.	The air tube is not securely connected into the main unit.	Make sure that the air tube is connected securely.
	Air is leaking from the arm cuff.	Replace the arm cuff with a new one.
Arm cuff deflates too soon.	The arm cuff is loose.	Apply the cuff correctly so that it is firmly wrapped around the arm.
Can not measure or readings are too low or too high.	The arm cuff has not been inflated sufficiently.	Inflate the cuff so that it is 30 to 40 mm Hg above your previous measurement result.

Nothing happens when you press the buttons.	The batteries are empty.	Replace the batteries with new ones.
	The batteries have been inserted incorrectly.	Insert the batteries with the correct (+/-) polarity.
Other problems.	<ul style="list-style-type: none"> <li>Press the O/I START button and repeat measurement.</li> <li>If the problem continues, try replacing the batteries with new ones.</li> </ul> If this still does not solve the problem, contact your OMRON retail outlet or distributor.	

## CHAPTER 4: GENERAL PROCEDURES FOR COLLECTING CAPILLARY BLOOD DROP SAMPLES FROM ADULTS AND CHILDREN

Capillary blood will be collected in NFHS-4 to test for the following biomarkers: haemoglobin (anaemia), random blood glucose (risk for diabetes), and HIV antibodies (prevalence of HIV infection). Capillary blood can be obtained from the palm side of the end of a finger. This chapter describes the materials needed for and, the steps involved in obtaining a capillary blood sample from adults and children.

### MATERIALS AND SUPPLIES FOR PERFORMING FINGERPRICK

The capillary blood drop(s) collected for biomarker testing will be drawn from a finger. The following supplies and materials will be used in performing the finger or heel prick:

- **Disposable gloves:** used to reduce the risk of bloodborne diseases. Gloves must be worn by the health investigators and by anyone else who may assist with the blood collection.
- **Absorbent paper sheets:** the surface area where your supplies will be placed while you conduct the blood collection. Be sure to place the plastic side of the absorbent sheet down/the absorbent side up.
- **Alcohol preps:** used for cleaning the skin prior to pricking the finger or heel.
- **Safety lancets:** the lancet is a single-use, disposable device used to prick the fingertip or heel (Figure 4.1). The needle is retractable: when the trigger is pressed, a surgical blade quickly protrudes from the device, punctures the skin, and then automatically retracts. The Unistik 3 Normal lancets have a gauge of 23G and a depth of 1.8mm. This lancet can be used for both children and adults
- **Preparation of lancet for use:**



Figure 4.1 Example of safety lancet



Hold the lancet by the sides, taking care **NOT** to press down on the release button. Just twist off the protective sterile cap full circle and lancet is ready to use.

- **Sterile gauze pads:** used to wipe away the first drop(s) of blood which helps to stimulate blood flow.
- **Adhesive bandages (plaster or Band-Aid):** used to cover the puncture site to minimize the risk of infection.

- **Plastic bag for waste:** Large bags that are provided to hold all of the biohazardous waste generated during the day. All waste bags are to be clearly labelled “biohazard”.

## STEPS IN OBTAINING CAPILLARY BLOOD FROM THE FINGER OF ADULTS FOR HAEMOGLOBIN, BLOOD GLUCOSE AND HIV TESTING

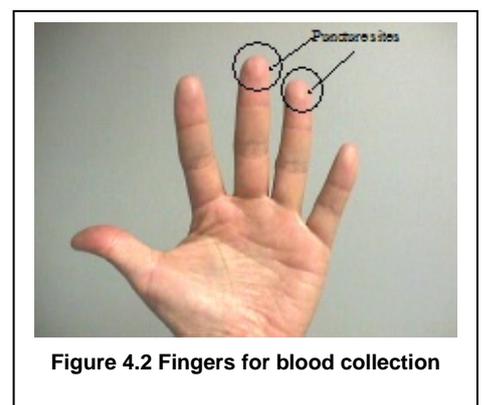
The following describes the steps that are involved in obtaining a capillary blood drop sample from the finger.

### 1. Complete general preparation

- If possible, find an indoor site to encourage privacy. If available, use a table or other piece of furniture with a flat surface to lay out your supplies. A couch, bed or mat should be readily available in the event the respondent feels faint and needs to lie down.
- If you find you must do the test outdoors, find a site in the full shade and away from rain, dust, and other environmental elements that might affect the sample.
- When and where possible, wash and dry your hands. **Always put on gloves** before beginning the collection of the blood sample from the first respondent.
- Take out a clean **absorbent paper sheet** and spread it over a flat surface where you will lay out your supplies.
- Refer to the **Biomarker Questionnaire** for adults to confirm the number of eligible individuals for whom blood samples will be collected. **After you have established the number of individuals to test, take out the appropriate equipment and general supplies for each respondent at a time.** You will want to have all general materials in easy reach when you begin collecting blood samples from the respondents.
  - **Note-** wait as long as possible before removing the microcuvettes, preferably right before pricking. These items should be taken out on an individual basis. In other words, take out one microcuvette to test a respondent. Do not remove multiple microcuvettes at the same time even if you will test more than one respondent. Take out a glucose strip from the container and insert in the glucometer following the manufacturer’s instruction.

### 2. Select and prepare the prick site

- Blood collection is usually easier if you **sit on the side of the respondent opposite to the hand** that you will collect blood from. For example, if you want to collect blood from the left hand, place yourself to the right side of the respondent.
- **Only use the third or fourth finger** for collecting the blood (Figure 4.2). Do not use a finger with a scar, a wound or cut, an infection, swelling, a deformity, or a



**Figure 4.2 Fingers for blood collection**

rash. Also, do not use a finger on which the respondent is wearing a ring, because the ring may disrupt the free flow of blood to the tip of the finger. You can ask the respondent to remove the ring.

- **Ask the respondent to warm her fingers by rubbing the palms together briskly until the skin becomes warm.** This will increase blood flow to the fingertip and improve the ease with which a sample can be obtained.



Figure 4.3 Clean the finger

- **Prepare the lancet for use**
  - Simply twist the blade slot cover 360° until the cover comes out.
  - **Do not remove the blade slot cover from the lancet other than as instructed above,** as this may cause the blade not to pierce the skin.
- **With an alcohol swab, clean the skin of the finger** thoroughly (Figure 4.3). If the swab is stained (with dirt), clean the finger a second time or until the swab is no longer stained. Allow the alcohol to air dry. Do not blow on the area to dry the alcohol. Blowing may deposit bacteria on the skin and contaminate the prick site.

#### Prick the Finger

- **Make sure that the finger is below the level of the respondent's heart** to increase the flow of blood to the finger. With your thumb, gently push the blood from the top knuckle toward the fingertip.

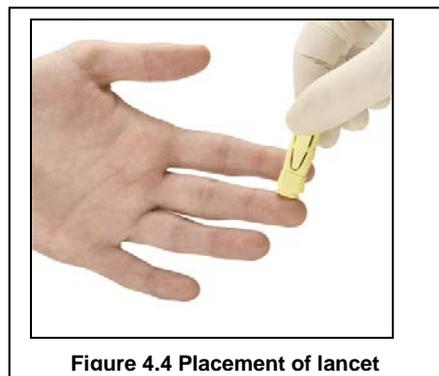


Figure 4.4 Placement of lancet

- When your thumb reaches the fingertip, **maintain a gentle pressure to trap the blood in the finger tip.**
- Place the lancet **firmly** against the skin with the **trigger facing upwards, so that the arrow preceding the trigger is visible** (Figure 4.4).

- **Note:** Avoid placing the lancet on the very tip of the finger or the sides beyond the palmar area or you will risk piercing the underlying bone. Proper puncture sites are shown in Figure 4.5.

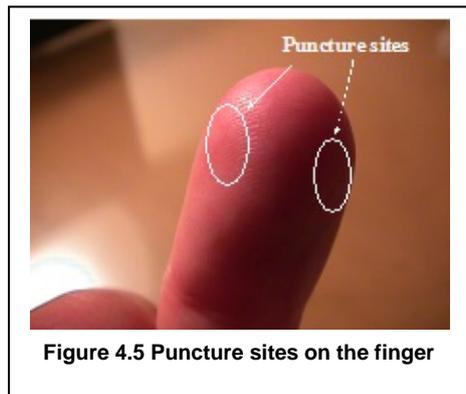


Figure 4.5 Puncture sites on the finger

- Use the lancet to **prick the skin** by placing the blade-slot surface against the area and pressing the trigger. The tip of the blade ejects through the blade slot, producing a micro-incision in the skin, and immediately retracts into the device. After pricking the skin, discard the lancet in the biohazard bag.

### 3. Collect the blood drops

- When the blood appears, use a sterile gauze pad to wipe away *the first one or two drops of blood depending on the tests being performed.*
- If the blood stops flowing before you have collected at least three blood drops on the filter paper card and/or done the anaemia testing, the pricking procedure may be repeated with the respondent's consent. For minors, you must get consent from the parent or adult responsible for the child. ***Do not reuse any of the supplies used for the first finger prick.***

### 4. After the blood collection, discard all used materials in the biohazardous waste bag.

## STEPS IN OBTAINING CAPILLARY BLOOD FROM CHILDREN 6-71 MONTHS OLD

Capillary blood will be collected in NFHS-4 from children 6-71 months old to test for **haemoglobin** only. Capillary blood can be obtained from the palm side of the end of a finger. If the blood is obtained from the finger, the steps in obtaining the blood sample are the same as those for an adult. However, if the child has very thin fingers that if used for the testing may result in injury to the underlying bone, the **heel** should be used instead. For children 6 – 12 months, a heel prick should be performed. The following describes the steps that are involved in obtaining a capillary blood drop sample from the heel.

The prick should be made outside a line drawn from the middle of the big toe to the heel or outside a line drawn from the area between the fourth and fifth toes to the heel (Figure 4.6). Take care to avoid the central area of the foot (to avoid injury to the nerves and tendons) or the centre of the heel (to avoid piercing the heel bone).



- **Prepare the lancet for use**
  - Simply twist the blade slot cover 360° until the cover comes out.
  - ***Do not remove the blade slot cover from the lancet other than as instructed above,*** as this may cause the blade not to pierce the skin.
- Hold the heel firmly (Figure 4.7). Apply moderate pressure near the puncture site. This can be done by wrapping the heel using your thumb and second finger.
- Clean the site with an alcohol swab. Make sure the site is dry before puncturing the skin with the lancet. In selecting a puncture site, avoid any areas of the skin that are broken or appear to be infected.

- Use the lancet for the skin puncture by placing the blade-slot surface against the area and pressing the trigger. Ensure the free flow of blood.
- Wipe away the first two drops of blood using a sterile gauze pad and collect the third drop for haemoglobin testing (see Chapter 5).
- After blood collection is complete, discard all materials used in the collection procedure in a labelled biohazardous waste container (bag).



Figure 4.7 Holding the infant's heel

#### PRECAUTIONS TO OBSERVE WHEN COLLECTING BLOOD SAMPLES<sup>4</sup>

This section describes the universal (general) precautions to be followed during blood collection<sup>5</sup>. You should take precautions when collecting blood to prevent exposure to blood borne infections, such as Hepatitis B or human immunodeficiency virus (HIV). When handling biological fluids, such as blood during the biomarker testing, the following rules should be followed to minimize your exposure to potentially biohazardous materials, thereby decreasing your chances of acquiring blood borne infections.

- **Wear gloves.** Gloves help to prevent skin and mucous-membrane exposure to blood. Gloves should be worn during blood collection, until the specimen(s) from a respondent is collected, and all waste materials produced during the collection are disposed. At that point, the gloves used should be treated as biohazardous waste. A new pair of gloves should be used with each respondent. **Gloves must never be reused!**

**Avoid penetrating injuries.** Although gloves can prevent blood contamination of intact and non-intact skin surfaces, they cannot prevent penetrating injuries caused by the instruments used for finger or heel pricks. Safety lancet devices reduce the risk of penetrating injuries.

- **Lancets** should not be used for purposes other than pricking the finger or heel. The lancets should not be broken or destroyed for curiosity or other purposes. Immediately after the testing is completed, the devices should be placed in the bag for biohazardous waste.
- **Wash** any skin surfaces or mucus membranes that become contaminated with blood immediately and thoroughly with running water or copious amounts of standing water.
- **Never eat or drink during the testing.** Since eating and drinking may result in you contaminating yourself, they are not permitted during haemoglobin and blood glucose

<sup>4</sup> Adapted from National Committee for Clinical Laboratory Standards (NCCLS) 1997

<sup>5</sup> For the universal precautions regarding bloodborne pathogens, see the U.S. Centers for Disease Control and Prevention guidelines and the U.S. Occupational Safety and Health Administration (OSHA) standards for protection from exposure to bloodborne pathogen.

measurements or collection of blood samples for HIV testing. And for similar reasons, you should not apply make-up during the biomarker testing.

- **Properly dispose of all biohazardous materials.** All materials coming in contact with blood must be placed in a biohazardous waste bag after use and disposed according to the survey's policy on infectious waste disposal. Take precautions when storing and transporting the waste during the fieldwork.

#### GOOD BLOOD COLLECTION PRACTICES

- **Good position in relation to the respondent.** Position yourself well before you puncture the respondent's finger.
- **Do not prick the finger if the hand is cold!** Warm the hands by asking the respondent to rub them together vigorously. In the case of a child, ask the mother to rub the child's hands.
- **Never "milk" the finger.** Excessive massaging or squeezing of the finger or heel will cause tissue juice to mix with and dilute the blood. This will result in erroneous test results, particularly yielding low levels of haemoglobin in blood. Instead, employ only mild pressure by using the thumb and the second and third fingers to make a "pad" at the puncture site.
- **Never allow alcohol to mix with the blood.** Alcohol, which is used to clean the puncture site, can mix with the blood and cause haemolysis of the sample leading to errors in the testing results. To avoid this problem, the finger or heel must be air dried completely before being punctured.
- **Avoid obstructing the blood flow to the finger tip.** It is important to hold the finger properly to allow for the accumulation of blood in the tip of the finger. Holding the finger too tightly can obstruct the blood flow to the finger, which will prevent you from getting an adequate volume of blood for testing.
- **Avoid shallow punctures.** A deep puncture should be made for better blood flow and to have a blood sample that is representative of that in the body.

#### Summary of Steps for Capillary Blood Collection

- For children between 6 and 12 months, a heel prick should be performed
- Set up blood collection supplies, making sure to select the correct lancet (adult or child)
- Clean the surface of finger/heel
- Prick the finger/heel
- Collect capillary blood
- Dispose of all used materials in the bag provided for biohazardous waste

## CHAPTER 5: HAEMOGLOBIN (ANAEMIA) TESTING

Red blood cells contain haemoglobin (Hb), an iron-rich protein that binds oxygen in the lungs and carries the oxygen to organs and tissues throughout the body. Anaemia is defined as a reduction in the normal number of red blood cells or a decrease in the concentration of Hb in the blood. Symptoms of anaemia range from pallor, fatigue and weakness, shortness of breath, and irregular heart rhythms. In NFHS-4, respondents will be screened for anaemia by measuring the level of Hb in their blood. Individuals who have an Hb level below a defined cut-off will be classified as anaemic.

Common causes of anaemia include:

- Iron deficiency from inadequate intake of foods containing iron, such as red meat;
- Intake of foods that contain non-bioavailable iron;
- Malaria and other parasitic infections (e.g., schistosomiasis; hookworm);
- Blood disorders (e.g. sickle cell anaemia; thalassaemia).

Anaemia is a common and significant global health problem. Consequences of anaemia include an increased risk of maternal and child mortality, impaired cognitive development in children, increased numbers of pre-term and low birth weight babies, and reduced work productivity in adults.

The measurement of Hb is the primary method of screening for anaemia. Hb measurement provides an opportunity to:

- Estimate the prevalence of anaemia in a nationally-representative sample;
- Link the levels of anaemia with demographic data so as to examine the socioeconomic, residential, and demographic differences in the prevalence of anaemia among populations;
- Design programs to prevent iron-deficiency anaemia among the populations most in need of intervention (e.g., iron fortification programs for women and young children living in rural districts).

