hment 1.
E SHEET)

Libra. Date September 23, 1993

ETHICAL REVIEW ~~COMMITTEE~~, ICDDR,B.

ipal Investigator Dr.S.M.Akramuzzaman Trainee Investigator (if any) _____

cation No.: 93-025(REVISED) Supporting Agency (if Non-ICDDR,B) _____

of Study: A study to determine the rtance of nosocomial transmission of les ~~to validate salivary IgM assay~~ diagnosis of recent measles infection.

Project status:
( ) New Study
( ) Continuation with change
( ) No change (do not fill out rest of form)

e the appropriate answer to each of the following (If Not Applicable write NA).

ource of Population:
a) Ill subjects (Yes) No
b) Non-ill subjects (Yes) No
c) Minors or persons under guardianship (Yes) No

es the study involve:
a) Physical risks to the subjects Yes (No)
b) Social Risks Yes (No)
c) Psychological risks to subjects Yes (No)
d) Discomfort to subjects Yes (No)
e) Invasion of privacy Yes (No)
f) Disclosure of information damaging to subject or others Yes (No)

es the study involve:
a) Use of records, (hospital, medical, death, birth or other) (Yes) No
b) Use of fetal tissue or abortus Yes (No)
c) Use of organs or body fluids (Yes) No

e subjects clearly informed about:
a) Nature and purposes of study (Yes) No
b) Procedures to be followed including alternatives used (Yes) No
c) Physical risks Yes No
d) Sensitive questions Yes No
e) Benefits to be derived (Yes) No
f) Right to refuse to participate or to withdraw from study (Yes) No
g) Confidential handling of data (Yes) No
h) Compensation &/or treatment where there are risks or privacy is involved in any particular procedure Yes No

5. Will signed consent form be required:
   (a) From subjects Yes No
   (b) From parent or guardian (if subjects are minors) (Yes) No
6. Will precautions be taken to protect anonymity of subjects Yes No
7. Check documents being submitted herewith to Committee:
   ___ Umbrella proposal - Initially submit an overview (all other requirements will be submitted with individual studies).
   ✓ Protocol (Required)
   ✓ Abstract Summary (Required)
   ✓ Statement given or read to subjects on nature of study, risks, types of questions to be asked, and right to refuse to participate or withdraw (Required)
   ___ Informed consent form for subjects
   ✓ Informed consent form for parent or guardian
   ✓ Procedure for maintaining confidentiality
   ✓ Questionnaire or interview schedule *

* If the final instrument is not completed prior to review, the following information should be included in the abstract summary:
1. A description of the areas to be covered in the questionnaire or interview which could be considered either sensitive or which would constitute an invasion of privacy.
2. Examples of the type of specific questions to be asked in the sensitive areas.
3. An indication as to when the questionnaire will be presented to the Cttee. for review.

e to obtain approval of the Ethical Review Committee for any changes ing the rights and welfare of subjects before making such change.

akram

Principal Investigator

Trainee

ENTERED 21 JUN 1993

## SECTION I - RESEARCH PROTOCOL

| | |
|---|---|
| Title: | A study to determine the importance of nosocomial transmission of measles and to validate salivary IgM assay for diagnosis of recent measles infection. |
| Principal Investigator: | Dr. S.M. Akramuzzaman, ICDDR,B |
| Co-investigators: | Dr. Darul Islam, Dhaka Shishu (Children) Hospital<br>Ms. Leanne Unicomb, ICDDR,B |
| Consultants: | Dr. FT Cutts, London School of Hygiene & Tropical Medicine<br>Dr. D. Mahalanabis, ICDDR,B (Project coordinator) |
| Starting date: | As soon as the protocol is approved. |
| Completion date: | Two and a half years from the starting date. |
| Total direct cost: | US $ 50,320 |
| Name of the division: | Clinical Sciences Division (CSD) |
| Signature of CSD Head: | Dmahalanabis. |
| Date: | 2-8-93 |

August 2, 1993

Abstract summary:

Anecdotal reports of nosocomial measles continue to be common. However, only a few studies have been directed towards measuring the importance of nosocomial measles transmission. To evaluate whether health facility visit is a risk factor for acquiring measles, a clinic based case-control study with 294 cases and 1176 hospital and neighborhood controls of 6-59 months of age will be conducted. If nosocomial measles infection is documented to be a risk factor for measles transmission, it will alert health care providers to adopt policies to immunize children at every contact with health facilities to reduce measles transmission. To reduce the chance of misclassification bias, we will confirm all measles cases serologically and measure measles antibodies in a sub-sample of the control group.

We also propose to determine the sensitivity and specificity of a method of detection of measles-specific IgM in saliva in comparison to serum antibody levels. This is a simple and non-invasive technique that can be potentially used in a variety of epidemiological investigations in future.

Research Review Committee _____

Ethical Review Committee _____

Director _____

## SECTION II - RESEARCH PLAN

### A. INTRODUCTION

#### 1. Objectives:

Main-objectives:
Measure the risk of acquiring measles infection through nosocomial transmission.

Validate salivary antibody capture assays for the diagnosis of recent measles infection.

Sub-objectives:
Obtain estimates of measles vaccine efficacy; obtain descriptive information on primary and secondary cases, and places of exposure of measles cases attending hospitals.

#### 2. Background and rationale:

Before the introduction of measles vaccine, measles caused an estimated 130 million cases and 3 million deaths globally per year. In the past decade, vaccination programs have increased measles vaccine coverage dramatically, but measles still causes an estimated 880,000 deaths per year.[1] Thus measles is responsible for more deaths than any other EPI diseases. As average coverage increases, it becomes more important to identify high-risk groups for vaccination, and to investigate patterns of measles transmission so that specific measles control activities can be conducted. One common method of measles transmission is nosocomial infection in children attending health facilities. Although WHO has been encouraging Ministries of Health to adopt policies of immunizing children at every contact with health facilities, the extent to which this is implemented at curative services varies widely, and anecdotal reports of nosocomial measles continue to be common.

In Abidjan, Cote d'Ivoire, a case-control study of measles at the major health center in the city estimated that two thirds of measles cases had been exposed to measles at a prior health facility visit.[2] In Yaounde, Cameroon, for children aged 6-23 months with measles, places of contact with measles cases were identified as a health center (42%), a neighbor's house (28%), the home (15%), the grandparent's village (15%), or unknown (10%). For children aged 2-6 years, places of contact were a neighbor's house (78%), the home (28%), a health center (6%), or unknown (11%)[3]. Measles transmission in emergency rooms played a prominent role in perpetuating outbreaks of measles in Los Angeles.[4] Davis et al reported that the proportion of measles cases in 30 states that involved transmission in medical settings increased from 0.7% in 1980 through 1982 to 3% for 1983 and 1984.[5] Foulon et al reported that 16% cases of measles who were admitted in a hospital in Paris acquired the infection in a medical setting.[6]

The role of uncontrolled nosocomial transmission in the propagation of a community outbreak of measles was also demonstrated by Raad et al.[7] Reports have also demonstrated that the frequency of complications such as otitis media, bacterial pneumonia, conjunctivitis and laryngitis[6] are more common in hospital acquired measles. The risk of hospitalization is also higher in unvaccinated hospital acquired measles cases.[7] Therefore, more attention needs to be given to methods of preventing spread of measles in medical facilities not only to reduce incidence of measles in the community, but also to reduce complication rate.

Well designed studies are required to demonstrate the importance of nosocomial transmission in the spread of measles, particularly in developing countries where such information is inadequate. If nosocomial measles infection is documented to be of public health significance, appropriate interventions will be designed and a follow-up study conducted to evaluate the success of the interventions in reducing nosocomial transmission.

