While countries in South Asia are making considerable progress in achieving the 4th Millennium Development Goal, that is, to reduce under-five mortality rates by two-thirds, reaching the finish line remains a challenge for most countries. Slow reductions in neonatal mortality are largely to blame for this. Public health programmes find it difficult to appropriately address the challenges presented by these deaths in the first month of life, which now constitute more than half of the deaths among under-five children in many of the South Asian countries.

These young infants often die within days of birth without ever being to a doctor, leaving behind no clues as to their causes of deaths. This paucity of information is one of the main obstacles to the development of appropriate interventions to reduce neonatal mortality. Infection is often speculated, but we nonetheless cannot identify the causal agents of the infections, making the planning and designing of comprehensive interventions tricky.

This is where ANISA comes in. It is a rigorous study across 4 community-based sites in Bangladesh, India and Pakistan following mothers through pregnancy and delivery so that the newborns can be monitored from birth. Community health workers are working in many communities in these three countries, assessing the babies immediately after birth at home and then another 9 times until the babies are 2-months old. If any sign of infections are seen in these babies, they are immediately sent to the study facilities where these sick babies are seen by ANISA-trained study doctors who decide whether they meet ANISA eligibility criteria, and provide treatment. Besides saving the lives of numerous neonates, this will enable us to improve our understanding of the aetiology of infections prevalent among neonates in South Asian communities.

ANISA, for the first time, is also bringing state-of-the-art technologies to the places where they are most needed and challenging the usual scenario of irrational distribution of technologies compared to the burden of ill-health and premature deaths. The extensive knowledge that this study will provide us will guide future treatment policies and help pave the way towards saving the lives of our children.
ANISA is being made possible by the collaboration of different organizations across several countries. It is this pleasant companionship between multidisciplinary groups that is vital for the achievement of ANISA goals.
**Collaborators**

- **University of Toronto**
  - SickKids Hospital
  - Data Analysis
  - Technical Support

- **ICDDR,B**
  - Data Management
  - Technical Support
  - Fund Management

- **CHILD HEALTH RESEARCH FOUNDATION**
  - Dhaka, Bangladesh
  - Overall Programme Co-ordination
  - Study Design and Development of Study Tools
  - Administration/Contracts
  - Financial Control
  - Research Dissemination

- **WHO**
  - Monitoring (Field)
  - Technical Support (Field)

- **OXFORD**
  - Monitoring (Lab)
  - Technical Support (Lab)

- **CDC**
  - Monitoring (Field & Lab)
  - Technical Support (Lab)
  - Data Analysis

- **ICDDR,B**
  - Data Management
  - Technical Support
  - Fund Management

- **MoH, GoB**

- **JHU**

- **Projahnmo**

- **Shimantik**

- **BANGLADESH | Sylhet**

- **INDIA | Shivgarh**

- **PAKISTAN | AKU**

- **URBAN**

- **RURAL**
In spite of a substantial decline in under 5 mortality over the past few decades, there has been little progress in addressing neonatal mortality, especially in developing countries. ANISA aims to produce vital data that can be used to reduce neonatal mortality by bridging the gap between modern diagnostic technology and resource-poor settings in an unparalleled fashion.

**INFECTION** accounts for an estimated 1.4 million neonatal deaths worldwide each year. About 60% of the deaths due to infection occur in the **FIRST WEEK** of life and the risks persist up to the 2nd month of life leaving an extremely narrow opportunity to intervene.

**NEONATAL MORTALITY** accounts for 3.6 million deaths, about 40-50% of under-five deaths in the world, annually.
What will ANISA do?

ANISA will determine the causes of neonatal sepsis, their population-based incidence and antibiotic resistance patterns in Bangladesh, India and Pakistan through community-based surveillance by using both standard and cutting edge diagnostic tests. ANISA also aims to identify the risk factors associated with infections and to describe clinical signs of neonatal infections.

and how? ANISA will ---

1. Conduct population-based surveillance of all pregnancies
2. Identify suspected cases of infections in young infants as early as possible
3. Collect blood, respiratory and/or CSF specimens from suspected cases
4. Determine bacterial aetiology by culture and TaqMan Low Density Array (TLDA)
5. Determine viral aetiology by TLDA
6. Determine antibiotic susceptibility patterns of bacterial isolates
7. Do qualitative controls for all pathogens
8. Maintain strict internal and external quality control and assurance
ANISA Community Health Workers (CHWs), with prior informed consent, visit the households of women of reproductive age to identify any new pregnancy. The mothers are offered the standard package of antenatal care available in their respective countries including distribution of iron, folic acid, counseling on immunization, birth preparedness, essential newborn care, and promotion of knowledge about danger signs in both mothers and newborns for appropriate care-seeking. Information is collected to understand the demographics and potential risk factors. All deliveries are reported and a postnatal visit is scheduled as soon as possible within the first 24 hours to ensure detection of danger signs without delay.