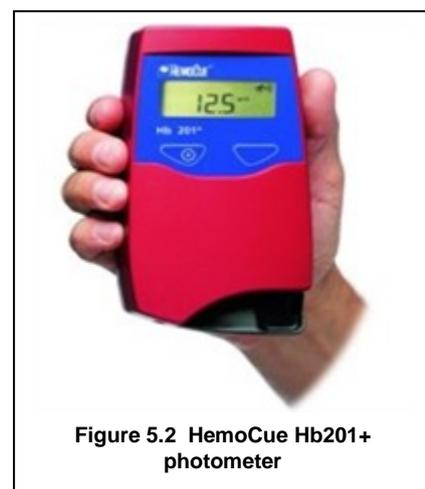
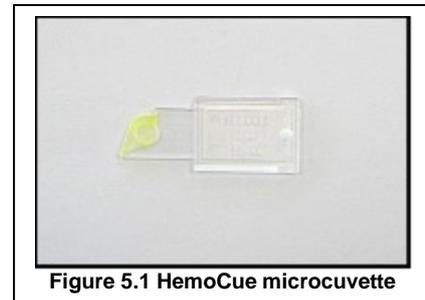
Haemoglobin measurement in NFHS-4 will be performed using the HemoCue photometer (Hb 201+). This widely used system measures the Hb concentration in a drop of blood obtained from by finger prick. The test is rapid, allowing results to be reported to the respondent immediately following the testing procedure. **Respondents found to have severe anaemia (an Hb level below 9 g/dl for pregnant women and below 7 g/dl for women who are not pregnant or don't know if they are pregnant and, men and children, will be referred to a health facility for further evaluation and treatment.**

This chapter discusses the materials needed and the procedure for haemoglobin measurement. Apart from discussing the materials required for haemoglobin measurement, directions are given regarding precautions to take during collection and testing, recording results in the **Biomarker Questionnaire**, and providing test results and information about anaemia to respondents.

## MATERIALS AND SUPPLIES FOR HAEMOGLOBIN MEASUREMENT

In addition to the supplies listed in Chapter 4, the following equipment and supplies are required for haemoglobin measurement:

- **Microcuvette:** a plastic disposable unit that serves as both a reagent vessel and a measuring device (Figure 5.1). The tip of the microcuvette contains a dry, yellow reagent (sodium azide). The microcuvette is designed to draw up the exact amount of blood needed for the test.
- **HemoCueHb 201 + photometer:** a device that uses the absorption of light to measure haemoglobin concentration from a single drop of blood collected in a microcuvette (Figure 5.2). Test results are presented on the photometer's electronic display. The HemoCue system is described in greater detail below.
- **Anaemia Brochure:** a one page document that provides basic information to the respondent about anaemia, including its definition, symptoms, causes, and methods of treatment and prevention. In addition, the respondents Hb results are recorded and classified within this document. See Appendix 7 or 8 for an example of an anaemia brochure.
- **Anaemia Referral Form for Severely Anaemic Respondents:** on this form is written the name and Hb results of severely anaemic respondents. It is to be given to individuals with Hb result indicative of severe anaemia. Respondents can take the referral form to a local, participating clinic or health centre, to receive proper treatment for their anaemia.



### THE HEMOCUE PHOTOMETER SYSTEM (Hb 201+)

Although the HemoCue system has proven to be durable and reliable under field conditions, there are some technical limitations to the system. One major limitation is that the reagent in the microcuvettes is sensitive to humidity. Thus, to minimize the degradation of the reagent, follow these instructions for the proper handling and storage of microcuvettes:

1. Record on the microcuvette container the date on which it is first opened;
2. Remove from the container only those microcuvettes required for immediate testing;
3. Remove the microcuvettes by holding the side opposite the tip;

4. Immediately after taking a microcuvette out of the container, snap the container lid back on tightly;
5. Keep the microcuvette container in a cool place and avoid exposing it to heat or direct sunlight.

Under these conditions, a microcuvette container can be stored for up to 3 months (90 days) after opening. Under field conditions, it is not advisable to store the microcuvettes in the opened container for more than a month. Microcuvettes from unopened containers can be used up to the expiration date on the container.

To ensure the HemoCueHb 201+ system operates properly, allow the photometer to come to the ambient temperature and protect it from direct sunlight. The device operates optimally between 18 and 30° C. The photometer has an internal electronic “SELFTEST”; every time the device is turned on, the device automatically verifies the performance of its optronic unit.

The photometer’s black microcuvette holder has three operating positions: 1) “pushed in” for measuring; 2) “pulled out” until ‘clicked’ for placing the microcuvette; 3) removed completely for cleaning.

Clean the microcuvette holder at the end of each day’s fieldwork. For cleaning, use an alcohol swab, or cotton wool/cotton-tipped swabs moistened with 70% alcohol. Check for dirt or dried blood on the swabs. Follow these procedures to clean the microcuvette holder (Figure 5.3):

1. Check that the analyser is turned off and the display window is blank.
2. Pull the microcuvette holder out of its loading position. Carefully press the small catch positioned in the upper right corner of the microcuvette holder.
3. While pressing the catch, carefully rotate the microcuvette holder towards the left as far as possible. Carefully pull the microcuvette holder away from the analyzer.
4. Clean the microcuvette holder with an alcohol swab or cotton wool moistened with 70% alcohol (ethanol or isopropyl alcohol).
5. Blood may get on the optronic system if you do not wipe the outside of the microcuvette before placing the microcuvette in the holder. If this happens, you will get an error message (E01-E05; E09-E30). Clean the optronic unit of HemoCue machine immediately when you get an error message before proceeding with measurements. Use a HemoCue cleaner to clean the optronic unit of the HemoCue device by pushing the swab into the opening of the microcuvette holder (Figure 5.3). Move the cleaner from side to side 5-10 times. If the swab of the cleaner is stained with blood or dirt, repeat the cleaning procedure with a new cleaner. **Do not use alcohol to clean the optronic unit.** It is important that the microcuvette holder is completely dry prior to



Figure 5.3 Cleaning microcuvette holder

reinserting it in the photometer. Furthermore, it is recommended to clean the optronic unit ones a week during fieldwork.

## COLLECTING BLOOD AND HAEMOGLOBIN MEASUREMENT

### **Children:** Follow the procedure below to measure haemoglobin level in children:

1. Determine if the child is 6 or more months old and eligible for haemoglobin measurement: For **Question 208**, refer to **Question 203** to determine if the child is less than 6 months old (i.e., the child was born in the month of interview or within the 6 months preceding the interview). If the child is less than 6 months old, the child is not eligible for haemoglobin measurement. Record '1' and continue to the next child. If the child is older than 6 months, proceed to **Question 209**.
2. Record the **Name** of the parent/responsible adult of the child in **Question 209**.
3. Read the statement in **Question 210** to seek consent for haemoglobin measurement of the child from the parent/responsible adult. Record the outcome of the consent process in **Question 211**; confirm that you read the statement to the parent/responsible adult and recorded their response accurately by signing in the space provided.
4. If consent was granted, collect a finger or heel stick blood sample from the child, following the procedure described in Chapter 4. Use a sterile gauze pad to wipe away the **first two** large blood drops from the finger or heel.
5. Conduct the haemoglobin measurement as follows:
  - o Step 1: Collect the capillary blood in the microcuvette:



Finger prick



Heel prick

Figure 4.4 Filling the microcuvette

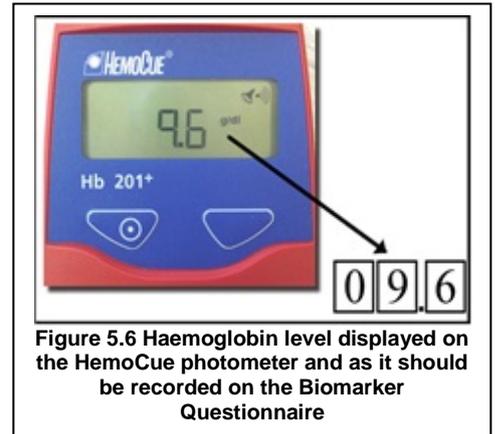
- Apply the tip of the HemoCue microcuvette to the middle of the blood drop. The microcuvette chamber will fill automatically by capillary action. The chamber needs to be filled completely (Figure 5.4). Never “top off” the microcuvette. Instead, if the microcuvette is not completely filled, use a fresh microcuvette and fill it with the next blood drop that forms.
- Wipe any surplus blood off both sides of the microcuvette “like butter from a knife,” using the clean end of a sterile gauze pad. Ensure that no blood is sucked out of the microcuvette when wiping the microcuvette.
- After filling the chamber, the microcuvette needs to be visually inspected for air bubbles. Since air bubbles may influence the haemoglobin measurement, any microcuvette containing air bubbles must be discarded. In such cases, with the



Figure 5.5 Placing the microcuvette in photometer

permission of the parent/ responsible adult, repeat the blood drop collection using a different finger (heel). Again, you must use new disposable supplies and follow all of the steps described previously in obtaining the new sample.

- Step 2: Obtain the haemoglobin level:
  - Place the microcuvette in its holder and gently push the holder into the photometer (Figure 5.5).
  - Reading the results: The microcuvette should be analysed immediately, and no later than ten minutes after being filled. The blood haemoglobin level in grams per decilitre (g/dl) is displayed after a delay of 15 to 45 seconds (Figure 5.6).



- Step 3: Stop bleeding at the site of the prick:
  - After the blood drop collection, wipe any remaining blood from the prick site with a sterile gauze pad. Press the gauze pad against the prick site until the blood flow has stopped completely.
  - Take an adhesive bandage from its wrapper and apply it to the prick site (Figure 5.7). Advise the mother, especially when the child is a toddler, to watch carefully that the child does not take off the bandage and put it in his/her mouth because it may choke the child.



- Step 4: Record the haemoglobin level and test result:
  - Record the haemoglobin level shown on the photometer (Figure 5.6) in the appropriate box in **Q. 212** of the biomarker section of the **Biomarker Questionnaire**. If there is no Hb result to record because the parent/responsible adult did not consent to the test, or there was some other problem, record the appropriate code in **Q. 212**. If more than one child in the household is eligible, check carefully that you are recording the haemoglobin level in the correct column of the Biomarker Questionnaire.
- Step 5: Collect biohazardous waste:
  - Place all biohazardous waste (e.g., lancets, microcuvettes, alcohol swabs, gauze, and gloves) into a plastic bag provided for field disposal of these items. At the end of the day, follow the procedures described in Chapter 9 for the proper disposal of these waste materials.

6. Record the child's haemoglobin level in the health card and anaemia brochure of either the parent or responsible adult. Inform the parent/responsible adult of the results and provide the parent with the brochure (see Appendix 6 or 7).
7. Provide a written referral to a health facility for medical treatment for any child with severe anaemia (haemoglobin level less than 7 g/dl; referral described at the end of chapter).

**Adults: Follow the procedure below for haemoglobin measurement in adults:**

1. Verify that the interviewer correctly entered the respondent's information in the Biomarker Questionnaire.
2. **For a respondent age 18-49 for women and 18-54 for men (or a respondent age 15-17 who has been in a union):**
  - a. Seek consent for measurement in **Questions 342/442** by reading the consent statement. If the adult does not consent to the haemoglobin measurement, record REFUSED in **Questions 343/443**, sign your name on the blank line. If the adult consents, record GRANTED in **Questions 343/443** and sign your name.
  - b. For a female respondent who has consented to measurement, record pregnancy status in **Questions 344**.
  - c. For a respondent age 15-17 who is not in a union, ask consent from the parent in **Questions 340/440**, and only if the parent consents to testing, can you proceed to request assent from the adolescent.
3. Prepare the equipment and supplies for the test for which consent has been granted as described in **Appendix 1**.
4. If consent was granted, collect blood from a finger prick following the procedure described in Chapter 4. Use a sterile gauze pad to wipe away the **first two** blood drops from the finger prick.
5. Conduct the anaemia test as described above for children:
  - Step 1: Collect the capillary blood in the microcuvette
  - Step 2: Obtain the haemoglobin level
  - Step 3: Stop bleeding at the site of the prick
  - Step 4: Record the haemoglobin level and test result in **Question 368/467**
  - Step 5: Collect biohazardous waste
6. Record the respondent's haemoglobin level in the anaemia brochure.
7. Inform the respondent of her/his haemoglobin level and provide the anaemia brochure. **For reasons of confidentiality, please provide each eligible adult who was tested with an informational brochure with their results recorded.**

8. Provide a written referral to a health facility for treatment for any respondent with severe anaemia (below 9g/dl for men and pregnant women and below 7g/dl for women who are not pregnant or don't know if they are pregnant and children).

#### PRECAUTIONS TO TAKE DURING HAEMOGLOBIN MEASUREMENT

Please take the following precautions while doing haemoglobin measurement:

- **Never remove a microcuvette from the container with fingers wet with alcohol.** This can result in alcohol coming into contact with the reagents inside the microcuvette which can inactivate the reagents. Using fingers wet with alcohol to handle other microcuvettes in the container can also affect them.
- **Never use the first two drops of blood for haemoglobin measurement.** If the respondent consents to the haemoglobin measurement, **wipe away the first two drops** of blood and then collect the third drop in the microcuvette. This ensures the free flow of blood and allows for the collection of blood with a representative concentration of red blood cells.
- **Avoid inadequate filling or re-filling of the microcuvette.** The compartment of the microcuvette that contains dry reagents (yellow portion) has to be completely filled. The microcuvette should be filled with a drop of blood in one continuous motion. A microcuvette that contains air bubbles should be discarded.
- **Wiping off blood on the microcuvette.** Blood on the exterior of the microcuvette should be removed. Failure to clean the exterior of the microcuvette can lead to an erroneously high haemoglobin reading.
- **Avoid keeping the microcuvette out for too long.** Keeping the microcuvette out of the container for too long before using it can lead to errors. Remove the microcuvettes from its container immediately before starting the testing procedure.
- **Avoid misalignment of the microcuvette in the photometer.** The microcuvette only fits into the photometer's microcuvette holder in one position. Therefore, place the microcuvette carefully in the holder and slowly push the holder inside the photometer to obtain a reading. Forcefully closing the microcuvette holder can cause blood to spray onto the optronic system, which can result in inaccurate Hb results.
- **Expired or improperly stored microcuvettes should not be used for testing.** While in the field, microcuvettes should not be used if more than 1 month has elapsed since the seal on the container was broken. The containers must be kept closed when not in use to avoid exposure to moisture, which can destroy the reagents.

#### PROVIDING HAEMOGLOBIN TEST RESULTS AND REFERRALS FOR SEVERE ANAEMIA

Before leaving the household, you will verbally report the results of the haemoglobin measurement for each person for whom a haemoglobin measurement was completed. In addition to verbally reporting the Hb results, Hb results will also be written in an informational brochure for each adult respondent in the household. For eligible children, their haemoglobin results should be recorded in

the brochure of their parent or responsible adult. When reporting the results, briefly explain to the respondent what his/her haemoglobin reading means, using the anaemia brochure as a guide. Please see Appendix 7 or 8 for an example of the anaemia brochure.

Respondents with severe anaemia should be informed about the effects of severe anaemia and encouraged to visit a health facility for follow-up medical attention. For each respondent with severe anaemia, you will fill out an Anaemia Referral Form (Appendix 3), on which you have recorded the haemoglobin level.