Another important research area identified by WHO is the development of simple laboratory techniques to diagnose measles infection. As the coverage of measles vaccine is increasing the accuracy of clinical diagnosis is diminishing with emergence of milder forms of the disease in the community. Failure to consider clinical misclassification of measles and other rashes, particularly in dengue endemic areas,[8] during concomitant rubella outbreaks[9] and in young infants, may result in inappropriate diagnostic conclusions and misdirected control efforts for both diseases. Also failure to identify correctly true measles cases may result in the loss of confidence by the community in the use of measles vaccine. In these circumstances a simple and non-invasive diagnostic tool which can be applied in the community is an urgent requirement for epidemiological investigations and surveillance. The Public Health Laboratory Service (UK) has developed a method for detecting IgM to measles in the saliva of recent cases by antibody capture. This salivary assay has been acceptable to parents, children and professionals and when used in controlled circumstances has proved both sensitive and specific in the UK.[10] Validation of this technique against serology (serum IgM) is required in developing country populations, as it is a potential non-invasive technique for the confirmation of measles which could assist in a variety of epidemiological studies on measles.

The Bangladesh Expanded Program on Immunization (EPI), which began operations in 1986[11], has achieved success in achieving national measles vaccination coverage of 54%.[12] Measles epidemics continue to occur;[13] but, there has been little information on the protective efficacy of measles vaccine administered in different settings, which is essential for a successful vaccination program. We have seen only one report of vaccine efficacy determination in a rural setting of Bangladesh where vaccine efficacy in 9-59 month old children was found be only 50%.[13] There has been an increase in the number of vaccinated measles cases attending Dhaka Shishu Hospital (Personal communication: Dr. Darul Islam) and the Clinical Research Centre (ICDDR,B) during this year, which indicates the need to assess measles vaccine efficacy in Dhaka city. The Poor vaccine

efficacy has also been reported from other developing countries, e.g., 44% from Kenya[14] where measles vaccine coverage was 80% and 55% from Bissau[15] where measles vaccine coverage was 81%. In the absence of laboratory back-up service for serological investigations which are expensive, the success of vaccination programs under field conditions can be assessed by vaccine efficacy determined by epidemiological means. Vaccine efficacy studies using the case-control method are most useful when personal immunization records are not generally available, but some other source such as records from one or more clinics, can be obtained. Thus an intensive effort can be made to determine the vaccination status of a limited number of cases and non-cases (controls) instead of concentrating on the whole population. If vaccine efficacy is found to be poor, further careful investigations will be required on proper maintenance of cold chain and vaccination techniques so that vaccination programs can be improved with reduction of incidence of measles in the vaccinated children. In vaccine efficacy determination, a highly specific case definition is required[16] this being a potential advantage of clinic-based studies where laboratory tests are available to assist in the classification of disease status. The data set generated from the nosocomial transmission study will also allow us to determine measles vaccine efficacy with adequate precision.

## B. SPECIFIC AIMS

1. To evaluate the risk of health facility visits for measles transmission, a matched case-control study among cases with measles and controls with other diseases will be conducted among children aged 6-59 months attending two large urban hospitals. All cases will be confirmed serologically. The odds of health facility visits will be compared between cases and controls during the 7-21 days prior to the development of their diseases. The study will have 80% power to detect an odds ratio of 2.0 at 5% level of statistical significance.

2. To measure the sensitivity and specificity of detection of measles specific IgM in saliva against serum IgM as the standard, salivary samples will be obtained from a sub-sample of children and compare with serum results for cases and non-cases.

## C. METHODS AND MATERIALS

### Study design:

This will be a clinic-based matched case-control study. The Clinical Research Center of the International Center for Diarrhoeal Disease Research, Bangladesh (ICDDR,B) and Dhaka Shishu (Children) Hospital will be used for selection of cases and controls. The ICDDR,B treats patients with diarrhoeal diseases and other related illnesses, about 60% of whom are children. Dhaka Shishu Hospital is the largest paediatric hospital in Dhaka city. Most of the patients of both these hospitals come from within and around Dhaka city and belong to middle and low socio-economic class.

Rodrigues et al[17] describe two conceptual approaches for the selection of controls. For the odds ratio from a case-control study to approximate the relative rate concurrent design is appropriate. In this, controls are selected concurrently from those still at risk (did not have measles in the past) when each new case is diagnosed. A child originally selected as a control can therefore, at a later date, be ascertained as a case. A child selected as both a control and a case is included in both groups during analysis. In this method it is necessary to obtain the past history of measles to select controls and exclude these controls from the analysis, in order to include only the children at risk of measles as controls. In this design the control children represent the person years at risk experience. Measles exhibits seasonality, but cases will be selected at the same time as their matched controls and the matched analysis will also match for time of selection thus yielding an unbiased estimate of the rate ratio.

For the odds ratio to approximate the relative risk an inclusive design[15] is appropriate. In this the controls are selected from among children regardless of whether or not they previously had measles. Here, the role of the control group is to estimate the proportion of the total population that is exposed. It represents those identified as being at risk at the start of the study. The odds ratio of exposure of cases to controls therefore yields an estimate of the risk ratio. Since the control group reflects the total population, a person ascertained as a case may also be selected as a control, and vice versa. Such children are included in the study as both cases and controls. In this method past history of measles in the controls will not be a criterion for exclusion. In an inclusive design for the measurement of vaccine efficacy, the risk ratio correctly measures the proportion of the exposed population that was totally protected, with one minus the relative risk yielding an estimate of protective efficacy of the measles vaccine. This model is thought to be appropriate for measles vaccine, where a certain proportion of vaccinated children are completely protected, and the non-responders have the same risk of disease as the unvaccinated. However, it will be of interest to evaluate the effect of using either a concurrent or an inclusive design on the estimate of vaccine efficacy obtained. For data collection, we will recruit controls irrespective of past history of measles. We will obtain a history of past measles on all children, and increase the sample size, so that if necessary controls with a past history of measles can be excluded from either analysis.

A set of hospital controls with diseases of similar severity to that of measles and another set of neighborhood controls, matched for age, will be used for each case to estimate the risk of nosocomial transmission. Data for children aged 9-59 months among the same sets of cases and controls will be used to determine measles vaccine efficacy. The primary value of using these two sets of controls lies in the detection of selection bias via comparison of estimates of effect from two sets of controls. If the effects do not differ, there will be no selection bias and this will strengthen the result. This will also provide direction for selection of controls for future case-control studies to study the effects of these risk factors.

**Sample size:**

1. **For the risk of nosocomial transmission:**

The null hypothesis that the true odds ratio equals unity implies that the proportions of exposed individuals among the cases and controls are equal. The sample size of each group (cases and controls) is therefore calculated using the formula for comparing two proportions:

$$n = \frac{2p(1-p)\ (Z_{\alpha/2} + Z_{\beta})^2}{D^2}$$

where $D = p_1 - p_2$

$p_1$ is the proportion of controls visiting health facility (exposed)

$p_1$ = 0.1, determined from a recent cross-sectional survey among all the hospitalized children (6-59 months of age) at the Clinical Research Centre of the ICDDR,B.

$p_2$ is the proportion of cases visiting health facility

We are interested in detecting an effect if health facility visit is associated with a 2 fold (or more) increase in occurrence of measles.

$$p_2 = \frac{0.1 \times 2}{1 + 0.1 \times (2 - 1)} = 0.18$$

$$p = (0.1 + 0.18)/2 = .14$$

For a two-sided significance level of 5%, $Z_{\alpha/2} = 1.96$,
and to achieve a significant result with power of 80%, $Z_{\beta} = 0.84$,
minimum sample size required:

$$n = \frac{2 \times 0.14\ (1 - 0.14) \times (1.96 + 0.84)^2}{(0.1 - 0.18)^2}$$

$$= 294$$

It will be easier to recruit controls than cases, in which case 4 controls can be recruited per case, with a consequent modest reduction in the number of cases required. Corresponding sample size for the case group would be given by:

$$n = (4 + 1) \times 196/(2 \times 4)$$

$$= 184$$

Adding 30% for taking into account of the potential confounders and another 30% for exclusion of controls with past measles during analysis, we will have to recruit about 294 cases and 294 x 4 = 1176 controls.