PREGNANCY SURVEILLANCE

Since most neonates die within days of birth without ever being to a doctor, ANISA aims to intervene in the narrow window of opportunity by monitoring mothers from the start of their pregnancies and determining expected delivery dates. Careful observation of pregnancy also serves to reveal the risk factors associated with neonatal morbidity and mortality.
SEPSIS SURVEILLANCE

ANISA monitors newborn babies for clinical signs indicating infections. Customized CHW-visit-schedule allows prompt detection of signs of neonatal sepsis; at the same time it prevents excessive interventions and overburdening of CHWs and the caregivers. All newborn babies are visited within 24 hours and the subsequent surveillance is based on regular, scheduled household visits. CHWs refer sick infants to the nearest designated facilities for physician’s evaluation, care and specimen collection. In addition to CHW's referred babies, the physicians at designated ANISA facilities also receive and assess the self-referred cases.
REFERRAL COMPLIANCE

Through their regular visits, the ANISA CHWs build a rapport with the mothers and the community as a whole. Consequently, most of the referrals to the study physicians are complied with, which by itself is one of the indicators of success.

PHYSICIAN’S ASSESSMENT

Designated study physicians assess the referred neonates who are enrolled if upon meeting the ANISA inclusion criteria after informed consent. This is followed by specimen collection briefly before treatment is provided to the infant.
After assessment and shortly before treatment, blood, respiratory and, if necessary, CSF specimens are collected from the neonates. This is one of the vital steps in the study and the maintenance of quality in terms of volume and purity is paramount for fulfilling ANISA’s goal of revealing aetiology of neonatal infections.
In addition to ensuring good laboratory practice in specimen collection and processing, ANISA has coupled conventional and sophisticated new generation molecular techniques to identify pathogens causing sepsis in newborns. Considering the limited volume of specimens and the possibility of a wide spectrum of infection causing organisms, ANISA has picked up a diagnostic method, TLDA, that can altogether identity 32 different pathogens within a few hours using minimal amount of blood and respiratory specimens.

With the objective of comprehensively covering a wide range of pathogens that commonly cause infections in South Asian newborns, the ANISA Technical Advisory Group has designed specific TaqMan Array Cards for this purpose.
Submission of LOI April 2009

Approval of Proposal October 2009

Phase 1 Method development December 2009 to May 2010

Submission of full proposal June 2009

Approval of phase 2 July 2010

Obtain IRB with FWA September 2010

Preparatory activities for field sites October 2010

Assessment of site initiation November 2010

Initiate Pilot phase July 2011

Centralized training March to June 2011

Approval of phase 2

Road Map ANISA
Sites were selected based on 1) proven track record of conducting community-based surveillance to identify and follow up suspected infection cases, 2) high neonatal mortality, 3) known denominators and potential to yield at least 1000 cases of suspected infections per year, 4) the ability to collect and transport specimens, 5) capacity to conduct laboratory studies and preserve specimens and isolates for further testing and analysis, and 6) willingness to use new diagnostic platforms and participate in external quality control (QC) and quality assurance.
Background

ANISA Bangladesh site is located in the rural areas of Kanaighat and Zakiganj in Sylhet, which is 300 kilometers North-East of capital city Dhaka. This site is run by Projahnmo (Program to Advance the Health of Newborns and Mothers), a partnership programme of Ministry of Health and Family Welfare of Bangladesh, ICDDR,B, Dhaka Shishu Hospital, Child Health Research Foundation, Shimantik and Johns Hopkins University. Projahnmo established Community-based surveillance in this area during 2002 and is still maintaining them through different projects. Specimens from this site will be collected in Government health facilities before being transported to CHRF laboratories in Sylhet and Dhaka for processing.
Background

ANISA India site is located at Shivgarh village in Uttar Pradesh state which accounts for a quarter of India’s neonatal deaths. The Shivgarh site was established in 2003 with support from USAID, N. Delhi Mission, Washington Mission and Saving Newborn Lives Initiative of Save the Children with a grant from the Bill & Melinda Gates Foundation. The Shivgarh site has maintained a Demographic Surveillance System (DSS) along with geo-referenced data points and relational database management system since 2003 which is serving as a reference gold standard for the Bill & Melinda Gates Foundation funded GC13 Health Metrics Project. In this site, specimens will be collected in Government health facilities before being transported to the “Empowerment Lab” at Lucknow for processing.