### **Summary of the steps involved in haemoglobin measurement:**

- Obtain consent
- Clean the finger or heel with an alcohol swab;
- Prick the finger or heel with the lancet;
- Wipe away the first two drops of blood;
- Collect the third blood drop in a microcuvette;
- Measure haemoglobin level in the blood sample using the HemoCue photometer;
- Stop the bleeding and apply an adhesive bandage to the puncture site;
- Record the haemoglobin level in the appropriate section of the Biomarker Questionnaire;
- Collect biohazardous waste;
- Inform the respondent of his/her haemoglobin level and provide an informational brochure on anaemia;
- Provide a written referral for follow-up medical attention for a respondent found to be severely anaemic

## **CHAPTER 6: BLOOD GLUCOSE TESTING**

### **INTRODUCTION**

Diabetes (diabetes mellitus) is a set of metabolic disorders that is characterized by hyperglycaemia (elevated blood glucose levels) due to defective insulin secretion, ineffective insulin action, or a combination of the two. Early symptoms of hyperglycaemia may include polyuria, polydipsia, weight loss and blurred vision. Long-term complications of diabetes cover retinopathy, loss of vision, kidney failure, and wounds that do not heal (or heal very slowly). As a consequence, diabetes is the major cause of non-traumatic amputations of the lower limbs, blindness and kidney disease worldwide. Glucose is derived primarily from digestion of carbohydrates and is the principal source of energy for cells. Insulin is required for the transfer of glucose from the blood into cells where it can be utilized for energy. Impairment of the glucose transporting mechanism, either as a result of insufficient insulin (autoimmune disease), cancer of the pancreas, viruses or drugs) or insulin that does not work properly (obesity, inactivity) may lead to diabetes

### **CAUSES OF DIABETES**

Diabetes is a poorly understood condition and the cause (s) of the disorder is/are not clearly known. Diabetes may be classified into two primary groups based on the aetiology of the disease: Type 1 diabetes and type 2 diabetes. Type 2 diabetes mellitus is the most common form of diabetes and is strongly linked to obesity and a sedentary life style. Individuals with a family history of diabetes are more likely to develop the condition than the general population. In addition to lifestyle factors, diabetes mellitus (type 2 diabetes) may result from trauma to the pancreas, endocrine disorders (hyperthyroidism; Cushing's syndrome), prescription drugs and infections (congenital rubella). The underlying cause of type 2 diabetes is a deficiency in insulin secretion and or insulin that is partially effective in removing glucose from the blood and into cells. Treatment options range from weight reduction to drugs that promote the secretion of insulin or enhance its effectiveness. Type 1 diabetes is characterized by a lack of insulin due to destruction of the beta cells of the pancreas. Individuals with type 1 diabetes require exogenous insulin for survival.

### **BLOOD GLUCOSE TESTING IN NFHS-4**

Women age 15 to 49 and men age 15-54 in a subsample of households are eligible for blood glucose testing in NFHS-4. Note that in households selected for men's interview, all women age 15 to 49 and men age 15 -54 are eligible for blood glucose testing. Health investigators should keep in mind that random blood glucose testing is done at any time of the day in combination with other biomarkers that the respondent has given her/his consent.

### **GENERAL PROCEDURE IN CONDUCTING THE RANDOM BLOOD GLUCOSE TEST**

This chapter discusses the materials needed and the procedure for random blood glucose testing. In addition, directions are given regarding precautions to take during blood collection and testing, recording results in the Biomarker Questionnaire, and providing test results to the respondents.

## MATERIALS AND SUPPLIES FOR BLOOD GLUCOSE TESTING

In addition to the Biomarker Questionnaire and supplies listed in Chapter 4, the following equipment and supplies are required for random blood glucose level measurement.

The FreeStyle Optium H glucometer with a glucose test strips will be used in NFHS-4 to measure random blood glucose. This equipment provides laboratory quality analysis of glucose with whole blood. The glucometer and test strip uses whole blood for glucose measurement and results are displayed as whole blood glucose. The standard way of reporting glucose measurement in blood is as plasma glucose. The blood plasma glucose level is about 11% higher than the level in whole blood. The conversion of whole blood glucose to plasma glucose will be calculated during data processing. The health investigators will record, in the biomarker questionnaire, the measured blood glucose value on the glucometer display.

**The components of the FreeStyle Optium H glucometer include the following:**

### 1. Glucometer (FreeStyle Optium H)

It is a device for measuring the concentration of glucose in the blood (Figure 6.1/6.2). It's measurement of blood glucose level is equivalent to the glucose levels using the glucose oxidase laboratory technique, in the range of 20-500mg/dl (1.1 – 27.8 mmol/L). The advantage of the glucometer is that results are available in 20 seconds on an LCD digital display which can be immediately communicated to respondents. A very small volume of blood is required for the blood glucose measurement. This device is suitable for use as point-of care equipment. A small drop of blood obtained from a finger prick is required by a disposable glucose test strip and is read by the glucometer. The glucometer then displays the results of the glucose measurement in mg/dl.



Figure 6.1 Glucometer

### 2. Disposable test strip

This is a plastic test strip with a small spot impregnated with glucose oxidase and other components (Figure 6.2). Each disposable test strip is used once and then discarded. Blood drop is filled into a glucose strip once strip is attached to glucometer. Usually they come in packs of 100 strips per vial along with a glucose test strip calibrator.



Figure 6.2 Disposable strips



Figure 6.3 Prepare glucometer for use

### 3. Calibration of glucometer

The Freestyle Optium **H** meter **must** be used with FreeStyle Optium H strips only. The meter requires calibrating for every new box of test strips using the calibrator strip supplied in box. Failure to calibrate properly will cause incorrect results.

- a. With the lot number facing upward, insert the contact bars of the calibrator into the monitor. The monitor will turn on automatically.
- b. The lot number of the calibrator strip and test strip foil package will appear in the display window.
- c. Check that the lot number matches on all the items; Meter display window, test strip calibrator, test strip instructions for use, test strip foil packet. If the Lot # on all these items matches, calibration is complete
- d. Use only the calibrator supplied with the test strips. Keep the calibrator until all the test strips in the box have been used then discard the calibration strip.

#### 4. MediSense Glucose Control Solutions (supplementary reagents not included in the kit)

- a. When you open a control solution bottle for the first time, count forward 90 days and write this date on the control solution bottle using a pen that won't smear or wipe off. Throw away any remaining solution after this date.
- b. Control solutions may be used with glucose test strips. **The control solution is used to confirm that the meter and test strips work together correctly.**
- c. Use only MediSense Glucose control solutions with this glucometer. Replace the cap securely on the bottle immediately after use.
- d. Do not use the control solution past the expiry date.
- e. Control solution results should fall within the control solution range printed on the test strip instructions for use. Check that the lot number printed on the test strip packet and instructions for use match.
- f. Low (Lo) and high (Hi) controls must be assayed. Control ranges are located on the package insert of Test strips. Retain package insert until box of test strips has been used.
- g. Repeat the test if control solution results are outside this range.
- h. Stop using the meter if control solution results are continually outside the range printed on the test strip instructions for use. Contact Customer Service.
- i. The Freestyle Optium H glucose device must be checked with the control solutions as required depending on usage.



Figure 6.4 Glucose control solutions

#### STEPS FOR MEASURING RANDOM BLOOD GLUCOSE LEVEL USING THE FREESTYLE OPTIUM H DEVICE

Open the foil test strip packet at the notch and tear down to remove the test strip.

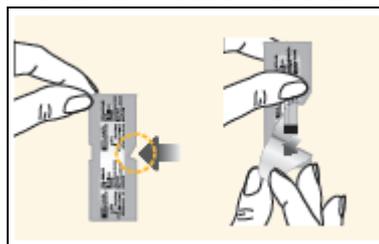


Figure 6.4 Prepare glucose strip for use

1. Insert the test strip into the meter with the contact bars (3 black lines) facing up until it stops. This turns on the meter. **Note:** *The meter turns off after 3 minutes of inactivity. Remove and reinsert the unused test strip to restart the meter.*



Figure 6.5 Insert glucose test strip in glucometer

- This display always appears when the meter is turned on. Do not use the meter if the display check screen does not exactly match the example. Contact Customer Service.

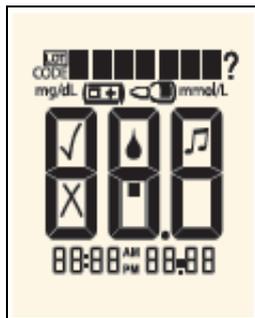


Figure 6.6 glucometer screen check

- The “Apply Sample” symbols  and  appear next, indicating the meter is ready for you to apply a sample to the test strip.



Figure 6.7 “Apply Sample” symbols on glucometer display

- Obtain a Blood Sample: Select a test site. Use the retractable safety lancet to obtain a blood sample.



Figure 6.8 Prick finger

7. Apply Blood to the test strip by bringing the blood drop to the white area at the end of the test strip. The blood is drawn into the test strip.

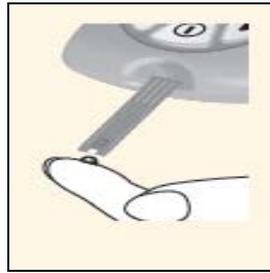


Figure 6.9 Collect blood in to glucose test strip

8. Hold the blood drop to the white area until the meter beeps (if sound is on) and the status bar appears on the display. This indicates the test strip has obtained enough blood.

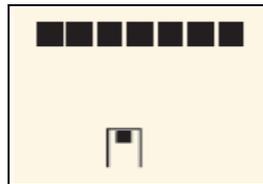


Figure 6.10 Test strip has required blood volume

9. The countdown appears on the display as the meter checks your glucose level. **Note:** Do not remove the test strip from the meter or disturb the test strip during the countdown.

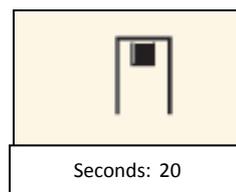


Figure 6.11 Countdown display during glucose measurement

10. The meter beeps (if sound is on) when the result appears on the display. The test is complete (see figure). Record this result in your biomarker questionnaire. The result is also stored in memory of the FreeStyle Optium H glucometer device.



Figure 6.12 Blood glucose results on display screen

11. Turn off the meter by removing the used test strip from the glucometer. Discard the used test strip in a biohazard bag. **Note:** You can also press and hold  for at least 2 seconds to turn off the meter. The meter also turns off after 60 seconds of inactivity.
12. The meter displays results in mg/dL or mmol/L. The unit of measurement is preset by the manufacturer. You cannot change this setting.
13. The meter displays results from 20–500 mg/dL (1.1–27.8 mmol/L). Low or high blood glucose results can indicate a potentially serious medical condition.
14. **In case of delay of more than 3 minutes of inactivity before applying blood to the glucose strip,** the glucometer turns off. Do not panic, simply remove and reinsert the unused strip into the glucometer to restart the meter.
15. Stop the bleeding at the prick site with a sterile gauze pad.
16. Record the results of the random blood glucose measurement in the biomarker questionnaire for women and men in **Question 370/469** respectively.
17. Record the respondent's random blood glucose level in the health card and in the blood glucose brochure. Explain the results to the respondent and (if adolescent) parent/responsible adult and provide a health card and an informational brochure on blood glucose. Each respondent should have a blood glucose brochure with their results recorded.
18. **Provide a referral form to a health facility for additional medical evaluation for any respondent with a random blood glucose level  $\geq 200$  mg/dl.**
19. Discard all biohazardous waste (used lancets, glucose strip, alcohol swabs, gauze, and gloves) into a plastic biohazard bag provided for field disposal of these items. At the end of the day, follow the procedures described in Chapter 9 for the proper disposal of waste materials.

## ERROR MESSAGES

### Error Messages

Message	What It Means	What To Do
E-1	The temperature is too hot or too cold for the meter to work correctly	<ol style="list-style-type: none"> <li>1. Move the meter and test strips to a location where the temperature is within the test strip operating range. (See test strip instructions for use for the appropriate range.)</li> <li>2. Wait for the meter and test strips to adjust to the new temperature.</li> <li>3. Repeat the test using a new test strip.</li> <li>4. If the error reappears, contact Customer Service.</li> </ol>
E-2	Meter error	<ol style="list-style-type: none"> <li>1. Turn off the meter.</li> <li>2. Repeat the previous testing steps.</li> <li>3. If the error reappears, contact Customer Service.</li> </ol>

Message	What It Means	What To Do
E-3	<p>The blood glucose level may be too low to be read by the system or</p> <p>There may be a problem with the test strip</p>	<ol style="list-style-type: none"> <li>1. Review the testing instructions.</li> <li>2. Repeat the test using a new test strip.</li> <li>3. If the error reappears, contact your healthcare professional <b>immediately</b>.</li> </ol>
E-4	<p>The blood glucose level may be too high to be read by the system or</p> <p>There may be a problem with the test strip</p>	<ol style="list-style-type: none"> <li>1. Repeat the test using a new test strip.</li> <li>2. If the error reappears, contact your healthcare professional <b>immediately</b>.</li> </ol>
E-5	Blood was applied to the test strip too soon	<ol style="list-style-type: none"> <li>1. Review the testing instructions.</li> <li>2. Repeat the test using a new test strip.</li> <li>3. If the error reappears, contact Customer Service.</li> </ol>

Message	What It Means	What To Do
E-6	Meter error	<ol style="list-style-type: none"> <li>1. Check that you are using the correct strip for this meter. (See test strip instructions for use to verify that your strip is compatible with this meter.)</li> <li>2. Repeat the test using a test strip for use with your meter.</li> <li>3. If the error reappears, contact Customer Service.</li> </ol>
E-7	No coding required or Test strip may be damaged, used or the meter does not recognise it	<ol style="list-style-type: none"> <li>1. Check that you are using the correct test strip for this meter. (See test strip instructions for use to verify that your strip is compatible with this meter.)</li> <li>2. Repeat the test using a test strip for use with your meter.</li> <li>3. If the error reappears, contact Customer Service.</li> </ol>
E-8 or E-9	Meter error	<ol style="list-style-type: none"> <li>1. Turn off the meter.</li> <li>2. Repeat the previous testing steps.</li> <li>3. If the error reappears, contact Customer Service.</li> </ol>

## TROUBLESHOOTING

### Troubleshooting

	What It Means	What To Do
The meter does not enter test mode after inserting a test strip.	Test strip is not inserted correctly or fully into the meter	<ol style="list-style-type: none"><li>1. With the contact bars (3 black lines) facing up, insert the test strip into the meter until it stops. This turns on the meter.</li><li>2. If the meter still does not enter test mode, contact Customer Service.</li></ol>
	No battery is installed Battery is installed incorrectly	<ol style="list-style-type: none"><li>1. Install battery with (+) facing up.</li></ol>
	Dead battery	<ol style="list-style-type: none"><li>1. Replace battery. Reset date and time, if necessary.</li></ol>

	What It Means	What To Do
The test does not start after applying the blood sample.	Blood sample is too small	<ol style="list-style-type: none"> <li>1. See test strip instructions for use for re-application instructions.</li> <li>2. If the countdown still does not start, remove the used strip and discard it correctly.</li> <li>3. Repeat the test using a new test strip.</li> <li>4. If the test still does not start, contact Customer Service.</li> </ol>
	Sample applied after meter turns off	<ol style="list-style-type: none"> <li>1. Remove the used strip and discard it correctly.</li> <li>2. Review the testing instructions.</li> <li>3. Repeat the test using a new test strip.</li> <li>4. If the test still does not start, contact Customer Service.</li> </ol>

	What It Means	What To Do
The test does not start after applying the blood sample.	Defective meter or test strip	<ol style="list-style-type: none"> <li>1. Remove the used strip and discard it correctly.</li> <li>2. Repeat the test using a new test strip.</li> <li>3. If the test still does not start, contact Customer Service.</li> </ol>

## MAINTENANCE OF THE GLUCOMETER

### Maintaining the Meter

#### Replacing the Battery

	<p>The meter comes with a CR 2032 lithium (coin cell) battery installed. It provides power for about 1,000 tests.</p>
	<p> • Appears on the display when the battery is low. • Causes the display light to fail to turn on.</p> <p>When  appears, you may still use the meter and the results will be accurate. The display light will not work. Replace the battery immediately.</p>
	<p>When  appears alone, the meter is not usable. The meter turns off automatically. Replace the battery immediately.</p>
	<ol style="list-style-type: none"><li>1. Gently push the battery cover in and up to remove it.</li></ol>



2. If the meter includes a plastic tab, pull on the tab to remove the battery. If the meter does not include a plastic tab, push the battery upwards to remove it.