**2. For validation of salivary IgM assay:**

For sensitivity: We wish to estimate the proportion (P) of measles-specific salivary IgM positives among clinically diagnosed and serologically confirmed recent measles cases. We like our estimate (p) to have a probability of 0.95 (sensitivity) with in ±0.05 of the true value (P), then

$$1.96\ SE(p) = 1.96\sqrt{[p(1-p)/n]} = 0.05$$

$$= 1.96\sqrt{[0.95(0.05)/n]} = 0.05$$

expresses the prior opinion that we would like 1.96 SE of our estimate to equal to 0.05. Then our sample estimate has 95% chance of being with in 5 percentage points of the true value (P). Now solving for n,

$$n = 73$$

Adding 10% for loss or damage of samples, we will need 80 clinically diagnosed and serologically confirmed recent measles cases.

For specificity: For estimation of specificity of 95% with 5% precision we will need 80 controls (calculated in the same way as above) who are serologically confirmed negatives for recent measles.

**3. For protective efficacy of measles vaccine:**

Our aim in this study is to estimate the odds ratio of 0.15 for measles associated with measles vaccination with an absolute precision of 0.1.

The approximate 95% confidence interval for the OR (odds ratio), is expected to extend from OR/f to ORxf where f is the error factor.

For vaccine efficacy (VE) of 85%, i.e. OR = 0.15, the upper bound of the 95% confidence interval of OR is expected to be 0.25.

$$OR \times f = 0.25$$

$$f = \frac{0.25}{0.15} = 1.67$$

We assume that $p_1$ = the proportion of controls vaccinated will be 0.5, that we expect the odds ratio to be about 0.15, and that we set f to 1.67, so that 95% confidence interval is expected to extend from 0.09 to 0.25.

$p_2$ = the proportion of measles cases vaccinated

$$= \frac{0.5 \times 0.15}{1 + 0.5 \times (0.15 - 1)}$$

$$= 0.13$$

The number required in each group of cases and controls is therefore given by[18]

$$n = (1.96/\log_e 1.67)^2 \times [1/0.5(1-0.5) + 1/0.13(1-0.13)]$$

$$= 187$$

It will be easier to recruit controls than cases, in which case 4 controls can be recruited per case, with a consequent modest reduction in the number of cases required. Corresponding sample size for the case group would be given by:

$$n = (4 + 1) \times 187/(2 \times 4)$$

$$= 117$$

Adding 30% for taking into account of the potential confounders and another 30% for exclusion of controls with past history of measles, we will have to recruit about 187 cases and 748 controls.

Even excluding up to 20% of children aged less than 9 months of age from cases selected for measuring the risk of nosocomial measles transmission, we will be able to use 235 of them for measuring protective efficacy of measles vaccine with the specified precision.

Therefore, 294 recent measles cases and 1176 hospital controls and the same number of neighborhood controls recruited for estimation of risk of nosocomial measles transmission will also allow us to estimate measles vaccine efficacy.

**Selection of cases:**

Eligibility criteria for cases:

- 6-59 months of age
- Appearance of measles rash within the last 6 (in order that IgM can be verified) weeks
- A standard definition for measles will be followed:[1]
  - generalized maculo-papular rash of 3 or more days duration, and
  - history of fever (HOT to touch) and
  - at least one of the following: cough, coryza or conjunctivitis.

Physicians posted at outpatient or inpatient departments will identify measles cases in the course of their normal duties. Trained Health Assistants will be stationed at outpatient and inpatient departments of the

hospitals and will guide measles cases to one of the Investigators. Health Assistants will also visit the short-stay observation and inpatient wards every day to identify children who develop measles during their stay on the wards. For cases which have clearly acquired measles while on the hospital ward (more than 7 days after admission), will not be included in the study. We will make a note of them, but there is no point taking controls since they will also have been in the hospital for the same length of time. Clinical diagnosis will be confirmed by eliciting the history of present illness and by thorough physical examination by the investigator who will also obtain informed consent as soon as the diagnosis is made. The health assistants will interview parents according to standardized and pre-tested questionnaire (Appendix) and take anthropometric measurements.

Information on cases will include age, socioeconomic status, crowding index, address, history of place and time of contact, sex of contact case, history of visiting a health center within 7-21 days before the appearance of rash, distance travelled and time taken to reach the health facility, duration since appearance of rash prior to reporting, vaccination history and previous use of health facilities. Vaccination history will be verified wherever possible by documentation.

**Selection of controls:**

Controls will be children (6-59 months of age) attending the hospitals for curative care. Inpatient controls will be selected for inpatient cases, being those admitted the same week as the cases. Outpatient controls will be selected for outpatient cases, being the next eligible children seen at the outpatient department, in the same week as the case. Controls will be selected from children who present with diseases which are not themselves transmitted nosocomially to any great extent, and which are perceived by mothers to have the same severity as measles (therefore as likely as cases to be taken to the health facility if they were to acquire measles).[19] Preliminary work will be conducted to have a idea of mothers' perceptions of the severity of different diseases by comparing a sample of measles cases and children with the following diseases with regard to distance travelled and time taken to reach the health facility, and duration of the episode prior to reporting. Diseases which could potentially be included as controls include acute diarrhoeal diseases, enteric fever, hepatitis, urinary tract infections, meningitis, PUO, thrush, skin infections. Respiratory tract infections including pertussis will be excluded because of potential for nosocomial transmission. Polio, tuberculosis and surgical cases will not be selected as controls since these conditions will not lead a mother to take the child to a health facility with the same probability as for measles because of different perceptions of severity and usefulness of medical care for these conditions. Controls will be matched to cases within the following age categories 6-8, 9-11, 12-23, 24-35, 36-47 and 48-59 months. The Health Assistants will identify suitable controls and the diagnosis of the present illness will be confirmed by one of the investigators by obtaining the history of present illness and by thorough physical examination. The investigator will also obtain informed consent. The Health Assistant will

interview parents of controls in the same location as the corresponding cases using a similar questionnaire.

Similar information as cases will be collected for controls (age, socioeconomic status, address, history of visiting a health center within 7-21 days before the onset of the disease for which she/he is attending the hospital, distance travelled and time taken to reach the health facility, duration of the episode prior to reporting, vaccination history and history of previous use of health services).

Neighborhood controls (6-59 months of age), matched within the same age categories of cases as before, will be selected in order to control for exposure to measles in cases and controls. After the case household is identified, the interviewer will proceed to the 'neighboring' household. This will be defined as that residence whose front door is physically closest to the one just visited. The process of visiting households will be repeated until a total of four children of appropriate age will be selected for a case. As an operating rule, all eligible children in the household contributing the fourth children for each case will be included, even if that results in 5-6 children for that case. Information on use of health services will be collected to determine the extent of the potential bias when cases are recruited at health facilities and controls are recruited at home.

In order to reduce the level of bias introduced into the study by the interviewer (particularly to reduce misclassification of exposure status) the Health Assistants and mothers will be kept blind of the hypothesis of the study and two different interviewers will interview the case and the matched controls respectively. However, interviewers will be swapped around so that each of them interviews cases and controls in sequence.

**Anthropometry:**

History and physical examination will be followed by Anthropometric measurements:

- Stature (accurate to the nearest 0.1 cm), using locally constructed stadiometers (for >2 years) and length boards (for $\leq$ 2 years).
- weight (accurate to the nearest 20 gms), using high quality weighing scales.
- mid-upper-arm-circumference (accurate to the nearest 0.1 cm), using a flexible and non-stretchable tape.

Methods of taking weight and height and their standardization will be followed as described by the WHO.[20] Anthropometry will be performed by well trained Health Assistants in the presence of a physician and an average of three measurements will be considered as the observed value.