3. Insert a new CR 2032, lithium (coin cell) battery: (+) facing up.



4. Slide the battery cover into place until it clicks.

**Note:** Dispose of used batteries correctly.



5. Press and hold **I** to turn the meter on. If the meter does not turn on, check to see that the battery was installed correctly.

If meter turns on, the meter may prompt you to reset the time and date. (See "Setting Up the Meter" in this user's manual.)

**Note:** Test results will not be lost even if the meter loses its time and date settings.

## CLEANING THE GLUCOMETER

### Cleaning the Meter

Avoid getting dirt, dust, blood, control solution or liquid in the meter test strip port. Clean the outside of the meter using a damp cloth and mild soap.

**Healthcare Professionals:**

Acceptable cleaning solutions include:

- 70% isopropyl alcohol or
- A mixture of 1 part ammonia, 9 parts water or
- A mixture of 1 part household bleach, 9 parts water

**IMPORTANT:**

**Do not** try to clean the test strip port.

**Do not** pour liquid into the test strip port or onto the buttons.

**Do not** immerse the meter in water or other liquid.

## CHAPTER 7: COMBINED HAEMOGLOBIN AND RANDOM BLOOD GLUCOSE TESTING

In about 85% of eligible households selected for the NFHS-4, women age 15 to 49 will be eligible for combined haemoglobin and blood glucose measurement. In chapters 5 and 6 above, the procedures for haemoglobin and blood glucose measurements have been described as independent tests.

The order of collection is very important and must be followed strictly. Blood is to be collected for haemoglobin measurement first, while collection of blood for blood glucose testing follows. Consent for each test must be obtained separately, following the order in the Biomarker Questionnaire. If consent has not been given to perform a test, follow the skip instructions. **You should complete the testing process for one eligible respondent before proceeding to the next eligible individual.**

### PROCEDURE FOR COMBINED HAEMOGLOBIN AND BLOOD GLUCOSE TESTING

1. The interviewer identifies all women eligible for haemoglobin and blood glucose testing from the household schedule and records their name, line number, age, and marital status in **Question 302** of the Biomarker Questionnaire. Women age 15-49 who are usual members of the household or who slept in the household the night before the day of the interview are eligible.
2. When instructed, the health investigator must check the respondent's age and marital status in **Question 302**. You must do this step because if the respondent is age 15-17 and not in a union, or has never been in a union, he/she is considered to be an adolescent and consent for testing must be obtained from the parent or adult responsible for the adolescent as well as the adolescent. If the respondent is in a union or has been in a union or is age 18-49, skip to the appropriate question.
3. **For a respondent age 18-49 or a respondent age 15-17 who is in a union, or has been in a union:**
  - Read the appropriate informed consent statement in the Biomarker Questionnaire separately for haemoglobin and blood glucose testing to the respondent and record the outcome of the consent request in the appropriate question.
  - For a female respondent who has consented to haemoglobin measurement, record pregnancy status in **Question 344**.
4. **For a respondent age 15-17 who has never been in a union (an adolescent):**
  - Record the **Name** of parent/responsible adult in the appropriate questions for haemoglobin and blood glucose measurement in **Question 307**.
  - Seek consent for haemoglobin and blood glucose measurements separately from the parent/responsible adult in the appropriate questions. If the parent/responsible adult does not consent to any or both tests, record REFUSED, sign your name on the blank line in the appropriate Questions. If the parent/responsible adult consents to

any or both tests, record GRANTED in the appropriate Questions and sign your name.

- Ask consent for the haemoglobin and blood glucose testing from the adolescent and record the outcome of the consent request in the appropriate Question. If the adolescent respondent does not consent to any testing, skip to the appropriate Question. Note that for adolescents both the parent/responsible adult must consent before testing can be done.
- For a female adolescent respondent who has consented to haemoglobin measurement, record pregnancy status in **Question 344**.

5. If consent was granted for both haemoglobin and blood glucose testing, the health investigator puts on a pair of gloves and prepares the work station with the appropriate biomarker supplies and equipment.
6. Prick a finger with a Unistik 3 Normal lancet and wipe away the first two large drops of blood.



7. Collect the third blood drop into a microcuvette for haemoglobin measurement by applying the tip of the microcuvette in the middle of the large third drop.
8. Clean the sides of the microcuvette on the sterile gauze and check for air bubbles. Haemoglobin level is read in the HemoCue machine in about 15 seconds.
9. Use the sterile gauze to wipe away the excess blood on the skin and allow a fourth drop to form.



10. Apply Blood to the test strip by bringing the blood drop to the white area at the end of the test strip. The blood is drawn into the test strip.
11. Hold the blood drop to the white area until the meter beeps (if sound is on) and the status bar appears on the display. This indicates the test strip has obtained enough blood.
12. The countdown appears on the display as the meter checks the glucose level. **Note:** Do not remove the test strip from the meter or disturb the test strip during the countdown.
13. The meter beeps (if sound is on) when the result appears on the display. The test is complete. Record this result in the biomarker questionnaire. The result is also stored in the memory of the FreeStyle Optium H glucometer device.

14. Wipe any remaining blood from the prick site with a sterile gauze pad. Press the gauze pad against the prick site until the blood flow has stopped completely.
15. Take an adhesive bandage from its wrapper and apply it to the prick site.
16. Record the **haemoglobin and blood glucose levels in Question 368 and 370 respectively**. If haemoglobin or blood glucose measurement was not obtained because the respondent did not consent to the test, or there was a technical problem with the testing, circle the appropriate code.
17. Place all biohazardous waste (used lancets, alcohol swabs, gauzes, microcuvette, glucose strip and gloves) into a clearly labelled "biohazard" plastic bag, which has been provided for field disposal of these items. Before leaving the enumeration area (cluster) the bags with biohazardous waste must be taken to a health facility in the enumeration area and incinerated or disposed of in the field following the recommended protocol in Chapter 9.
18. Record the test results in the health card and in the appropriate informational brochures. Each eligible respondent should receive the appropriate brochures with their test results recorded.
19. Explain to the respondent what her haemoglobin and blood glucose results mean with the brochure as a guide.
20. Provide a written referral to a health facility for treatment for any respondents with severe anaemia (below 9 g/dl for pregnant women and men and below 7 g/dl for women who are not pregnant or don't know if they are pregnant).
21. Provide a written referral to a health facility for additional medical evaluation for any respondents with a random blood glucose level  $\geq 200\text{mg/dl}$ .

## **CHAPTER 8: COMBINED HAEMOGLOBIN, BLOOD GLUCOSE TESTING AND DRIED BLOOD SPOT (DBS) COLLECTION FOR HIV TESTING**

Acquired Immune Deficiency Syndrome (AIDS) is a disorder of the immune system caused by the human immunodeficiency virus (HIV). People with AIDS are vulnerable to the development of life-threatening conditions from relatively innocuous infections that people with a properly functioning immune system do not typically develop.

Most of the current information on HIV prevalence in India comes from surveillance of women attending antenatal clinics or of other special populations such as commercial sex workers or individuals treated at health facilities for sexually transmitted infections. However, surveillance data do not yield an estimate of the prevalence of HIV among the general population. For example, the antenatal surveillance system excludes men and non-pregnant women. Since HIV is transmitted principally through sexual contact, obtaining an estimate of HIV prevalence for both men and women will provide a better estimate of the current level of HIV in India than is available from other sources. Therefore, to estimate the national prevalence of HIV in the NFHS-4, a nationally-representative sample of women 15-49 and men 15-54 will be tested.

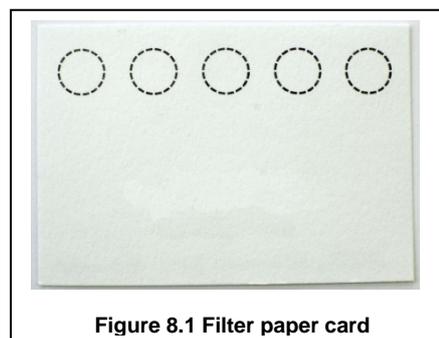
HIV testing itself does not take place in the field. Instead, blood samples are collected from a finger prick on a filter paper card, dried, transported to the designated laboratories for logging in and checking. The dried blood spots (DBS) will be stored at the laboratory until field work is completed and then processed for testing in batches.

HIV testing in NFHS-4 is anonymous. Personal identifiers are delinked from the DBS samples and respondents who agree to be tested are not told their results. Instead, respondents are given a voucher for voluntary counselling and HIV testing at a nearby health facility. For individuals living in communities in which there is no health facility with the capacity to provide counselling and testing within [15] km distance, arrangements will be made to bring mobile VCT to the community.

## MATERIALS AND SUPPLIES FOR DBS COLLECTION FOR HIV TESTING

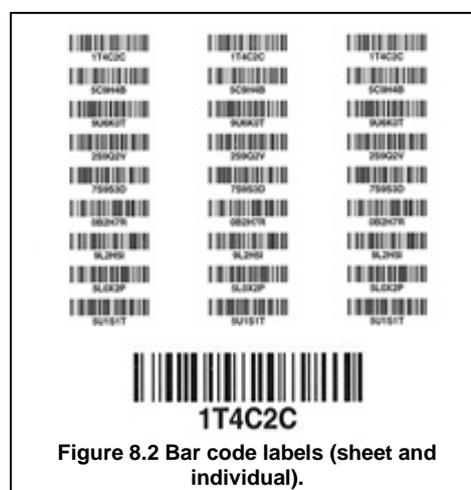
In addition to the supplies listed in Chapter 4, the following materials are required for DBS collection for HIV testing:

- **Filter paper card:** You will use special filter paper cards (Figure 8.1) to collect the blood samples. Each card has five pre-printed circles that hold about 100 µl of blood when filled.



- **The filter paper cards must be kept clean and dry at all times.** Water, dust, sweat from your hands, or other environmental contaminants can affect HIV testing. **Use gloves at all times when handling the filter paper cards.**
- The filter paper cards come in packets of 100 cards. Before opening a new packet, put on gloves. Open the packet and place the cards in 3-4 smaller zip-loc bags; place these smaller bags in a large zip-loc bag. The bags must be stored stacked so as to avoid compressing the filter paper. **Place a few desiccant packets and a humidity indicator card in each small bag before sealing it. You must also place desiccants and a humidity indicator card in the larger zip-loc bag.** If the humidity indicator card and/or desiccants change colour before the cards have all been used, replace the humidity indicator card and desiccants following the instructions described in the **Desiccant packets** and **Humidity indicator card** sections below.
- **Note:** Keeping the unused filter paper cards with desiccants in a zip-loc bag will prevent moisture from being absorbed on the filter paper card which will prevent over-saturation or merging of circles when blood is collected.

- **Bar code labels:** because the HIV testing in the NFHS-4 is anonymous, respondents' names are never written on the filter paper cards. Instead, bar code labels are used to identify the DBS samples and link them to the interview data. You will be provided with sheets of "peel-off" adhesive bar code labels (Figure 8.2). The barcodes are arranged in rows; the codes on each label are the same across one row. **A different row of bar code labels is to be used for each respondent for whom a DBS sample is collected.**



- **Drying box:** a plastic box with a cardboard rack positioned inside.
  - For proper use, stand the box vertically (the filter paper cards will be positioned horizontally). The drying boxes are to be used for overnight drying of DBS samples. They are not to be used for long-term sample storage.

- **Desiccant packets:** drying agents used to absorb moisture from the air to keep the filter paper cards dry (Figure 8.3). The granules inside the packets change colour from blue to pink as they absorb moisture.



Figure 8.3 Desiccants

- Change the desiccants when the granules change to a pink colour or as indicated by the humidity indicator card.
- Treat used desiccants as biohazardous waste and throw them away in a biohazardous waste bag.

- **Humidity indicator cards:** cards that allow closer monitoring of the level of moisture than monitoring the colour of the desiccant packets alone.

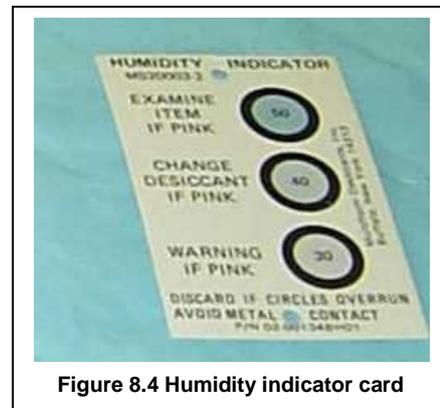


Figure 8.4 Humidity indicator card

- There are three circles on the humidity indicator card (Figure 8.4). If the circle at the bottom of the card (labelled 30%) turns pink, it indicates a relatively high level of humidity and is a warning to carefully monitor the humidity level. If the middle circle (labelled 40%) turns pink, replace the desiccant packets and humidity card. If the top circle (50%) turns pink, you must examine the DBS cards as their quality might have been compromised due to the high humidity in the bag. You should replace the desiccant packets and humidity card with fresh ones.

- **Glassine paper or weighing paper:** thin, glossy, semi-opaque paper squares used to protect the dried blood spots on the filter paper cards during storage.
- **Low gas-permeable bags (small zip-loc bags):** special small zip-loc bags used for storing the DBS samples prior to transfer to the lab. These bags are specially manufactured to reduce the exposure of their contents to air and moisture. These bags are expensive and should never be used for other purposes, such as carrying food or adhesive bandages. The bags have a sliding “zipper” that is used to close and seal the bag.

- **Large zip-loc bags or Cluster bag:** A large zip-loc bag will be provided for each of the NFHS-4 sample clusters in which you will work. These bags will be used to hold the small zip-loc bags with DBS samples from the cluster during storage and transport to the laboratories.
- **DBS Transmittal Sheet:** accompanies the DBS samples to the laboratory. The purpose of this sheet is to track the samples from the field to their arrival in the laboratory for testing so as to ensure that the number of DBS samples sent to the laboratory matches the number of samples collected in the field. A bar code with the same unique identifier as the bar code label attached to the DBS sample is attached to the DBS Transmittal Sheet and in the space provided in the Biomarker Questionnaire (**Question 371 for women or Question 470 for men**). See Appendix 9 for an example of the DBS Transmittal Sheet.
- **Voucher/Coupon for free HIV Integrated Counselling and Testing Centre (ICTC) Services:** contains information regarding HIV, ICTC services, and a list of local ICTC centres where respondents can receive HIV testing. See Appendix 5 for an example of a Voucher/Coupon for free ICTC services.

## COMBINED PROCEDURE FOR HAEMOGLOBIN AND GLUCOSE TESTING AND DBS COLLECTION FOR HIV TESTING

This section focuses on the steps involved in collecting the blood samples for HIV, haemoglobin and blood glucose testing. When blood is collected for HIV, haemoglobin and blood glucose testing, the order of collection is very important and must be followed strictly. Blood is first collected to prepare dried blood spots for HIV testing, while collection of blood for haemoglobin measurement precedes that for blood glucose testing.

Consent for each test must be obtained separately, following the order in the Biomarker Questionnaire. If consent has not been given to perform a test, follow the skip instructions. **You should complete the testing process for one respondent before proceeding to the next eligible individual.**

**Follow the procedure below to collect blood for DBS, haemoglobin and blood glucose testing.**

1. The interviewer identifies all women and men from the household schedule who are eligible for haemoglobin, blood glucose and DBS collection. In approximately 15% of clusters, women age 15-49 and men 15-54 years who are usual members of the household or who slept in the household the night before are eligible.
2. Interviewer records the **Name, Line Number, Age, and Marital Status** of all eligible women and men in **Question 302** and **Question 402**, respectively, in the Biomarker Questionnaire.
3. When instructed, the health investigator must check the respondent's age and marital status in **Question 302/403**. You must do this step because if the respondent is age 15-17 and not in a union, or has never been in a union, he/she is considered to be an adolescent and consent for testing must be obtained from the parent or adult responsible for the adolescent as well as the adolescent. If the respondent is in a union or has been in a union or is age 18-49, skip to **Question 342/442**.

**For a respondent age 18-49 for women and 18-54 for men or a respondent age 15-17 who is in a union, or has been in a union:**

- Read the informed consent statement for anaemia in **Question 342/442** to the respondent and record the outcome of the consent request in **Question 343/443**. If a female respondent does not consent to haemoglobin measurement, follow skips to **Question 345**.
- For a female respondent who consents to haemoglobin measurement, record pregnancy status in **Question 344**.
- Read the informed consent statement for blood glucose in **Question 349/448** to the respondent and record the outcome of the consent request in **Question 350/449**. If the respondent does not consent to blood glucose testing, follow skips to **Question 353/453**.

- Read the informed consent statement for **HIV testing** to the respondent in **Question 358/457**. Record the outcome of the consent request in **Question 359/458**; confirm that you read the statement to the respondent and recorded their response accurately by signing in the space provided and entering your interviewer number. If the respondent does not consent to HIV testing, follow the skips to **Question 367/466**.
- Read the informed consent statement for **additional testing** to the respondent in **Question 364/463**. Record the outcome of the consent request in **Question 365/464**; confirm that you read the statement to the respondent and recorded their response accurately by signing in the space provided.
- If the respondent did not grant consent for additional testing, write “**No additional tests**” on the filter paper card.

**For a respondent age 15-17 who has never been in a union (an adolescent):**

- Check and record name of his/her parent/responsible adult in **Question 307/407**.
- Seek consent for haemoglobin measurement from the parent/responsible adult in **Question 340/440**. If the parent/responsible adult does not consent to the adolescent’s haemoglobin measurement, in **Question 341/441** record REFUSED, sign your name on the blank line, and go to **Question 347/446**. If the parent/responsible adult consents to the testing, in **Question 341/441** record GRANTED, sign your name, and go to **Question 342/442**.
- Ask assent for the haemoglobin measurement from the adolescent in **Question 342/442** and record the outcome of the consent request in **Question 343/443**. If female adolescent respondent does not consent to haemoglobin measurement, skip to **Question 345**.
- For a female adolescent respondent who has consented to haemoglobin measurement, record pregnancy status in **Question 344**.
- Seek consent for **blood glucose testing** from the parent/responsible adult by reading the consent statement in **Question 347/446** and if consent is granted, proceed to request consent from the adolescent in **Question 349/448** and record the consent outcome. Ensure you correctly follow the skip instructions in the Biomarker Questionnaire.
- Seek consent for **HIV testing** from the parent/responsible adult by reading **Question 356/455**. If the parent/responsible adult does not consent to the HIV test, record REFUSED in **Question 357/456**, sign your name on the blank line, and go to **Question 367/466**. If the parent/responsible adult consents to the testing, record GRANTED in **Question 357/456** and sign your name.

- Read the informed consent statement for HIV testing to the adolescent respondent in **Question 358/457**. Record the outcome of the consent request in **Question 359/458**; confirm that you read the statement to the respondent and record their response accurately by signing in the space provided.
  - Seek consent to store blood for **additional testing** from the parent/responsible adult by reading **Question 362/461**. If the parent or responsible adult does not consent to additional testing, record REFUSED in **Question 363/462**, sign your name on the blank line, and go to **Question 366/465**. If the parent/responsible adult consents, record GRANTED in **Question 363/462** and sign your name.
  - Read the informed consent statement for **additional testing** to the adolescent respondent in **Question 364/463**. Record the outcome of the consent request in **Question 365/464**; confirm that you read the statement to the adolescent respondent and recorded their response accurately by signing in the space provided.
  - If either the parent or responsible adult, or the adolescent respondent does not grant consent for additional testing, write **“No additional tests”** on filter paper card.
4. Prepare the equipment and supplies **ONLY for the tests for which consent has been granted.**
5. If consent for HIV testing was granted, identify the next available complete set (**row**) of bar code labels. **Wearing a pair of gloves**, carefully remove a new filter paper card from the plastic zip-loc bag in which you have stored the cards. Make sure to handle the card in such a way that you do not touch the areas within the pre-printed circle. ***Never handle a card with your bare hands as you may transfer sweat, dirt or other contaminants on to the card.***
- Place the card with the pre-printed circles face-up on the clean absorbent sheet that you have spread out on a flat surface. Discard the card if it drops on the floor or ground or if it becomes dirty in any other manner.
  - Take the **first bar code label** from the first complete row on the sheet of bar code labels and paste it in the appropriate column of the Biomarker Questionnaire containing the line number of the respondent from whom you will collect blood for HIV testing (**Question 371** for women or **Question 470** for men).
  - Take the **second bar code label** from the same row on the sheet of bar code labels and paste it at the bottom of the filter paper card. Do not cover up or touch any part of the pre-printed circles.
  - Take the **third bar code label** from the **same row** on the sheet of bar code labels and paste it on the Blood Sample Transmittal Sheet for the cluster in which you are working.

- **DO THE ABOVE STEPS CAREFULLY.** The bar code label is the only means of identifying the blood sample and for linking the HIV test results to the interview data. Mistakes will result in mismatches later on. **CHECK THAT THE THREE MATCHING BAR CODE LABELS HAVE BEEN PLACED ON THE FILTER PAPER CARD, THE BIOMARKER QUESTIONNAIRE, AND THE TRANSMITTAL SHEET BEFORE YOU PROCEED TO COLLECT BLOOD DROPS FROM THE RESPONDENT.**

**6. Note that the blood collection and test order for combined DBS, haemoglobin and blood glucose testing is as follows:** After pricking the finger:

- First large blood drop: wipe away.
- Second, third and fourth blood drops on the filter paper card. Wipe off excess blood on skin.
- Fifth blood drop into the microcuvette.
- Sixth blood drop into the glucose strip.
- Seventh and eighth blood drops in the last two circles of the filter paper card.

**7. Obtaining Blood from the Finger as follows:**

- Follow the steps for producing a finger stick blood sample as described in Chapter 4. Use a sterile gauze pad to wipe away the first blood drop. While maintaining a firm grip on the finger, press gently on the side of the finger from which you are taking the blood sample to form a large second drop (Figure 8.5). Be careful to **avoid 'milking' or 'squeezing' the finger** as this could affect the test results. Wait until the drop is large enough to fill one of the pre-printed circles on the card.

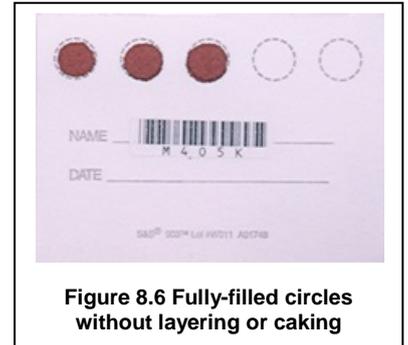


**Figure 8.5 Large drop of blood**

- **Position of Filter Paper Card**
  - Move the card underneath the finger, with the pre-printed side of the card facing the pricked finger.
  - The card must not be pressed against the prick site on the finger. Make sure that the respondent's finger does not touch the card at any point when you are collecting the blood spots.
- **Blood Collection on the card:**
  - 1) Let the blood drop fall freely in the centre of the pre-printed circle. In case the blood drop does not fall readily, you may touch the filter paper gently against a **LARGE blood drop** (but not the skin). In one step, a **large blood drop** should be allowed to soak through and completely fill the circle.

2) You must continue to collect drops of blood until you have fully saturated three circles on the filter paper card (Figure 8.6). Three blood spots are obligatory; however, we recommend that all five circles be filled.

3) To enhance blood flow, gently apply intermittent pressure to the area surrounding the prick site to get a third drop. Allow sufficient time for a large blood drop to form before filling the next circle on the filter paper card. Again, avoid milking or squeezing the finger.



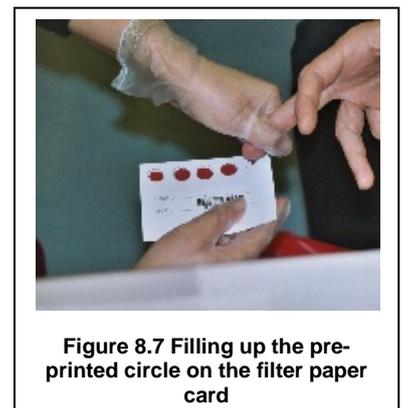
4) If the blood flow stops or decreases before you fully saturate three circles, you will need to do another finger prick. **Whenever this is necessary, you should explain to the respondent that you were unable to obtain an adequate sample and ask permission to obtain blood from another finger.** Use fresh supplies and a **different finger** for the second finger prick.

5) After collecting three spots on the filter paper card, place the filter paper card with the blood spots on the absorbent paper sheet away from other items. Wipe away and excess blood on the skin using the sterile gauze.

- Apply gentle pressure to form a **fifth blood drop** which will be collected in to the microcuvette for haemoglobin measurement in the HemoCue Hb 201+ device.

- Wipe away excess blood on the skin and apply gentle pressure to form a **sixth blood drop** which will be allowed to touch the correct end of the glucose strip for glucose measurement.

- Wipe off excess blood on the skin and continue to fill the remaining two circles on the filter paper card with the seventh and eighth blood drops (Fig 8.7).



- After the blood drop collection, wipe any remaining blood from the prick site with a sterile gauze pad. Press the gauze pad against the prick site until the blood flow has stopped completely.

- Take an adhesive bandage from its wrapper and apply it to the prick site.

8. Record the **haemoglobin results** in **Question 368/467**. If a haemoglobin measurement was not obtained because the respondent did not consent to the test, or there was a technical problem with the testing, circle the appropriate code.

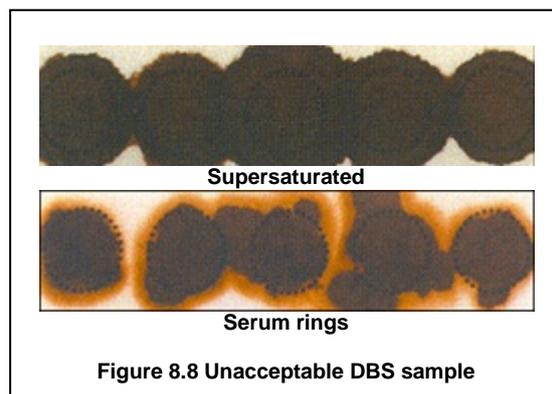
9. Record the **blood glucose results** in **Question 370/469**. If a blood glucose measurement was not obtained because the respondent did not consent to the test, or there was a technical problem with the testing, circle the appropriate code.
10. Place the filter paper card with blood spots in the drying box:
  - The drying box should always be placed vertically on a flat surface before opening. After opening it, carefully pick up the filter paper card with the wet blood spots and place the card in a horizontal position in one of the slots in the drying rack affixed to the inside of the box. The blood spots should face towards the back of the drying box.
  - The placement of the box is especially important for filter paper cards that have not dried completely. Keep the box in a vertical position so that the cards do not fall out of their slots. To prevent blood that has not dried completely from spreading, place the box vertically.
  - ***Avoid touching or smearing the blood spots on other cards in the box when you are storing a new card. Never put more than one filter paper card in the same slot in the drying rack.*** Allow the blood spots to dry overnight at ambient temperature. ***Never set any materials on top of the open box as they might contaminate the filter paper cards stored inside.***
  - If the collection process in a household is interrupted for any reason, you should close the box to prevent any possible contamination.
  - After all DBS samples have been collected from all eligible respondents in the household, carefully close the box. The drying box should be kept in a position so that the filter paper cards remain in their slots inside the box.
11. **Collect biohazardous waste**
  - Place all biohazardous waste (lancets, alcohol swabs, gauzes, and gloves) into a clearly labelled “biohazard” plastic bag, which has been provided for field disposal of these items. Before leaving the enumeration area (cluster) the bags with biohazardous waste must be taken to a health facility in the enumeration area and incinerated.
12. Record the respondent’s haemoglobin and blood glucose results in the appropriate informational brochures.
13. Inform the respondent of her/his haemoglobin and blood glucose results and provide the anaemia and blood glucose brochures.

14. Provide a written referral to a health facility for treatment for any respondents with severe anaemia (below 9 g/dl for men and pregnant women and below 7 g/dl for women who are not pregnant or don't know if they are pregnant).
15. Provide a written referral to a health facility for medical evaluation for any respondents with random blood glucose level  $\geq 200\text{mg/dl}$ .
16. Provide an informational brochure on HIV/AIDS. The brochure will include a voucher for free HIV testing, and a list of the nearest participating ICTC centres.

#### PRECAUTIONS TO TAKE DURING DBS COLLECTION FOR HIV TESTING

With respect to DBS collection for HIV testing, there are a number of precautions that should be strictly followed:

- **Always use the pre-printed side of the card to collect the blood spots.**
- **Do not 'layer' the sample in an attempt to fill in the circle.**
  - There may be times when a drop of blood will not completely fill the circle. If a circle is not completely saturated, the next drop or just a portion of the next drop of blood may be used to saturate the circle if the drop is obtained immediately. If the first drop starts to dry due to any interruption in getting the subsequent drop, you must begin filling another circle. Layering or application of successive drops of blood to a dried or partially dried blood spot causes caking.
- **Do not overfill the circles.** Overfilling the circles can cause super saturation (top panel of Figure 8.8), which is unacceptable.
- **Do not place the drying box horizontally until blood dries.** Placing the box horizontally before the blood dries can cause serum rings (bottom panel of Figure 8.8), which is also unacceptable. The blood should not extend beyond the pre-printed area, as shown in Figure 8.8.
- Try to have the blood drop fall exactly in the centre of the pre-printed circle. However, if by accident the drop falls outside of the circle and is not large enough, then let the next drop of blood fall again exactly in the centre of the original drop and not in the centre of the pre-printed circle. Note: all circles should be uniformly filled.
- **Protect the filter card from contamination.** Do not allow water or other contaminants to come into contact with the filter paper card before or after collecting the blood samples.



- **Do not place the specimens in the small zip-loc bags until the blood has dried thoroughly** (chocolate brown). Insufficient drying adversely affects the quality of the samples and consequently, the test results.
- **Taking the filter paper card out of the storage bag.** The filter paper card should be the last item taken out of the package before starting the blood collection procedure.

## STORING AND TRANSFERRING THE DRIED BLOOD SPOTS

The dried blood spot (DBS) samples must be properly maintained until they are picked up and taken to the laboratory. They should never be exposed to direct sunlight, stored in a humid environment, or exposed to dust, dirt or other environmental contaminants. During storage, it is important to regularly monitor the level of humidity in the stored samples by observing a change in colour of the desiccant granules or the humidity indicating card. The following describes the steps that should be followed in storing the DBS samples in the field and their subsequent and transfer to the laboratories.

### 1. Storage:

Each morning, before you go to the field, you must remove the filter paper cards with the DBS samples that you collected on the previous day from the drying box and prepare them for storage as follows:

1. Put on a pair of gloves and carefully open the drying box. Check that the blood spots on each filter paper are completely dried (chocolate brown).
2. Remove each filter paper card on which the spots have dried from the drying box separately. Be careful not to touch the blood spots.
3. Gently fold a piece of glassine paper over the blood spots and put one filter paper card into a small (low gas-permeable) zip-loc bag. Put one desiccant packet and a humidity indicator card behind the filter paper card, with the circles on the humidity card and the 'window' in the desiccant sachet facing out so that the circles on the humidity indicator card and granules, respectively, are visible. It is important that the desiccant packet and the humidity indicator card do not touch the blood spots. Close the zipper, gently pushing out any excess air in the bag as you are zipping it, being careful not to press on the blood spots. **DBS samples (the blood spots) should not be allowed to come in direct contact with other DBS samples during handling, shipment or storage.**
4. Continue to package each of the filter paper cards from the previous day which have dried overnight, putting one filter paper card into one small zip-loc bag with one desiccant packet and one humidity indicator card.