**Validation of diagnosis by serum and salivary IgM:**

One ml of venous blood will be drawn from ante-cubital veins of all cases to confirm the clinical diagnosis of measles by serum IgM assay at ICDDR,B virology laboratory. Blood will be delivered into sterile tubes and left to clot for 1-2 hours at room temperature. Serum will be separated by centrifugation and will be preserved $<-60^0$ or lower until assay is performed. Blood will also be collected from a sub-sample of consecutive hospital controls (according to recruitment) until 80 serologically negative controls are found for validation of salivary IgM assay. Sera will be tested for anti-measles IgM using a commercially available enzyme linked immunosorbent assay (ELISA) kit (Behring Enzygnost Anti-measles-virus /IgM). In a large scale field evaluation during an epidemic (10,148 reported cases) the ELISA kit (Behring) was found to be highly sensitive (97.2%) among clinically documented subjects with significant increase in complement-fixing antibody.[21] The test (ELISA kit, Behring) also agreed well with immunofluorescence (97% agreement) for confirmation of clinically diagnosed measles cases during an epidemic.[22] Since an excess of IgG or rheumatoid factor (IgM-RF) may interfere in the IgM enzyme immunoassay, measles-IgM ELISA will be carried out by using sera diluted in rheumatoid factor absorbent (sheep anti-human IgG; RF absorbent, Behring) to precipitate IgG and IgG-linked IgM-RF. Each run will include low-positive, high-positive and negative controls. Serum IgM results will be expressed as positives or negatives based on the criteria given by the manufacturer.

In addition, saliva samples will be collected by saliva swabs from above sub-sample of 80 serologically confirmed consecutive (according to recruitment) cases and the sub-sample of 80 serologically negative controls immediately after collection of blood samples to assess the sensitivity and specificity of the salivary IgM assay compared to serum IgM. The saliva swab consists of a 3 cm cylinder of expanded polystyrene foam on a handle of tubular cardboard and can be used like a toothbrush. Saliva samples will be taken by rubbing a sponge swab around the gums (like a toothbrush) for about 60 seconds until saturated. This is particularly suitable for collecting salivary specimens from young children. The swabs will be vortexed in 1 ml phosphate buffered saline (PBS) containing 10% fetal calf serum (FCS) and 0.1% Tween20 (T). This will be stored without further treatment at $-30^0$C until testing. Measles virus-specific salivary IgM will be detected by antibody capture radioimmunoassay at Public Health Laboratory Service (UK) using methods described by Perry KR et al.[10] The test results will be expressed as reactive (positive) or unreactive (negative) based on the ratio of the total bound radioactivity of each specimen to the mean radioactivity bound of the 4 negative serum controls (Test:Negative >3.0 = reactive). This test was found to be 100% sensitive in clinically diagnosed and serologically confirmed measles cases between 1-5 weeks after the onset of symptoms and was found to be 100% specific when tested in a group of blood donors.[10]

For objective evaluation, the tests will be performed in a blinded fashion. To eliminate the chance of arising bias in generating the data the clinicians responsible for making diagnosis will be kept blind of the results of the

IgM tests and the laboratory testers will be kept blind of the clinical diagnosis of the cases and controls till the study is over.

**Management of cases and controls:**

All cases and hospital-controls will be appropriately managed either on out-patient or in-patient basis according to the routine practice of the respective hospitals, this includes administration of vitamin A. All unvaccinated study children (including neighborhood controls) will be vaccinated with appropriate EPI vaccines and their mothers will be made aware of the dangers of EPI diseases and advised to vaccinate other children in their households.

**General data management plan:**

Data management will progress according to the following scheme:

- Data entry forms for interviewer administered questionnaire and for other variables such anthropometry and laboratory results will be designed, standardized and pre-tested.
- Interviewer will collect data and will complete questionnaires.
- Interviewer will check the questionnaire thoroughly as soon as the interview is complete and will correct any errors, returning to the respondent if necessary.
- One of the investigators will check questionnaire, reviewing 10% of sample respondents.
- A data entry clerk will enter the data in a microcomputer using Epi-info.
- A different data entry clerk will enter data into a microcomputer a second time.
- The two data files will be compared to find any typing errors, and errors will be corrected.
- Either at the time of data entry (interactive checking), or afterwards (batch checking), data will be checked to ensure that they are within the allowable range, and that they are consistent from one question to another. The batch checking will be made separately in cases and controls as the distributions may be quite different. Any errors found will be corrected.
- When data are cleaned, new variables will be created from the raw data.
- Data of different questionnaires and forms will be linked to one other.

**Data analysis plan:**

<u>For risk of nosocomial measles transmission & protective efficacy of vaccine:</u>
For the hospital visit as a risk factor of measles with four controls matched to each case, the observations will be represented as matched case-control quintuplets (case 1, control 4). Treating each matched quintuplets as a separate stratum and using the methods for stratified data we can obtain a Mantel-Haenszel estimate of the odds ratio as :

$$\psi = \frac{\text{Total no. of unexposed controls who have an exposed case}}{\text{Total no. of exposed controls who have an unexposed case}}$$

where, exposure will be defined as prior visit to a health facility and a case is a child who will have measles.

Similarly for determination of protective efficacy of measles vaccine with four controls matched to each case, the observations will be represented as matched case-control quintuplets (case 1, control 4). Treating each matched quintuplet as a separate stratum and using the methods for stratified data as above we can obtain a Mantel-Haenszel estimate of the odds ratio as:

$$\psi = \frac{\text{Total no. of unvaccinated controls who have an vaccinated case}}{\text{Total no. of vaccinated controls who have an unvaccinated case}}$$

Vaccine efficacy (VE) will be determined by using the following formula:

$$VE\ (\%) = (1-RR) \times 100, \quad \text{where, } RR = \frac{\text{Risk of measles in the vaccinated}}{\text{Risk of measles in the unvaccinated}}$$

$$= (1 - \psi) \times 100$$

A $X^2$ test for the null hypothesis that the true odds ratio is 1 will be obtained by comparing the observed and expected number of cases using the Mantel-Haenszel $X^2$ test for stratified analysis. Confidence limits of odds ratios will be obtained from the variance of the logarithm of the Mantel-Haenszel estimate of the odds ratio for stratified data.

The additional variables, such as socio-economic conditions, not matched for, may confound the effects of the exposures of interest, i.e., prior visit to health facilities. The effect of these potential confounders on the exposures of interest, the mutual confounding of these and other variables and their interactions will be examined by conditional logistic regression analysis.

For sensitivity and specificity of salivary IgM assay:

The proportion of serologically confirmed measles cases ($N_1$) having positive salivary IgM will the sensitivity (SEN) of the salivary IgM assay with standard error:

$$\text{Standard error of sensitivity} = \sqrt{[SEN(1-SEN)/N_1]}$$

Similarly, the proportion serologically negative controls ($N_2$) having negative salivary IgM will the specificity (SPE) of the salivary IgM assay with standard error:

$$\text{Standard error of specificity} = \sqrt{[SPE(1-SPE)/N_2]}$$

## D. STUDY SCHEDULE:

1. Recruitment of personnel and their training. Procurement of supplies and materials. Preparation of questionnaire and data forms, and pre-testing them. Standardization of all procedures - 3 months.

2. Selection of cases and controls, mothers' interview, physical examination, anthropometry, collection of laboratory specimens, laboratory studies and data entry - 2 years.

3. Data analysis and report writing - 3 months.

## BUDGET

A study to determine the importance of nosocomial transmission of measles, to validate salivary IgM assay for diagnosis of recent measles and to determine measles vaccine efficacy.

| Personnel Cost: | Grade | % of effort | No. | Salary per month | Total salary (for 24 months) |
|---|---|---|---|---|---|
| Dr. SM Akramuzzaman | NOB | 25% | | 300 | 7,200 |
| Dr. Darul Islam | - | 10% | | - | - |
| Ms. Leanne Unicomb | - | 5% | | - | - |
| Dr. FT Cutts | - | 5% | | - | - |
| Dr. D Mahalanabis | - | 5% | | - | - |
| Physician | Spl. Gr. | 100% | 1 | 150 | 3,600 |
| Health Assistants | Spl. Gr. | 100% | 3 | 150 X 4 | 14,400 |
| Laboratory Technician | Spl. Gr. | 100% | 1 | 150 | 3,600 |
| Data Entry Technician | Spl. Gr. | 100% | 1 | 150 | 3,600 |
| Health Workers | Spl. Gr. | 100% | 3 | 20 X 4 | 1,920 |
| Sub-total | | | | | US $ 34,320 |

Local Travel

| | |
|---|---|
| Daily visit to Dhaka Shishu Hospital (to collect records & samples) | 1,000 |
| Visits to the houses of neighborhood controls and some clinics | 1,000 |
| Sub-total | US $ 2,000 |

Supplies and Materials

| | |
|---|---|
| Hospital supplies (cotton, needle, syringes, lancets, solution, leukoplast etc.) | 400 |
| Stationery and office supplies (book register, binders, files, pencils, fastener, paper, ribbon, stapler etc.) | 500 |
| Glassware (tubes, seal etc.) | 100 |
| Sub-total | US $ 1000 |

| | |
|---|---|
| **International travel** | |
| To discuss data analysis and report writing in London | 4,000 |
| Visit of consultant | 2,500 |
| Sub-total | US $ 6,500 |
| **Other costs:** | |
| Rent, communication and utilities (postage, telephone, telegram etc.) | 200 |
| Printing & publication (printing of forms, reprints etc.) | 300 |
| Transportation of saliva samples | 500 |
| Repairs and maintenance (maintenance and repair of equipments) | 100 |
| Sub-total | US $ 1,100 |
| **Interdepartmental Services:** | |
| Pathological tests: Serum IgM (saliva IgM assay: free of cost) | 2,000 |
| Medical Illustrations | 200 |
| Telex/Fax | 100 |
| Maintenance charges | 200 |
| Xerox/mimeograph | 300 |
| Transport subsidy | 100 |
| Sub-total | US $ 2,900 |
| **Capital expenditure** | |
| Weighing scales | 300 |
| Length boards | 200 |
| Microcomputer | 2,000 |
| Sub-total | US $ 2,500 |
| **Total direct cost** | US $ 50,320 |
| **Indirect cost (13% of direct cost)** | US $ 6,542 |
| **Total Project Cost** | US $ 56,862 |

**Abstract summary for the Ethical Review Committee**

Anecdotal reports of nosocomial measles continue to be common. However, only a few studies have been directed towards measuring the importance of nosocomial measles transmission. To evaluate whether health facility visit is a risk factor for acquiring measles, a clinic based case-control study with 294 cases and 1176 hospital and the same number of neighborhood controls of 6-59 months of age will be conducted. If nosocomial measles infection is documented to be a risk factor for measles transmission, it will alert health care providers to adopt policies to immunize children at every contact with health facilities to reduce measles transmission. To reduce the chance of misclassification bias, we will confirm all measles cases serologically and measure measles antibodies in a sub-sample of the control group.

We also propose to determine the sensitivity and specificity of a method of detection of measles-specific IgM in saliva in comparison to serum antibody levels. This is a simple and non-invasive technique that can be potentially used in a variety of epidemiological investigations in future. We are addressing the following items for necessary information for the committee:

1. The incidence of measles and mortality from this disease is the highest in children bellow five years of age. The likelihood of acquiring measles from health facilities is high in these children because of their frequent visits to health facilities for obtaining treatment for diarrhoea, ARI and other diseases. The rationale of including two sets of controls is to strengthen the validity of the study by excluding the possibility of selection bias. However, no blood or salivary samples will be collected from neighborhood controls.

2. The study will not cause any risk to the children.

3. Measles cases and hospital controls will be provided with the best possible clinical care available in the participating hospitals. In addition all study children will be given a dose of high potency vitamin A capsule on recruitment into the study. All unvaccinated study children including the neighborhood controls will also be immunized with appropriate EPI vaccines and their parents will be advised to vaccinate other children in their households.

4. Confidentiality of childrens' records will be maintained. Unique study numbers will be used during analysis.

5. Informed consent in a consent form will be obtained from the legal guardian or parents before inclusion in the study. They will be fully informed and explained about the objectives and benefits of the study. For cases and hospital controls the consent will be obtained at the hospital and for neighborhood controls the consent will be obtained at the residences of the children.

6. A short interview of the mothers or legal guardians of all cases and controls will be taken by the health assistants on their childrens' present health, past illnesses, socio-economic and environmental conditions and history of contact with other measles cases. For measles cases and hospital controls the interview will be conducted at the hospital and for neighborhood controls the same will the conducted at their residences. The interview will last for approximately fifteen minutes.

7. There will be immediate benefit to the cases and hospital controls as they will be appropriately treated for their illnesses free of cost. Furthermore, unvaccinated children will be given appropriate EPI vaccines. The long term benefits to the society include identification of health facility visit as a potential risk factor for acquiring measles for which appropriate control measures could be adopted and identification of salivary IgM as a potentially simple and non-invasive technique for diagnosis of recent measles that could be used in clinical and epidemiological investigations.

8. Information obtained from the questionnaire, anthropometric measurements, laboratory results of serum and salivary IgM assay and hospital records will be used for data analysis. To reduce the chance of misclassification bias, small amount of blood (1 ml) will be collected by from ante-cubital veins of all cases and a sub-sample of hospital controls to serologically confirm and rule out the diagnosis of measles in all cases and the controls respectively. This simple procedure is routinely performed on all admitted children in these hospitals for their treatment. Saliva will be collected from a sub-sample of cases and hospital controls. This will be done by rubbing a sponge swab around the gums (like a toothbrush) for about a minute. This is also a very safe and simple procedure that can be performed without any discomfort to the children and now this is being performed in other studies in the Clinical Research Centre of the ICDDR,B.

## REFERENCES


1. Expanded Program on Immunization. Measles control in the 1990s: plan of action for global measles control. World Health Organization. WHO/EPI/GEN/92.3.

2. Klein-Zabban ML, Foulon G, Gaudebout C, Badoual J, Assi Adou J. Frequence des rougeoles nosocomiales dans un centre de protection maternelle et infantile d'Abidjan. Bull WHO 1987; 65:197-201.

3. Guyer B. Transmission de la rougeole a Yaounde en 1975. Medecine Tropicale 1976;36:451-53.

4. Farizo KM, Stehr-Green PA, Simpson DM, Markwitz LE. Pediatric emergency room visits as a risk factor for acquiring measles. Pediatrics 1991;87:74-79.

5. Davis RM, Orenstein WA, Frank JA, et al. Transmission of measles in medical settings 1980 through 1984. JAMA 1986;255:1295-98.

6. Foulon G, Cottin JF, Matheron S, Perronne C, and Bouvet E. Transmission and severity of measles acquired in medical settings. JAMA 1986;256:1135.

7. Raad II, Sheretz, Rains CS, Cisick JL et al. The importance of nosocomial transmission of measles in the propagation of a community outbreak. Infect Control Hosp Epidemiol 1989;10:161-6.

8. Dietz VJ, Nieburg P, Gubler DJ, and Gomez I. Diagnosis of measles by clinical case definition in dengue-endemic areas: implications for measles surveillance and control. Bull WHO 1992;70(6):745-50.

9. Lee MS, King CC, Jean JY et al. Sero-epidemiology and evaluation of passive surveillance during 1988-89 measles outbreak in Taiwan. Int J Epidemiol 1992;21:1165-74.

10. Perry KR, Brown DWG, Perry JV et al. The detection of measles, Mumps and rubella antibodies in saliva using antibody capture radioimmunoassay. J Med Virology (In press, J Med Virology, 1993).

11. Government of Bangladesh/ World Bank/ SIDA/ USAID/ UNICEF/ WHO. Joint review of the Expanded Program on Immunization in Bangladesh: summary report of findings, key issues and recommendations. Unpublished document, Ministry of Health and Family Planning, Dhaka, Bangladesh, 1989.

12. Expanded Program on Immunization. Report of the 14th Global Advisory Group Meeting, World Health Organization, Geneva, 1992.

13. Akramuzzaman SM. Epidemic measles in rural Bangladesh: The need for intensification of measles vaccine coverage and an effective vaccine. MSc. Dissertation. University of London. London School of Hygiene and Tropical Medicine, 1992.

14. Kenya PR. Measles and mathematics: "control or eradication". East Afr Med J 1990;67(12):856-63.

15. Aaby P, Knudsen K, Jensen TG et al. Measles incidence, vaccine efficacy and mortality in two urban African areas with high vaccination coverage. J Infect Dis;162(5):1043-8.