- When you have packed all of the filter paper cards, put them into the large zip-loc bag that has been labelled for the cluster in which the samples were collected (Figure 8.9). Note that the Cluster Sample (zip-loc) Bag itself should also contain a few desiccant packets.

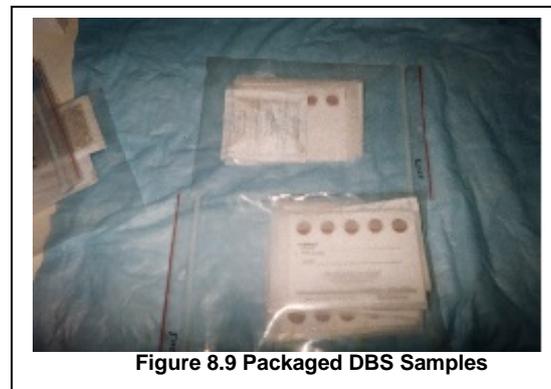


Figure 8.9 Packaged DBS Samples

5. Check the humidity indicator cards for the individually packaged DBS samples that you have previously placed in the Cluster bag before adding newly packaged DBS samples to an existing Cluster Sample Bag. **The build-up of humidity can damage the quality of the sample.**
  - A bottom circle that is pink (30% humidity) indicates a warning of increasing humidity. If the middle circle (40% humidity) is pink, gently open the small zip-loc bag, remove the desiccant packet and replace it with a fresh desiccant packet. Replace the humidity indicator card as well. If the top circle (50% humidity) is pink, examine the DBS cards as this indicates a high build-up of moisture which can degrade the DBS sample. Further, if any of the circles on the humidity indicator card have merged together so that they are not completely separated, remove the indicator card and replace it with a fresh indicator card. Close the zipper, gently pushing out any excess air in the bag as you are zipping it. **Please review the storage instructions, using the humidity card as a guide.**
6. Check the condition of the desiccant packets and humidity indicator card before closing the zipper on the Cluster Sample Bag. Replace as needed.
  - If you have additional Cluster Sample Bags for completed clusters that have not yet been collected by a Field Supervisor, examine all of the samples in those bags in the same manner, every couple of days, as long as they are with your team in the field.

## 2. Transfer:

The purpose of the DBS Transmittal Sheet (see Appendix 9) is to account for the samples that have been collected in each Cluster.

1. Fold the DBS Transmittal Sheet along the dotted lines (so that the bar-code labels are not folded), and keep it in the Sample Cluster Bag along with the DBS samples for that cluster.
2. When you have completed the cluster, remove the packaged DBS samples from the Cluster Sample Bag (do not open the small zip-loc bags).
3. One by one, check the bar codes on the labels on the filter paper cards against the bar codes affixed to the back side of the DBS Transmittal Sheet. For each DBS sample, put a check mark in the column labelled health investigator for each corresponding bar code found on the

transmittal sheet. Count the number of DBS samples and record in the boxes provided in Column (3) on the front side of the transmittal sheet in the column labelled TOTAL COUNT OF BLOOD SAMPLES. If there are any discrepancies, you must attempt to account for them. Use Column (7) to explain. Sign your name in Column (4) and the date in Column (6).

The team's field supervisor will follow behind you, re-verify the samples, and sign his/her name in the FIELD TEAM SUPERVISOR row.

Periodically, a sample pick up person will visit the teams to collect the DBS samples for the completed clusters. When he/she collects the DBS samples, he/she will recount the DBS samples for each of the completed clusters and sign the DBS Transmittal Sheet. The samples and transmittal sheet will be transported to designated laboratories for logging in and storage.

### **Summary of the steps involved in collecting blood for HIV testing, haemoglobin and random blood glucose testing for adults:**

- Check Biomarker Questionnaire for individuals eligible for testing;
  - Seek voluntary consent for haemoglobin and blood glucose testing and blood collection for HIV testing from the respondent (if the respondent is age 15 to 17 and unmarried seek voluntary consent from the parent/responsible adult and the respondent).
- Place the bar code labels in the Biomarker Questionnaire, on a filter paper card and the DBS Transmittal Sheet;
- Clean and prick the respondent's finger with an adult lancet;
- Wipe away the first drop of blood;
- Fill the pre-printed circles on the filter paper card with the second, third, and fourth drops of blood – you will fill 3 circles on the filter paper card;
- Wipe away any excess blood on the skin;
- Collect the fifth blood drop in a microcuvette;
- Collect the sixth blood drop on the glucose strip;
- Collect the seventh and eighth blood drops on the filter paper, if possible;
- Test the blood sample for haemoglobin with the HemoCue photometer;
- Test the blood sample for blood glucose in the glucometer;
- Stop the bleeding at the prick site;
- Record the haemoglobin level on the Biomarker Questionnaire
- Place the filter paper card in the drying box;
- Collect biohazardous waste.
- Inform the respondent of his/her haemoglobin and glucose results and provide an informational brochures on anaemia and blood glucose ;
- Provide a written referral for follow-up medical attention for respondents found to be severely anaemic or with random blood glucose levels  $\geq 200\text{mg/dl}$ ;
- Provide all respondents with an informational brochure on HIV/AIDS, vouchers for free HIV testing, and a list of nearby ICTC centres.

## CHAPTER 9: BIOHAZARDOUS WASTE DISPOSAL

Any material coming in contact with blood or serum (lancets, microcuvettes, glucose strips, alcohol swabs, gauze, and gloves) is considered to be biohazardous (hazardous to other humans). Safe disposal of such material is very important to prevent the transmission and spread of various blood borne diseases, such as Hepatitis B and HIV, among survey personnel and within the study community. Biohazardous waste has to be collected in a special container during the blood collection and testing, securely stored and transported, and safely disposed at the end of each day of fieldwork.

If possible, commercially available biohazardous waste disposal containers should be used for waste disposal. These types of containers are red and have a special logo warning about biohazardous content. They can be securely closed for safe storage and transportation during the fieldwork.

There are two options for disposal:

1. Take the biohazardous waste to the nearest health facility for disposal in an incinerator (**preferred option**). The health facilities should employ standard procedures for biohazardous waste disposal.
2. Follow the procedures outlined below for burning the waste in the field.

### MATERIALS AND SUPPLIES

The following items are required in the field for disposal of biohazardous materials:

- Kerosene
- Four percent sodium hypochlorite solution (4% Bleach: Disinfectant)
- Matches
- Spade or other tool for digging a small pit
- Ziploc-type polyethylene bags
- Forceps
- Puncture-resistant container labelled "Biohazard" (for example, a wide-mouth plastic jar).
- Scissors

### PROCEDURES FOR FIELD DISPOSAL OF BIOHAZARDOUS WASTE

At the end of each blood collection and testing within the household, all materials used during the testing (gloves, microcuvettes, glucose strips, lancets, alcohol swabs, and gauze pads) are to be placed in a biohazard bag (plastic). At the end of each day's work or at least every two days, the biohazardous waste should be disposed of following the procedure below.

Before beginning the biohazardous waste disposal procedure, determine a place where the waste can be safely destroyed. An open field area with loose soil is preferable, since the materials need to be burnt and buried. To reduce the risk of spreading a fire, avoid starting a fire in drought areas, and keep away from other flammable materials.

**Follow the procedure below to safely dispose of biohazardous waste in the field:**

**Step 1:** At the end of each day, bring the sharps container (plastic jar) with biohazardous materials to the area selected for the waste disposal. Wearing gloves, add a half litre of 4 percent sodium hypochlorite solution (disinfectant) into the sharps container (plastic jar) with the biohazardous materials (see figure 9.1). After adding the hypochlorite solution, close the container (jar) so it is airtight. Keep the jar in an upright position for five minutes. After that, invert the plastic jar and keep in that position for an additional five minutes. This step is necessary to ensure that all of the materials in the sharps container (plastic jar) are disinfected by complete immersion in the 4 percent sodium hypochlorite solution.



**Figure 9.1 Adding sodium hypochlorite**



**Figure 9.2 Transferring contaminated materials**

**Step 3:** A forceps can be used to transfer any material that sticks to the walls of the plastic jar to the polyethylene bag (Figure 9.3).



**Figure 9.3 Removing remaining materials**



**Figure 9.4 Digging a pit for bag**

**Step 4:** Dig a small hole with a spade, and put the polyethylene bag containing the biohazardous materials in the pit (Figure 9.4).

**Step 5:** Use scissors to make a hole at the bottom of the polyethylene bag (Figure 9.5).



**Figure 9.5** Making a hole in the bag



**Figure 9.6** Draining off hypochlorite solution

**Step 6:** Drain off the hypochlorite solution from the polyethylene bag (Figure 9.6).

**Step 7:** Put waste paper on top of the polyethylene bag containing biohazardous materials (Figure 9.7).



**Figure 9.7** Putting waste paper on top of bag



**Figure 9.8** Pouring kerosene on bag

**Step 8:** Pour kerosene on the bag (Figure 9.8).

**Step 9:** Burn the polyethylene bag containing the biohazardous materials in the pit (Figure 9.9).



**Figure 9.9** Burning contaminated materials



**Figure 9.10** Ascertaining that contaminated materials are completely burned

**Step 10:** Wait until all of the contents are burned (Figure 9.10).

**Step 11:** Cover the pit with soil (Figure 9.11).



**Figure 9.11** Covering the pit with soil

It is the responsibility of the team's field supervisor to ensure proper disposal of biohazardous waste by the health investigators. It is unacceptable that the materials used during the testing in one fieldwork cluster are carried by the team to the next cluster. Biohazardous materials must be destroyed at the end of the day.

## **UNIVERSAL PRECAUTIONS IN THE COLLECTION OF BLOOD SAMPLES**

Universal precautions, as defined by the U.S. Centers for Disease Control and Prevention (CDC), are a set of precautions designed to prevent transmission of the human immunodeficiency virus (HIV), the Hepatitis B virus (HBV), and other blood borne pathogens when providing first aid or health care.

Below are excerpts from two documents on universal precautions: 1) CDC's "Perspectives in Disease Prevention and Health Promotion Update: Universal Precautions for Prevention of Transmission of Human Immunodeficiency Virus, Hepatitis B Virus, and Other Blood borne Pathogens in Health-Care Settings,"<sup>6</sup> and 2) OSHA's "Occupational Exposure to Blood borne Pathogens—Precautions for Emergency Responders."<sup>7</sup>

### **UNIVERSAL BLOOD PRECAUTIONS (U. S. Centers for Disease Control and Prevention (CDC))**

In 1983, CDC published a document entitled Guideline for Isolation Precautions in Hospitals (1) that contained a section entitled Blood and Body Fluid Precautions. The recommendations in this section called for blood and body fluid precautions when a patient was known or suspected to be infected with blood borne pathogens. In August 1987, CDC published a document entitled Recommendations for Prevention of HIV Transmission in Health-Care Settings (2). In contrast to the 1983 document, the 1987 document recommended that blood and body fluid precautions be consistently used for all patients, regardless of their blood borne-infection status. This extension of blood and body fluid precautions to all patients is referred to as Universal Blood and Body Fluid Precautions or Universal Precautions. Under universal pre-cautions, blood and certain body fluids of all patients are considered potentially infectious for human immunodeficiency virus (HIV), Hepatitis B virus (HBV), and other blood borne pathogens.

Universal precautions are intended to prevent parenteral, mucous membrane, and non-intact skin exposures of health-care workers to blood borne pathogens. In addition, immunization with the HBV vaccine is recommended as an important adjunct to universal precautions for health-care workers who have exposure to blood.

Since the recommendations for universal precautions were published in August 1987, CDC and the US Food and Drug Administration (FDA) have received requests for clarification of the following issues: 1) body fluids to which universal precautions apply, 2) use of protective barriers, 3) use of gloves for phlebotomy, 4) selection of gloves for use while observing universal precautions, and 5) need for making changes in waste management programs as a result of adopting universal precautions.

Universal precautions apply to blood and to other body fluids containing visible blood. Occupational transmission of HIV and HBV to health-care workers by blood is documented. Blood is the single most abundant source of HIV, HBV, and other blood borne pathogens in the occupational setting. Infection-control efforts for HIV, HBV, and other blood borne pathogens must focus on preventing exposure to blood and delivering HBV immunization.

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<sup>6</sup> Morbidity and Mortality Weekly Report 1988; 37(24):377-388.

<sup>7</sup>U.S. Department of Labor, Occupational Safety and Health Administration. 1998 (Revised) OSHA 3106.

Protective barriers reduce the risk of exposure of the health-care worker's skin or mucous membranes to potentially infective materials. For universal precautions, protective barriers reduce the risk of exposure to blood, body fluids containing visible blood, and other fluids to which universal precautions apply. Examples of protective barriers include gloves, gowns, masks, and protective eyewear. Gloves should reduce the incidence of contamination of hands, but they cannot prevent penetrating injuries due to needles or other sharp instruments. Masks and protective eyewear or face shields should reduce the incidence of contamination of mucous membranes of the mouth, nose, and eyes.

Universal precautions are intended to supplement, rather than replace, recommendations for routine infection control, such as hand washing and using gloves to prevent gross microbial contamination of hands. Because specifying the types of barriers needed for every possible clinical situation is impractical, some judgment must be exercised.

The risk of nosocomial transmission of HIV, HBV, and other blood borne pathogens can be minimized if health-care workers use the following general guidelines:

1. Take care to prevent injuries when using needles, scalpels, and other sharp instruments or devices; when handling sharp instruments after procedures; when cleaning used instruments; and when disposing of used needles. Do not recap used needles by hand; do not remove used needles from disposable syringes by hand; and do not bend, break, or otherwise manipulate used needles by hand. Place used disposable syringes and needles, scalpel blades, and other sharp items in puncture-resistant containers for disposal. Place puncture-resistant containers as close to the use area as is practical.
2. Use protective barriers to prevent exposure to blood, body fluids containing visible blood, and other fluids to which universal precautions apply. The type of protective barrier(s) should be appropriate for the procedure being performed and the type of exposure anticipated.
3. Wash hands and other skin surfaces that are contaminated with blood or other body fluids immediately and thoroughly.

Gloves should reduce the incidence of blood contamination of hands during phlebotomy (drawing blood samples), but they cannot prevent penetrating injuries caused by needles or other sharp instruments. The likelihood of hand contamination with blood containing HIV, HBV, or other blood borne pathogens during phlebotomy depends on several factors: 1) the skill and technique of the health-care worker, 2) the frequency with which the health-care worker performs the procedure (other factors being equal, the cumulative risk of blood exposure is higher for a health-care worker who performs more procedures), 3) whether the procedure occurs in a routine or emergency situation (where blood contact may be more likely), and 4) the prevalence of infection with blood borne pathogens in the patient population. The likelihood of infection after skin exposure to blood containing HIV or HBV will depend on the concentration of virus (viral concentration is much higher for Hepatitis B than for HIV), the duration of contact, the presence of skin lesions on the hands of the health-care worker and, for HBV, the immune status of the health-care worker. Although not accurately quantified, the risk of HIV infection following intact-skin contact with infective blood is certainly much less than the 0.5% risk following percutaneous needle stick exposures (5). In universal

precautions, all blood is assumed to be potentially infective for blood borne pathogens, but in certain settings (e.g., volunteer blood-donation centres) the prevalence of infection with some blood borne pathogens (e.g., HIV, HBV) is known to be very low. Some institutions have relaxed recommendations for using gloves for phlebotomy procedures by skilled phlebotomists in settings where the prevalence of blood borne pathogens is known to be very low.

Institutions that judge that routine gloving for all phlebotomies is not necessary should periodically reevaluate their policy. Gloves should always be available to health-care workers who wish to use them for phlebotomy. In addition, the following general guidelines apply:

1. Use gloves for performing phlebotomy when the health-care worker has cuts, scratches, or other breaks in his or her skin.
2. Use gloves in situations where the health-care worker judges that hand contamination with blood may occur; for example, when performing phlebotomy on an uncooperative patient.
3. Use gloves for performing finger or heel sticks on infants and children.
4. Use gloves when persons are receiving training in phlebotomy.

*Selection of Gloves* . The Center for Devices and Radiological Health, FDA, has the responsibility of regulating the medical-glove industry. Medical gloves include those marketed as sterile surgical gloves or non-sterile examination gloves made of vinyl or latex. General-purpose utility ("rubber") gloves are also used in the health-care setting, but they are not regulated by the FDA since they are not promoted for medical use. There are no reported differences in barrier effectiveness between intact latex and intact vinyl, which are used to manufacture gloves. Thus, the type of gloves selected should be appropriate for the task being performed. The following general guidelines are recommended:

1. Use sterile gloves for procedures involving contact with normally sterile areas of the body.
2. Use examination gloves for procedures involving contact with mucous membranes, unless otherwise indicated, and for other patient care or diagnostic procedures that do not require the use of sterile gloves.
3. Change gloves between patient contacts.
4. Do not wash or disinfect surgical or examination gloves for reuse. Washing with surfactants may cause wicking, i.e., the enhanced penetration of liquids through undetected holes in the gloves. Disinfecting agents may cause deterioration.
5. Use general-purpose utility gloves (e.g., rubber household gloves) for house-keeping chores involving potential blood contact and for instrument cleaning and decontamination procedures. Utility gloves may be decontaminated and reused but should be discarded if they are peeling, cracked, or discoloured, or if they have punctures, tears, or other evidence of deterioration.

## References

- Garner, J.S. and B.P. Simmons. Guideline for isolation precautions in hospitals. *Infect Control* 1983; 4:245-325.
- Centers for Disease Control. Recommendations for prevention of HIV transmission in health-care settings. *MMWR* 1987; 36 (suppl no. 2S).
- Immunization Practices Advisory Committee. Recommendations for protection against viral hepatitis. *MMWR* 1985; 34:313-24,329-35.
- Department of Labor, Department of Health and Human Services. Joint advisory notice: Protection against occupational exposure to hepatitis B virus (HBV) and human immunodeficiency virus (HIV). Washington, D.C: U.S. Department of Labor, U.S. Department of Health and Human Services, 1987.
- Centers for Disease Control. Update: Acquired immunodeficiency syndrome and human immunodeficiency virus infection among health-care workers. *MMWR* 1988; 37:229-34,239.
- Garner, J.S. and M.S. Favero. Guideline for hand washing and hospital environmental control, 1985. Atlanta: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control, 1985; HHS publication no. 99-1117.

## STANDARD PRECAUTIONS AGAINST BLOODBORNE PATHOGENS (U. S. Occupational Safety and Health Administration (OSHA))

Universal precautions are designed to protect health-care workers and patients from exposure to bloodborne pathogens and other potentially infectious body substances. They are mandated by the United States Occupational Safety and Health Administration (OSHA) and include the following.

1. Wear personal protective equipment (aprons, gowns, gloves, goggles, face shields, masks, and CPR devices) when exposure to blood, blood droplets, and other body fluids is anticipated; these precautions are always mandated during invasive procedures.
2. Wear gloves when doing patient care if skin is cut, abraded, or chapped; when collecting or handling specimens or body fluids; cleaning specimen containers; or decontaminating. If you anticipate contact with mucous membranes, non-intact skin, GI or GU tract, or active bleeding wounds; or anticipate invasive procedures such as venipuncture or vascular access procedures, then universal precautions must be observed.
3. Gowns, aprons, scrubs, or lab coats must cover exposed skin areas when there is a potential for splashing blood or body fluids on clothing; however, this protection is not required for routine-care situations in which blood or body substances are not likely to be present. Perform all procedures in such a way such that splashing, spattering, or droplet formation is minimized.
4. Prevent injuries that can be caused by needles, scalpels, and other “sharps.” Dispose of all these in puncture-resistant containers. Do not recap, bend, break by hand, or remove needles from disposable syringes. Tape “piggyback” needle devices in place to prevent accidental dislodging.
5. Remove torn or punctured gloves promptly. Wash hands and other skin surfaces immediately and thoroughly if contaminated with blood or other body fluids.
6. Eating, drinking, applying cosmetics or lip balm, and handling contact lenses are not permitted in work areas where there is a reasonable likelihood of exposure to blood or other body substances.
7. Health-care workers should protect and always take care of themselves first. Presume that all patients have hepatitis B or HIV. In cases of suspected HIV or hepatitis B (HBV) exposure, identify, obtain consent, and test for exposure if the patient consents to testing. If the patient refuses consent or outcome is positive, the health-care worker must receive HIV-antibody testing immediately. Advise HIV-negative person who has been exposed to seek medical evaluation of any acute febrile illness within 12 weeks of exposure to HIV and to retest in 6 to 12 weeks and 6 months after exposure. Some institutions offer prophylactic drug therapy or hepatitis B vaccinations to their employees. If exposed or injured, the healthcare worker must make the decision to accept drug therapy within a few hours of the incident.

8. The following are definitions of infection-control terminology:

**Bloodborne pathogens:** Organisms that can be transmitted from one person to another by exposure to the infected person's blood. The major pathogens include hepatitis B virus, hepatitis C virus, human immunodeficiency virus (HIV; the AIDS virus), and treponemesthat cause syphilis.

**Body substances:** Any fluids or solids that come out of or off of the human body. Examples include saliva, sputum, urine, faeces, wound drainage, and all the fluids referred to as "other potentially infectious materials" (see later).

**Exposure incident:** The contact of blood or other body substances with an employee's mucous membranes (eyes, mouth), non-intact skin (skin with cuts, abrasions, dermatitis, or other); or contact by piercing or puncturing mucous membranes or skin with a contaminated item.

**Regulated (infectious) waste:**Items caked or saturated with blood, or other potentially infectious materials; contaminated sharps; pathologic and microbiologic waste.

**Other potentially infectious materials (OPIM):** Body substances specifically designated by the CDC and OSHA that may transmit bloodborne pathogens include semen, vaginal secretions, cerebrospinal fluid, synovial fluid, pleural fluid, amniotic fluid, saliva in dental procedures, or any body substance that is visibly contaminated with blood, and all body substances in situations for which it is difficult or impossible to determine whether blood is present.

## APPENDIX1: BIOMARKER TESTING CONSENT STATEMENTS

### CONSENT FOR BLOOD PRESSURE MEASUREMENT

I would like to measure your blood pressure. This will be done three times, with an interval of about five minutes between measurements. This is a harmless procedure. It is used to find out if a person has high blood pressure. If it is not treated, high blood pressure may eventually cause serious damage to the heart. The results of this blood pressure measurement will be given to you after the measurement process is completed. I will explain the meaning of your blood pressure numbers. If your blood pressure is high, we will suggest that you consult a health facility or doctor since we cannot provide any further testing or treatment during the survey. You can also decide at any time not to participate in the blood pressure measurement. The result will be kept strictly confidential and will not be shared with anyone other than members of our survey team.

Do you have any questions?

You can say yes to the test or you can say no. It is up to you to decide.

**Will you allow me to measure your blood pressure?**

### CONSENT FOR ANAEMIA MEASUREMENT

As part of this survey, we are asking people all over the country to take an anaemia test. Anaemia is a serious health problem that usually results from poor nutrition, infection, or chronic disease. This survey will assist the government to develop programs to prevent and treat anaemia.

For the anaemia testing, we will need a few drops of blood from a finger. The equipment used to take the blood is clean and completely safe. It has never been used before and will be thrown away after each test. The blood will be tested for anaemia immediately, and the result will be told to you right away. The result will be kept strictly confidential and will not be shared with anyone other than members of our survey team.

Do you have any questions?

You can say yes to the test, or you can say no. It is up to you to decide.

**Will you take the anaemia test?**

### **CONSENT FOR RANDOM BLOOD GLUCOSE TESTING**

As part of this survey, we are also measuring the level of sugar in the blood. If it is not treated, a high level of blood sugar may increase the risk for heart disease and stroke. For the blood glucose testing, we will need a few drops of blood from a finger. The equipment used to take the blood is clean and completely safe. It has never been used before and will be thrown away after each test.

The blood will be tested for glucose immediately, and the result will be told to you right away. The result will be kept strictly confidential and will not be shared with anyone other than members of our survey team. The results of this blood glucose test will be given to you with an explanation of the meaning of your blood glucose numbers. If your blood glucose is high, we will suggest that you consult a health facility or doctor since we cannot provide any counselling, further testing or treatment during the survey.

Do you have any questions about the blood glucose measurement so far? If you have any questions about the procedure at any time, please ask me.

You can say yes or no to having your blood glucose measured now.

**Will you allow me to proceed to take your measurement?**

### **CONSENT FOR DBS COLLECTION**

As part of the survey we also are asking people all over the country to take an HIV test. HIV is the virus that causes AIDS. AIDS is a very serious illness. The HIV test is being done to see how big the AIDS problem is in India.

For the HIV test, we need a few (more) drops of blood from a finger. The equipment used to take the blood is clean and completely safe. It has never been used before and will be thrown away after each test. No names will be attached so we will not be able to tell you the test results. No one else will be able to know your test results either. If you want to know whether you have HIV, I can provide you with a list of nearby facilities offering counselling and testing for HIV. I will also give you a voucher for free services for you (and for your partner if you want) that you can use at any of these facilities.

Do you have any questions?

You can say yes to the test, or you can say no. It is up to you to decide.

**Will you take the HIV test?**

### **CONSENT FOR ADDITIONAL TESTING**

APPENDIX 2: NFHS-4 BLOOD PRESSURE REFERRAL FORM

**National Family Health Survey (NFHS-4): Blood Pressure Referral Form**

**(Blood Pressure Referral Form to be given when adult's systolic pressure is greater than 200mm Hg and/or diastolic pressure is greater than 109 mm Hg)**

During NFHS-4 \_\_\_\_\_ (NAME)'s blood pressure was measured on \_\_\_ / \_\_\_ / \_\_\_\_.  
His/her systolic pressure was \_\_\_\_ and his/her diastolic pressure was \_\_\_\_ mg/dL, which is severely elevated.

Date \_\_\_\_\_ Signature \_\_\_\_\_

-----CUT HERE-----

**National Family Health Survey (NFHS-4): Blood Pressure Referral Form**

**(Blood Pressure Referral Form to be given when adult's systolic pressure is greater than 200mm Hg and/or diastolic pressure is greater than 109 mm Hg)**

During NFHS-4 \_\_\_\_\_ (NAME)'s blood pressure was measured on \_\_\_ / \_\_\_ / \_\_\_\_.  
His/her systolic pressure was \_\_\_\_ and his/her diastolic pressure was \_\_\_\_ mg/dL, which is severely elevated.

Date \_\_\_\_\_ Signature \_\_\_\_\_

**National Family Health Survey (NFHS-4): Anaemia Referral Form**

(Anaemia Referral Form to be given when adult's haemoglobin level is less than 7.0 g/dL for women and children or less than 9.0 g/dL for men and pregnant women)

During NFHS-4 \_\_\_\_\_ (NAME), was tested for anaemia on \_\_\_\_ / \_\_\_\_ / \_\_\_\_.

His/her level of haemoglobin was \_\_\_\_ . \_\_\_\_ g/dL, which indicates he/she has severe anaemia.

**THIS PERSON NEEDS MEDICAL ATTENTION FOR THE ANAEMIA IN A HEALTH FACILITY RIGHT AWAY.**

Date \_\_\_\_\_ Signature \_\_\_\_\_

-----CUT HERE-----

**National Family Health Survey (NFHS-4): Anaemia Referral Form**

(Anaemia Referral Form to be given when adult's haemoglobin level is less than 7.0 g/dL for women and children or less than 9.0 g/dL for men and pregnant women)

During NFHS-4 \_\_\_\_\_ (NAME), was tested for anaemia on \_\_\_\_ / \_\_\_\_ / \_\_\_\_.

His/her level of haemoglobin was \_\_\_\_ . \_\_\_\_ g/dL, which indicates he/she has severe anaemia.

**THIS PERSON NEEDS MEDICAL ATTENTION FOR THE ANAEMIA IN A HEALTH FACILITY RIGHT AWAY.**

Date \_\_\_\_\_ Signature \_\_\_\_\_

**National Family Health Survey (NFHS-4): Blood Glucose Referral Form**

**(Blood Glucose Referral Form to be given when adult's random glucose level is greater than 200mg/dL)**

During NFHS-4 \_\_\_\_\_(NAME)'s random blood glucose level was tested on \_\_\_ / \_\_\_ / \_\_\_.

His/her random blood glucose level was \_\_\_ mg/dL, which is abnormally high.

Date \_\_\_\_\_ Signature \_\_\_\_\_

-----CUT HERE-----

**National Family Health Survey (NFHS-4): Blood Glucose Referral Form**

**(Blood Glucose Referral Form to be given when adult's random glucose level is greater than 200mg/dL)**

During NFHS-4 \_\_\_\_\_(NAME)'s random blood glucose level was tested on \_\_\_ / \_\_\_ / \_\_\_.

His/her random blood glucose level was \_\_\_ mg/dL, which is abnormally high.

Date \_\_\_\_\_ Signature \_\_\_\_\_

(*LETTER NUMBER*)

Date: \_\_\_\_\_

To

**The Incharge**

Integrated Counselling and Testing Centre (ICTC), *Address*

**Subject: Referral Letter for Voluntary Counselling and Testing of HIV**

Dear **Sir/Madam**,

Presently the fourth National Family Health Survey (NFHS-4) is being conducted under the stewardship of the Ministry of Health and Family Welfare (MOHFW). The International Institute for Population Sciences (IIPS), Mumbai, is designated as nodal agency for coordinating the NFHS-4 project and (*ORGANIZATION*) is conducting the fieldwork for NFHS-4 in (*STATE*).

As a part of this survey, health investigators visit sampled households in selected villages, towns and cities throughout India. During the survey, women and men in randomly selected household are asked to give a capillary blood sample from a fingerstick for HIV testing in one of five reputed laboratories. The HIV prevalence results from the tests will be used by the National AIDS Control Organisation (NACO) and the Ministry of Health and Family Welfare for further refining programme strategies.

The HIV testing in NFHS-4 is anonymous, i.e., no individual identifiers are associated with the blood samples or test results and, therefore, respondents will not be provided with information about their own HIV status. However, we are giving this referral letter to respondents to receive an HIV test at a health facility, so that they will have an opportunity to receive voluntary counselling and HIV testing at a health facility without charge if they desire.

**The bearer of this letter Ms/Mr -----was eligible for anonymous HIV testing in NFHS-4 and hence may please be provided with necessary services for voluntary counselling and testing for HIV free of charge.**

**If you have any questions about the NFHS-4 survey, you may contact:**

**(ADD CONTACT INFORMATION FOR FIELD AGENCY CONDUCTING NFHS-4)**

Thanking you and with regards,

Yours sincerely,

**(In-charge of Organization)**  
(Organization)

## APPENDIX 6: NFHS-4 BLOOD PRESSURE AND DIABETES BROCHURE FOR ADULTS

### What IS Diabetes?

Diabetes is a set of metabolic disorders characterized by **elevated blood glucose** levels due to defective insulin secretion and/or ineffective insulin action.

### What is MEASURED?

The level of glucose in blood is measured (usually on more than one occasion) to determine if the person has diabetes. The measurement is usually done on a **fasting blood sample**, about 8 to 12 hours after eating, or on a **random blood sample**.