16. Cutts FT, Smith PG, Colombo S, et al. Field evaluation of measles vaccine efficacy in Mozambique. Am J Epidemiol 1990; 131:349-55

17. Rodrigues LC, and Kirkwood BR. Case control design in the study of common diseases. Int J Epidemiol 1990;19:205-13.

18. Schlesselman JJ. Case-control analysis - Design, Conduct, Analysis. Oxford University Press, New York, 1985.

19. Cousens SN, Feachem RG, Kirkwood BR, Mertens TE, Smith PG. Case-control studies of childhood diarrhoea: iv: choice of control group. CDD/EDP/90.1, World Health Organization, Geneva 1990.

20. Measuring change in nutritional status. Guidelines for assessing the nutritional impact of supplementary feeding programs for vulnerable group. World Health Organization. Geneva, 1983.

21. Ozzane G, and D'Halewyn M. Performance and reliability of the Enzygnost Measles Enzyme-Linked Immuno-Sorbent Assay for detection of measles virus-specific immunoglobulin M antibody during a large measles epidemic. J Clin Microbiol 1992;30(3):564-569.

22. Rossier E, Miller H, McCulloch B, Sullivan L, and Ward K. Comparison of immunofluorescence and enzyme immunoassay for detection of measles-specific Immunoglobulin M antibody. J Clin Microbiol 1991;29(5):1069-71.

CONSENT FORM - I
(FOR CASES)

FOR STUDIES OF NOSOCOMIAL TRANSMISSION OF MEASLES AND VALIDATION OF MEASLES-SPECIFIC SALIVARY IgM ASSAY

A large number of children in Bangladesh suffer from measles each year and many of them die from it's complications. A large proportion of them probably acquire measles during their visits to clinics and hospitals. The ICDDR,B is carrying out research to measure the risk of health facility visits for measles transmission so that appropriate measures could be adopted to interrupt measles transmission. This study will also allow us to validate a test with saliva in comparison to that with blood for diagnosis of recent measles. If this test is found to be sensitive, it would be a simple and non-invasive technique that could assist in a variety of epidemiologic investigations. Since your child has recently suffered from measles, inclusion of your child in this study will enable us to conduct above investigations. If you agree to enrol your child in this study, the following procedures will be performed.

A health assistant will interview you on your child's present health, past illnesses, socio-economic and environmental conditions and history of contact with other measles cases. She will measure height, weight and arm circumference of your child and a physician will examine your child thoroughly. Your child will be ensured of the best possible treatment available in this hospital for her/his illness free of cost. One ml (1/5th volume of a tea spoonful) of blood will be collected once from her/his finger prick which is a very simple and safe procedure and is being routinely performed on all admitted children in this hospital for their treatment. This will be done to confirm that your child has suffered from measles recently. In addition, a sample of saliva may be collected from your child's mouth by rubbing a sponge swab around the gums (like a toothbrush) for about a minute. This is also a very simple and safe technique which is now being performed for other studies in this hospital.

If at any time you wish to withdraw your child from the study, you are free to do so and even then she/he will receive adequate treatment.

If you are willing to include your child in this study, please sign or put your left thumb impression bellow.

Signature of the investigator

Signature/left thumb impression of the legal guardian

Signature of the witness

Date

সম্মতি - পত্র - ①
(হাম রোগীর জন্য)

# হাসপাতালে রোগীদের মাঝে হাম বিস্তার ও মুখের লালার সাহায্যে হাম নির্ণয়ের গবেষণা

প্রতি বছর বাংলাদেশের অসংখ্য শিশু হামে ভোগে এবং এদের মধ্যে অনেকেই এ রোগের কারণে মারা যায়। বিভিন্ন রোগের চিকিৎসার জন্য হাসপাতাল বা ক্লিনিকে থাকা অবস্থায় অন্য হাম রোগীর কাছ থেকে অনেক শিশুরই হাম হতে পারে। হাসপাতাল বা ক্লিনিকে শিশুদের নিয়ে যাওয়ার পর তাদের হাম হওয়ার ঝুঁকি কতটুকু তা জানার জন্য আন্তর্জাতিক উদরাময় গবেষণা কেন্দ্র গবেষণা করছে। রক্ত পরীক্ষার সাথে মুখের লালা পরীক্ষার তুলনা করে আমরা দেখব যে, লালা পরীক্ষার সাহায্যে হাম নির্ণয় করা যায় কিনা। যদি লালা পরীক্ষাটি সঠিক হিসাবে প্রমাণিত হয়, তাহলে চিকিৎসায় ও গবেষণায় রক্ত পরীক্ষার পরিবর্তে এ পরীক্ষাটি অনেক সহজ ও নিরাপদ হবে। যেহেতু আপনার শিশু হামে ভুগছে/ভুগেছিল, সেহেতু আপনার শিশুর আমাদের গবেষণায় অংশগ্রহনের মাধ্যমে এই পরীক্ষাগুলি করালে আমরা উপকৃত হব। আপনি যদি আপনার শিশুকে এই গবেষণায় অংশগ্রহন করাতে চান তাহলে আমরা আপনাকে নীচে উল্লেখিত কিছু প্রশ্ন করব ও আপনার শিশুকে পরীক্ষা করব।

একজন স্বাস্থ্যকর্মী আপনার শিশুর বর্তমান ও অতীত স্বাস্থ্য, আপনার পারিবারিক ও আর্থিক অবস্থা এবং আপনার শিশু হাম রোগীর কাছে গিয়েছিল কিনা ইত্যাদি বিষয়ে কিছু কথা বলবে। তাতে আপনার ১৫ মিনিট সময় নষ্ট হতে পারে। একই স্বাস্থ্যকর্মী আপনার শিশুর ওজন, উচ্চতা ও হাতের মাপ নিবে এবং একজন ডাক্তার তাকে খুব ভালভাবে পরীক্ষা করবে। আপনার শিশুর বিনামূল্যে হাসপাতালে চিকিৎসার সব প্রয়োজনীয় ব্যবস্থা করা হবে। আপনার শিশুর হাতের শিরা হতে সামান্য পরিমান রক্ত (১ মি.লি যা এক চা চামচের ১/৫ এর সমান) শুধুমাত্র একবার নেয়া হবে। এটুকু রক্ত হাসপাতালে ভর্তি প্রায় সব শিশুর কাছ থেকে নিয়মিতভাবে নেয়া হয়ে থাকে। আপনার শিশুর হাম হয়েছিল কিনা এ ব্যাপারে নিশ্চিত হওয়ার জন্যই এ পরীক্ষাটি করা হবে। একই সংগে একটা কাঠির আগায় খুব

নরম কাঠি দিয়ে আপনার শিশুর মুখ থেকে সামান্য পরিমাণ লালা নেয়া হ'তে পারে। এতে আপনার শিশুর কোন কষ্ট বা ক্ষতি হবে না। এই পরীক্ষা এ মুহূর্তে এই হাসপাতালে অন্যান্য গবেষণাতেও করা হচ্ছে।

আপনি যে কোন সময় এই গবেষণা থেকে আপনার শিশুকে বের করে নিতে পারবেন। এ গবেষনা থেকে বের করে নিলেও আপনার শিশুকে প্রয়োজনীয় চিকিৎসা করা হবে।

এই গবেষণায় আপনার শিশুকে অন্তর্ভুক্ত করতে চাইলে নীচে স্বাক্ষর করুন/ বাম হাতের বৃদ্ধাঙ্গুলীর ছাপ দিন।

গবেষণাকারীর স্বাক্ষর

অভিভাবকের স্বাক্ষর/বাম হাতের বৃদ্ধাঙ্গুলির ছাপ

তারিখ-

সাক্ষীর স্বাক্ষর

## CONSENT FORM - II
(FOR HOSPITAL CONTROLS)

FOR STUDIES OF NOSOCOMIAL TRANSMISSION OF MEASLES AND VALIDATION OF MEASLES-SPECIFIC SALIVARY IgM ASSAY

A large number of children in Bangladesh suffer from measles each year and many of them die from it's complications. A large proportion of them probably acquire measles during their visits to clinics and hospitals. The ICDDR,B is carrying out research to measure the risk of health facility visits for measles transmission so that appropriate measures could be adopted to interrupt measles transmission. This study will also allow to validate a test with saliva in comparison to that with blood for diagnosis of recent measles. If this test is found to be sensitive, it would be a simple and non-invasive technique that could assist in a variety of epidemiologic investigations. A child attended this hospital just before your one who recently had measles. Although your child did not have measles recently, inclusion of your child in this study will allow us to conduct above investigations by comparing the characteristics of that child with recent measles to those of your one. If you agree to enrol your child in this study, the following procedures will be performed.