### What CAUSES diabetes?

In most cases the cause of diabetes is not known. Some of the possible causes are:

- ◇ Poor diet
- ◇ Obesity
- ◇ Viral infection
- ◇ Age
- ◇ Stress
- ◇ Smoking
- ◇ Family history
- ◇ Genetics
- ◇ Lack of physical activity

### TEST RESULTS

Date \_\_\_\_\_

Name: _____			
<b>BLOOD GLUCOSE (Random)</b>			
_____ mg/dL			
<b>Blood glucose category</b>			
	<i>Normal</i>	<i>Abnormal</i>	<i>Referral</i>
Random Blood glucose	< 200 mg/dL	≥ 201 mg/dL	No Yes

BLOOD PRESSURE				
SYSTOLIC		DIASTOLIC		
BLOOD PRESSURE CATEGORY	Systolic (mm Hg)	And	Diastolic (mm Hg)	Referral: Recheck within
Normal (Optimal)	<120	And	<80	1 Year
Normal (Mildly high)	<130	And	<85	1 Year
Normal (Moderately high)	130 - 139	And/Or	85 - 89	2 Months
Abnormal (Mildly elevated)	140 - 159	And/Or	90 - 99	1 Month
Abnormal (Moderately elevated)	160 - 179	And/Or	100 - 109	1 Week
Abnormal (Severely elevated)	≥180	And/Or	≥110	Immediately

Note!!! Your blood pressure was measured by a trained examiner. These measurements were obtained as part of a survey and do not represent a medical diagnosis. Interpretation of these measurements must be made by a physician.

### What IS Blood Pressure?

Arterial blood pressure is the force exerted by the blood on the walls of the arteries as the heart pumps (contracts) and relaxes.

Elevated high blood pressure is dangerous because it can lead to serious health problems such as kidney failure, and death from coronary heart disease and stroke.

### What is MEASURED?

The **systolic pressure** is the maximum pressure in an artery at the moment when the heart is beating and pumping blood through the body.

The **diastolic pressure** is the lowest pressure in an artery at the moments between beats when the heart is resting.

A person is considered to have high blood pressure (hypertension) if either one of the two pressures is above normal.

### What CAUSES high Blood Pressure?

The exact causes of high blood pressure are unknown, but several factors may play a role in its development including:

- ◇ Smoking
- ◇ Overweight / Obesity
- ◇ Lack of physical activity
- ◇ Family history
- ◇ Genetics
- ◇ High salt and/ or alcohol intake
- ◇ Older age
- ◇ Stress

### MEASURES OF PREVENTION.....

#### Can Diabetes be PREVENTED?

Diabetes can be prevented by:

- ◇ Reducing carbohydrates in the diet
- ◇ Regular exercise

#### Can high Blood Pressure be PREVENTED?

High blood pressure can be prevented by:

- ◇ Reducing weight
- ◇ Increasing physical activity
- ◇ Increasing consumption of fruits and vegetables
- ◇ Reducing salt and alcohol consumption
- ◇ Stopping smoking

## Implementing Agency

[CONTACT INFORMATION: ADDRESS]

## National Family Health Survey NFHS-4

Name \_\_\_\_\_ Date \_\_\_\_\_

[IMPLEMENTING AGENCY] is conducting the National Family Health Survey in which testing of blood glucose and measuring of blood pressure are included. The study will help us identify whether there are problems with diabetes and hypertension among women and men in India.

We appreciate you allowing us to interview and test you.

Thank you for your cooperation.

*Please look inside for the results of your blood glucose and blood pressure tests.*

APPENDIX 7: NFHS-4 ANAEMIA BROCHURE FOR WOMEN AND CHILDREN

**What IS Anaemia?**

Anaemia is a serious health condition in which there are not enough red blood cells or haemoglobin in the blood.

Haemoglobin is a substance in the blood that carries oxygen to the brain, muscles, disease-fighting organs and other parts of the body. Iron is important for making haemoglobin.

**What are the SYMPTOMS of Anaemia?**

Some of the symptoms of anaemia are:

- tiredness
- headaches
- dizziness
- poor appetite
- heart palpitations
- shortness of breath

**Why is Anaemia DANGEROUS?**

Anaemia is dangerous because it reduces one's resistance to infections.

- severe anaemia can lead to heart failure
- during childbirth, anaemic women are more likely to die from excessive bleeding
- anaemic children have low birth weight, poor learning capacity, and less resistance to infections than other children

**TEST RESULTS**

Date \_\_\_\_\_

Woman	Child 1	Child 2	Child 3
_____	_____	_____	_____
<b>Haemoglobin level (write) and Anaemia Classification (circle one)</b>			
_____ g/dL	_____ g/dL	_____ g/dL	_____ g/dL
<b>Severe</b> (less than 7.0 g/dL)	<b>Severe</b> (less than 7.0 g/dL)	<b>Severe</b> (less than 7.0 g/dL)	<b>Severe</b> (less than 7.0 g/dL)
<b>Moderate</b> (7.0-9.9 g/dL)	<b>Moderate</b> (7.0-9.9 g/dL)	<b>Moderate</b> (7.0-9.9 g/dL)	<b>Moderate</b> (7.0-9.9 g/dL)
<b>Mild</b> (not pregnant) (10.0-11.9 g/dL) (pregnant) (10.0-10.9 g/dL)	<b>Mild</b> (10.0-10.9 g/dL)	<b>Mild</b> (10.0-10.9 g/dL)	<b>Mild</b> (10.0-10.9 g/dL)
<b>Normal</b> (not pregnant) (12.0 g/dL or more) (pregnant) (11.0 g/dL or more)	<b>Normal</b> (11.0 g/dL or more)	<b>Normal</b> (11.0 g/dL or more)	<b>Normal</b> (11.0 g/dL or more)
<b>Height (cm)</b>			
_____	_____	_____	_____
<b>Weight (Kg)</b>			
_____	_____	_____	_____

**Severe Anaemia:** You have a seriously low level of haemoglobin in your blood. You need to see a doctor or visit a health centre immediately for treatment. Eat more foods rich in iron and treat malaria and worms immediately.

**Moderate Anaemia:** Your anaemia may be caused by iron deficiency, worms, excessive bleeding or malaria. You should visit a doctor or health centre as soon as possible. Eat more foods rich in iron.

**Mild Anaemia:** You need more daily iron. Treat malaria and worms immediately. Eat more foods rich in iron.

**Iron Rich Foods**



To increase the body's use of iron, eat more fruits and vegetables rich in vitamin C:



**What CAUSES Anaemia?**

Anaemia is caused by loss of blood mainly due to:

- parasites, especially hookworms
- excessive menstrual losses
- chronic diseases such as ulcers or tuberculosis
- a lack of iron in the diet
- inability of the body to absorb iron from food

**How can Anaemia be PREVENTED?**

- Eat a diet adequate in iron-rich foods such as dark green leafy vegetables, liver, meat, or fish, and fruits rich in vitamin C.
- Avoid giving tea to infants and young children.
- Avoid taking coffee or tea with meals.
- Prevent and treat worms.
- Prevent malaria by using insecticide-treated bednets.
- Pregnant mothers and infants should take iron tablets or syrup.
- Limit the number of births by spacing children and delaying the first pregnancy.

**Implementing Agency**

[CONTACT INFORMATION: ADDRESS]



**National Family Health Survey  
NFHS-4**

Name \_\_\_\_\_ Date \_\_\_\_\_

NFHS-4 will help us to find out the anaemia and nutritional status among men, women and young children in India.

We appreciate that you allowed us to interview you and to test you (and your child) for anaemia.

Thank you for your cooperation.

**Please look inside for the results of your anaemia test, and height and weight measurements.**

APPENDIX 8: NFHS-4 ANAEMIA BROCHURE FOR MEN AND CHILDREN

**What IS Anaemia?**

Anaemia is a serious health condition in which there are not enough red blood cells or haemoglobin in the blood.

Haemoglobin is a substance in the blood that carries oxygen to the brain, muscles, disease-fighting organs and other parts of the body. Iron is important for making haemoglobin.

**What are the SYMPTOMS of Anaemia?**

Some of the symptoms of anaemia are:

- tiredness
- headaches
- dizziness
- poor appetite
- heart palpitations
- shortness of breath

**Why is Anaemia DANGEROUS?**

Anaemia is dangerous because it reduces one's resistance to infections.

- severe anaemia can lead to heart failure
- during childbirth, anaemic women are more likely to die from excessive bleeding
- anaemic children have low birth weight, poor learning capacity, and less resistance to infections than other children

**TEST RESULTS**

Date \_\_\_\_\_

Man	Child 1	Child 2	Child 3
_____	_____	_____	_____
<b>Haemoglobin level (write) and Anaemia Classification (circle one)</b>			
_____ g/dl	_____ g/dl	_____ g/dl	_____ g/dl
<b>Severe</b> (less than 9.0 g/dL)	<b>Severe</b> (less than 7.0 g/dL)	<b>Severe</b> (less than 7.0 g/dL)	<b>Severe</b> (less than 7.0 g/dL)
<b>Moderate</b> (9.0-11.9 g/dL)	<b>Moderate</b> (7.0-9.9 g/dL)	<b>Moderate</b> (7.0-9.9 g/dL)	<b>Moderate</b> (7.0-9.9 g/dL)
<b>Mild</b> (12.0-12.9 g/dL)	<b>Mild</b> (10.0-10.9 g/dL)	<b>Mild</b> (10.0-10.9 g/dL)	<b>Mild</b> (10.0-10.9 g/dL)
<b>Normal</b> (13.0 g/dL or more)	<b>Normal</b> (11.0 g/dL or more)	<b>Normal</b> (11.0 g/dL or more)	<b>Normal</b> (11.0 g/dL or more)
<b>Height (cm)</b>			
_____	_____	_____	_____
<b>Weight (Kg)</b>			
_____	_____	_____	_____

**Severe Anaemia:** You have a seriously low level of haemoglobin in your blood. You need to see a doctor or visit a health centre immediately for treatment. Eat more foods rich in iron and treat malaria and worms immediately.

**Moderate Anaemia:** Your anaemia may be caused by iron deficiency, worms, excessive bleeding or malaria. You should visit a doctor or health centre as soon as possible. Eat more foods rich in iron.

**Mild Anaemia:** You need more daily iron. Treat malaria and worms immediately. Eat more foods rich in iron.

**Iron Rich Foods**



**dark green leafy vegetables**  
(spinach, kale, mustard, beet, parsley, amaranth)



**meat, liver or fish**



**lentils and beans**

To increase the body's use of iron, eat more fruits and vegetables rich in vitamin C:

lemons, guavas, mangoes, oranges



vegetables: green peppers, cabbage

**What CAUSES Anaemia?**

Anaemia is caused by loss of blood mainly due to:

- parasites, especially hookworms
- excessive menstrual losses
- chronic diseases such as ulcers or tuberculosis
- a lack of iron in the diet
- inability of the body to absorb iron from food

**How can Anaemia be PREVENTED?**

- Eat a diet adequate in iron-rich foods such as dark green leafy vegetables, liver, meat, or fish, and fruits rich in vitamin C.
- Avoid giving tea to infants and young children.
- Avoid taking coffee or tea with meals.
- Prevent and treat worms.
- Prevent malaria by using insecticide-treated bednets.
- Pregnant mothers and infants should take iron tablets or syrup.
- Limit the number of births by spacing children and delaying the first pregnancy.

**Implementing Agency**

[CONTACT INFORMATION: ADDRESS]



**National Family Health Survey**

**NFHS-4**

Name \_\_\_\_\_ Date \_\_\_\_\_

NFHS-4 will help us to find out the anaemia and nutritional status among men, women and young children in India.

We appreciate that you allowed us to interview you and to test you (and your child) for anaemia.

Thank you for your cooperation.

**Please look inside for the results of your anaemia test, and height and weight measurements.**

**APPENDIX 9: NFHS-4: Dried Blood Spot (DBS) TRANSMITTAL SHEET**

**NFHS-4: Dried Blood Spot (DBS) TRANSMITTAL SHEET (FRONT)**

**KEEP IN LARGE ZIPLOC BAG WITH SAMPLES UNTIL FINAL SIGNATURE OBTAINED**

HEALTH INVESTIGATOR CODE	CLUSTER NUMBER								
<table border="1" style="width: 100%; height: 20px;"> <tr> <td style="width: 25%;"></td> <td style="width: 25%;"></td> <td style="width: 25%;"></td> <td style="width: 25%;"></td> </tr> </table>					<table border="1" style="width: 100%; height: 20px;"> <tr> <td style="width: 25%;"></td> <td style="width: 25%;"></td> <td style="width: 25%;"></td> <td style="width: 25%;"></td> </tr> </table>				

PERSON SENDING/RECEIVING SAMPLES	TIME TO FILL IN FORM	TOTAL COUNT OF BLOOD SAMPLES	SIGNATURE CONFIRMING THAT EACH BLOOD SAMPLE IS PRESENT—SEE BACK OF FORM	SIGNATURE CONFIRMING THAT THE NUMBER OF BLOOD SAMPLES MATCHES COL. 3	DATE	NOTES (NOTE ANY DISCREPANCY IN NUMBERS OF SAMPLES)		
(1)	(2)	(3)	(4)	(5)	(6)	(7)		
HEALTH INVESTIGATOR	WHEN CLUSTER IS COMPLETED	<table border="1" style="width: 100%; height: 20px;"> <tr> <td style="width: 50%;"></td> <td style="width: 50%;"></td> </tr> </table>						
FIELD TEAM SUPERVISOR	AFTER INTERVIEWER HAS DONE HIS/HER COUNT	<table border="1" style="width: 100%; height: 20px;"> <tr> <td style="width: 50%;"></td> <td style="width: 50%;"></td> </tr> </table>						
SAMPLE PICK UP PERSON	WHEN SAMPLES ARE PICKED UP IN FIELD	<table border="1" style="width: 100%; height: 20px;"> <tr> <td style="width: 50%;"></td> <td style="width: 50%;"></td> </tr> </table>						
RECEIVER AT [LABORATORY]	UPON ARRIVAL AT [LABORATORY]	<table border="1" style="width: 100%; height: 20px;"> <tr> <td style="width: 50%;"></td> <td style="width: 50%;"></td> </tr> </table>						

**HEALTH INVESTIGATOR:** Upon completion of a cluster, verify that the barcode label on each blood sample (filter paper card) collected and stored in the large zip-loc bag labeled with the cluster number corresponds to a barcode number pasted to the back of this transmittal sheet (and vice-versa). Note any discrepancies in Column (7). Count and record the total number of blood samples in Column (3). Sign your name in Column (4) and the date in Column (6). Fold and store this transmittal sheet in the large zip-loc bag.

**FIELD TEAM SUPERVISOR:** After the interviewer has verified the blood samples, you will conduct a second verification. Count the number of blood samples, record the total in Column (3). Verify that the bar code number on each blood sample (filter paper card) collected and stored in the large zip-loc bag labeled with the cluster number corresponds to a barcode number pasted to the back of this transmittal sheet (and vice-versa) and sign Column (4). Verify that your count of blood samples is the same as the number reported by the interviewer and sign Column (5). Note any discrepancies in Column (7). Record the date in Column (6). Now scan the barcodes on the transmittal sheet into CAPI. Fold and store this transmittal sheet in the large zip-loc bag.

**SAMPLE PICK UP PERSON:** Before leaving a cluster, you will verify the number of blood samples collected in the completed cluster. For the completed cluster, count and record in Column (3) the total number of blood samples stored in the large zip-loc bag labeled with that cluster number. Note any discrepancies in Column (7). Sign your name in Column (5) and the date in Column (6). Fold and store this transmittal sheet in the large zip-loc bag.

**RECEIVER AT [LABORATORY]:** Upon receiving blood samples from the project office, verify that the barcode number on each blood sample (filter paper card) collected and stored in the large zip-loc bag labeled with the cluster number corresponds to a barcode number pasted to the back of this transmittal sheet (and vice-versa). Note any discrepancies in Column (7). Count and record the total number of blood samples in Column (3). Sign your name in Column (4) and the date in Column (6). Photocopy both sides of this form and return the original to the project office.

**Note: This form will be destroyed under the direction of the Laboratory Director after all blood samples have been completely processed and a final HIV test result has been determined for each usable sample.**