A health assistant will interview you on your child's present health, past illnesses, socio-economic and environmental conditions and history of contact with other measles cases. Your child will be ensured of the best possible treatment available in this hospital for her/his illness free of cost. One ml (1/5th volume of a tea spoonful) of blood will be collected once from her/his finger prick which is a very simple and safe procedure and is being routinely performed on all admitted children in this hospital for their treatment. This will be done to confirm that your child has not suffered from measles recently. In addition, a sample of saliva may be collected from your child's mouth by rubbing a sponge swab around the gums (like a toothbrush) for about a minute. This is also a very simple and safe technique which is now being performed for other studies in this hospital.

If at any time you wish to withdraw your child from the study, you are free to do so and even then she/he will receive adequate treatment.

If you are willing to include your child in this study, please sign or put your left thumb impression bellow.

Signature of the investigator

Signature/left thumb impression of the legal guardian

Signature of the witness

Date

সম্মতি-পত্র-(২)
(হাসপাতাল কর্তৃপক্ষ-এর জন্য)

## হাসপাতালে রোগীদের মাঝে হাম-বিস্তার ও মুখের লালার সাহায্যে হাম নির্ণয়ের গবেষণা

প্রতি বছর বাংলাদেশের অসংখ্য শিশু হামে ভোগে এবং এদের মধ্যে অনেকেই এ রোগের কারণে মারা যায়। বিভিন্ন রোগের চিকিৎসার জন্য হাসপাতাল বা ক্লিনিকে থাকা অবস্থায় অন্য হাম রোগীর কাছ থেকে অনেক শিশুর হাম হতে পারে। হাসপাতাল বা ক্লিনিকে শিশুদের নিয়ে যাওয়ার পর তাদের হাম হওয়ার ঝুঁকি কতটুকু তা জানার জন্য আন্তর্জাতিক উদরাময় গবেষণা কেন্দ্র গবেষণা করছে। রক্ত পরীক্ষার সঙ্গে মুখের লালা পরীক্ষার তুলনা করে আমরা দেখব যে, লালা পরীক্ষার সাহায্যে হাম নির্ণয় করা যায় কিনা। যদি লালা পরীক্ষাটি সঠিক প্রমাণিত হয়, তাহলে চিকিৎসা ও গবেষণায় রক্ত পরীক্ষার পরিবর্তে এ পরীক্ষাটি অনেক সহজ ও নিরাপদ হবে। আপনার বাচ্চার আগে যে শিশুটি এই হাসপাতালে এসেছিল তার হাম হয়েছিল। যদিও আপনার বাচ্চার হাম হয়নি, তবুও সে এই গবেষণায় যোগ দিলে হামে ভোগা শিশুটির সাথে আপনার বাচ্চার তুলনা করে পরীক্ষা-নিরীক্ষার দ্বারা হাম বিস্তারের কারণ ও মুখের লালার সাহায্যে এ রোগ নির্ণয় সম্পর্কে জানতে পারব। আপনি যদি আপনার শিশুকে এই গবেষণায় অংশগ্রহণ করাতে চান তাহলে আমরা আপনাকে নীচে উল্লেখিত কিছু প্রশ্ন করব ও আপনার শিশুকে পরীক্ষা করব।

একজন স্বাস্থ্যকর্মী আপনার শিশুর বর্তমান ও অতীত স্বাস্থ্য, আপনার পারিবারিক ও আর্থিক অবস্থা এবং আপনার শিশু হাম রোগীর কাছে গিয়েছিল কিনা ইত্যাদি বিষয়ে কিছু কথা বলবে। তাতে আপনার ১৫ মিনিট সময় নষ্ট হতে পারে। একই স্বাস্থ্যকর্মী আপনার শিশুর ওজন, উচ্চতা ও হাতের মাপ নেবে এবং একজন ডাক্তার তাকে খুব ভালভাবে পরীক্ষা করবে। আপনার শিশুর বিনামূল্যে হাসপাতালে চিকিৎসার সব প্রয়োজনীয় ব্যবস্থা করা হবে। আপনার শিশুর হাতের শিরা হতে সামান্য পরিমাণ রক্ত (১ মি.লি যা এক চা চামচের ১/৫-এর সমান) শুধুমাত্র একবার নেয়া হবে। এটুকু রক্ত হাসপাতালে ভর্তি প্রায় সব শিশুর কাছ থেকে নিয়মিতভাবে নেয়া হয়ে থাকে। আপনার শিশুর হাম হয়েছিল কিনা এ ব্যাপারে

নিশ্চিত হওয়ার জন্যই এ পরীক্ষাটি করা হবে। একই সঙ্গে একটা কাঠির আগায় খুব নরম পাশ্ব দিয়ে আপনার শিশুর মুখ থেকে সামান্য পরিমাণ লালা নেয়া হতে পারে। এতে আপনার শিশুর কোন কষ্ট বা ক্ষতি হবে না। এ পরীক্ষা এই মুহূর্তে এই হাসপাতালে অন্যান্য গবেষণাতেও করা হচ্ছে।

আপনি যে কোন সময় এই গবেষণা থেকে আপনার শিশুকে বের করে নিতে পারবেন। এ গবেষণা থেকে বের করে নিলেও আপনার শিশুকে প্রয়োজনীয় চিকিৎসা করা হবে।

এই গবেষণায় আপনার শিশুকে অন্তর্ভুক্ত করতে চাইলে নীচে স্বাক্ষর করুন/বাম হাতের বৃদ্ধাঙ্গুলির ছাপ দিন।

গবেষণাকারীর স্বাক্ষর

অভিভাবকের স্বাক্ষর/বাম হাতের বৃদ্ধাঙ্গুলির ছাপ

সাক্ষীর স্বাক্ষর

তারিখ-

CONSENT FORM - III
(FOR NEIGHBORHOOD CONTROLS)

FOR THE STUDY OF NOSOCOMIAL TRANSMISSION OF MEASLES

A large number of children in Bangladesh suffer from measles each year and many of them die from it's complications. A large proportion of them probably acquire measles during their visits to clinics and hospitals. The ICDDR,B is carrying out research to measure the risk of health facility visits for measles transmission so that appropriate measures could be adopted to interrupt measles transmission. Recently, a child in your neighborhood has suffered from measles. Although your child has not suffered form measles recently, a comparison of the information for the child with measles and your child regarding the recent visit to a health facility will allow us to determine the risk of health facility visit for measles transmission. If you agree to enrol your child in this study, the following procedures will be performed.

A health assistant will interview you at your house on your child's present health, past illnesses, socio-economic and environmental conditions, recent visit to a hospital or a clinic and history of contact with measles cases. She will measure height, weight and arm circumference of your child and a physician will thoroughly examine her/him. In addition, your child will receive appropriate vaccines in order to prevent measles and other killer diseases free of cost.

If at any time you wish to withdraw your child from the study, you are free to do so and even then she/he will receive the above preventive measures.

If you are willing to include your child in this study, please sign or put your left thumb impression bellow.

_____
Signature of the investigator

_____
Signature/left thumb impression of the legal guardian

_____
Signature of the witness

Date_____

সম্মতি পত্র – ৩
(প্রতিবেশী কর্ণেল-এর জন্য)

## হাসপাতালে রোগীদের মাঝে হাম বিস্তারের গবেষণা

প্রতি বছর বাংলাদেশের অসংখ্য শিশু হামে ভোগে এবং এদের মধ্যে অনেকেই এ রোগের কারণে মারা যায়। বিভিন্ন রোগের চিকিৎসার জন্য হাসপাতালে বা ক্লিনিকে থাকা অবস্থায় অন্য হাম রোগীর কাছ থেকে অনেক শিশুরই হাম হতে পারে। হাসপাতাল বা ক্লিনিকে শিশুদের নিয়ে যাওয়ার পর তাদের হাম হবার ঝুঁকি কতটুকু তা জানার জন্য আন্তর্জাতিক উদরাময় গবেষণা কেন্দ্র গবেষণা করছে। কয়েকদিন আগে আপনার বাসার পাশেই একটি শিশুর হাম হয়েছিল। যদিও আপনার বাচ্চার হাম হয়নি, তবুও হামে ভোগা শিশুটির ও আপনার বাচ্চার অল্পদিনের মধ্যে হাসপাতালে যাওয়ার সব খবরাখবর তুলনা করে হাসপাতালে গেলে হাম হওয়ার ঝুঁকি বাড়ে কিনা, তা আমরা নির্ণয় করতে পারব। আপনি যদি আপনার শিশুকে এই গবেষণায় অংশগ্রহন করতে চান, তাহলে আপনাকে আমরা নীচে উল্লেখিত কিছু প্রশ্ন করব ও আপনার শিশুকে পরীক্ষা করব।

একজন স্বাস্থ্যকর্মী আপনার শিশুর বর্তমান ও অতীত স্বাস্থ্য, আপনার পারিবারিক ও আর্থিক অবস্থা এবং আপনার শিশু হাম রোগীর কাছে গিয়েছিল কিনা ইত্যাদি বিষয়ে কিছু কথা বলবে। তাতে আপনার ১৫ মিনিট সময় নষ্ট হতে পারে। একই স্বাস্থ্যকর্মী আপনার শিশুর ওজন, উচ্চতা ও হাতের মাপ নিবে এবং একজন চিকিৎসক তাকে খুব ভালভাবে পরীক্ষা করবে। সাথে সাথে আপনার শিশুকে বিনামূল্যে হাম ও অন্যান্য জীবন রক্ষাকারী রোগের জন্য টীকা দেয়া হবে।

আপনি যে কোন সময় এই গবেষণা থেকে আপনার শিশুকে বের করে নিতে পারবেন। আপনার শিশুকে এ গবেষণা থেকে বের করে নিলেও তার প্রয়োজনীয় টীকা দেয়া হবে।

এই গবেষণায় আপনার শিশুকে অংশগ্রহন করাতে চাইলে নীচে স্বাক্ষর করুন / বাম হাতের বৃদ্ধাঙ্গুলির ছাপ দিন।

গবেষণাকারীর স্বাক্ষর

অভিভাবকের স্বাক্ষর/বাম হাতের বৃদ্ধাঙ্গুলির ছাপ

সাক্ষীর স্বাক্ষর

## Appendix

## QUESTIONNAIRE (FOR CASES & CONTROLS)

1. ID number: ____/____/____/
   (Items 2-4 will apply to measles cases & hospital controls)
2. Hospital number : ____/____/____/____/____/____/____/
3. Recruitment hospital: DSH ____ ICDDR,B ____
   OPD ____ Observation ward ____ IPD ____ ICU ____
4. Date of recruitment: ____/____/____/ (date/month/year)
5. Interviewer's code: ____/____/
6. Child's name: _____
7. Child's date of birth: ____/____/____/ Age ____/____/ months
   child's birthplace: home ____ hospital ____
   born pre-term: no ____ yes ____ low birth weight: no ____ yes ____
8. Sex : M ____ F ____
9. Feeding history: Ever breast-fed: no _____ yes _____
   stopped _____ continuing _____
   if stopped, when: ____/____/ months of age
   Supplementary foods: no _____ yes _____
   if yes, what: milk _____ cereals _____ rice-curry _____
   when started: ____/____/ months of age
10. Respondent's age: ____/____/ years
    respondent's relationship to the child: mother ____ other(state)_____
11. Head of Household's name:_____
    HHH's relationship to child: father____ other (state)_____
12. Occupation of the Head of the Household: _____
13. Occupation of mother: _____
14. Education of the Head of the Household (years of schooling):____/____/
15. Mother's education (years of schooling):____/____/
16. Family income per month: taka ____/____/____/____/____/
17. Type of construction of house: floor _____
    roof _____
    wall _____
18. Number of rooms in the house: ____/____/
19. Number of persons more than 12 years of age: ____/____/
20. Number of children (≤12 years): ____/____/
21. Number of children sleeping together in the same room with the child: ____/
22. For how long you lived in Dhaka ? ____/____/ years
    where did you live before ? _____
23. Address: house no _____ road _____ road no_____
    locality _____ thana _____
    description of the direction of the house in relation to the nearest main road or a important landmark _____
    _____
    _____
24. Distance of the health facility from the home: ____/____/ miles
25. Time required to travel to the health facility: ____/____/____/ minutes
    method of transport _____
26. Date of onset of measles rash (for cases): ____/____/____/

27. Signs and symptoms of illness (during interview):

| | | | Duration (days) |
|---|---|---|---|
| rash : | No___ | Yes___ | ____/____/ |
| fever : | No___ | Yes___ | ____/____/ |
| coryza : | No___ | Yes___ | ____/____/ |
| cough : | No___ | Yes___ | ____/____/ |
| respiratory hurry: | No___ | Yes___ | ____/____/ |
| respiratory distress: | No___ | Yes___ | ____/____/ |
| conjunctivitis: | No___ | Yes___ | ____/____/ |
| stomatitis : | No___ | Yes___ | ____/____/ |
| watery diarrhea: | No___ | Yes___ | ____/____/ |
| mucoid diarrhea: | No___ | Yes___ | ____/____/ |
| bloody diarrhea: | No___ | Yes___ | ____/____/ |
| anorexia : | No___ | Yes___ | ____/____/ |
| oedema : | No___ | Yes___ | ____/____/ |

28. Do you remember the child meeting another child with measles?

No ____ Yes ____

if yes, date of exposure ____/____/____/

sex of index case M ____ F ____

age of index case ____/ ____/ months

index case attends school? Yes ____ No ____

place of exposure:

home ____ neighbor ____

school ____ state which _____

health center ____ OPD/IPD _____

state which health centre _____

private clinic ____ OPD/IPD _____

state which private clinic _____

hospital ____ OPD/IPD

state which hospital _____

different locality ____ state which _____

unknown ____

29. Do any people in your household have measles ?

No ____ Yes ____ if yes, date of onset ____/____/____/ of the first case

sex M ____ F ____ age ____/ ____/____/ months

survived ____ died ____

date of onset ____/____/____/ of the second case

sex M ____ F ____ age ____/ ____/____/ months

survived ____ died ____

date of onset ____/____/____/ of the third case

sex M ____ F ____ age ____/ ____/____/ months

survived ____ died ____

30. When was the most recent visit before this one that this child visited a health facility ? (List dates of visits and reasons for each visits, starting with the most recent visit and working backwards for the 3 months prior to this visit. Ask about visits where the child was accompanying other persons to a health facility as well. Check prescriptions or vaccination cards or clinic records.)

| Date | | | Health facility | Reason | Documented or based on history |
|---|---|---|---|---|---|
| | | | | | |
| | | | | | |
| | | | | | |

31. Vaccination history:

| Vaccine | Children with cards Dates | | | Children without cards Yes | No |
|---|---|---|---|---|---|
| BCG | | | | | |
| DPT 1 | | | | | |
| Polio 1 | | | | | |
| DPT 2 | | | | | |
| Plio 2 | | | | | |
| DPT 3 | | | | | |
| Polio 3 | | | | | |
| Measles | | | | | |

name of the clinic of measles vaccination: _____
date of measles vaccination: ____/____/____/ (for children without cards: from clinic records, if available).

32. Maternal tetanus toxoid immunization history:

TT 1___ TT 2___ TT3___ TT4___ TT5__

33. Blood sample taken? venous ____ capillary____ none ____

34. Saliva sample taken? yes ___ no ___