Principal Investigator

Dr. Vishwajeet Kumar is a physician with advanced public health training from Johns Hopkins Bloomberg School of Public Health. His scientific work is centred on developing and evaluating innovative models for improving maternal and newborn health. He has been working in this site for last 8 years.

Site description

<table>
<thead>
<tr>
<th>Study area</th>
<th>Shivgarh</th>
</tr>
</thead>
<tbody>
<tr>
<td>Population of study area</td>
<td>350,000</td>
</tr>
<tr>
<td>Birth cohort (over 2 years)</td>
<td>14,000</td>
</tr>
<tr>
<td>Neonatal mortality</td>
<td>66/1,000 live births</td>
</tr>
<tr>
<td>Number of CHW</td>
<td>One CHW for every 7,000 population</td>
</tr>
</tbody>
</table>

Expected Numbers

| Neonates with potential infections (10%) | 1400 |
| Enrollment (over 2 years, 70% compliance) | 980 |
| Bacterial isolates (5% of enrolled neonates) | 49 |
| Viral isolate (30% of enrolled neonates) | 294 |
| Population based viral incidence rate | 21/100 live birth |
| Bacterial isolates population based bacterial incidence rate | 3.5/1000 live birth |
Background

ANISA study sites at urban Pakistan are low-income communities with high neonatal mortality rates located in peri-urban coastal fishing villages in Karachi (4 sites) about an hour drive from the Aga Khan University (AKU) campus and one urban squatter settlement (Bilal Colony) at a distance of 30 minutes from AKU. Among them Bilal Colony, Rehri Goth and Ibrahim Hyderi have extensive experience with neonatal research, having served as the only community sites for the Young Infant Clinical Signs Study II. Surveillance for pregnancy and newborn illnesses was established in 2003. All five sites are currently serving as the sites for Saving Newborn Lives (SNL)-sponsored antibiotic therapy trial for neonatal infections (with support from the Bill & Melinda Gates Foundation).

Pakistan
Urban site - Karachi

Principal Investigator
Dr. Anita Zaidi is Professor and Chair, Department of Paediatrics and Child Health at Aga Khan University (AKU). Dr. Anita’s work has focused on neonatal infections, epidemiology of childhood vaccine-preventable illnesses and improving diagnostic and molecular methods of detecting infectious agents in children.

Site description

<table>
<thead>
<tr>
<th>Study area</th>
<th>Bilal colony, Ibrahim Hyderi Colony, Rehri Goth, Ali Akber Shah Goth and Bhains Colony</th>
</tr>
</thead>
<tbody>
<tr>
<td>Population of study area</td>
<td>270,000</td>
</tr>
<tr>
<td>Birth cohort (over 2 years)</td>
<td>19,000</td>
</tr>
<tr>
<td>Neonatal mortality</td>
<td>34.4/1,000 live births will update</td>
</tr>
<tr>
<td>Number of CHW</td>
<td>One CHW for every 1617 population</td>
</tr>
</tbody>
</table>

Expected Numbers

| Neonates with potential infections (8%) | 1520 |
| Enrollment (over 2 years, 70% compliance) | 1050 |
| Bacterial isolates (5% of enrolled neonates) | 59 |
| Population based bacterial incidence rate | 3.1/1000 live birth |
| Number of Viral isolate (30% of enrolled neonates) | 315 |
| Population based viral incidence rate | 21/100 live birth |
Background
The rural Pakistan site, Matiari, is located 400 kilometers from Karachi. It has developed a prospective cluster randomized trial focusing on the integrated management of birth asphyxia, low birth weight infants and suspected neonatal infections (especially pneumonia) with oral Amoxicillin. This site has experience of conducting multiple community based surveillance and clinical trial funded by NIH, WHO and BMGF. Specimens will be collected at home by experienced medical teams and transported to field lab for blood culture and the rest to AKU lab for molecular tests.
Centralized Management
The ANISA team has worked hard to ensure that data collectors, study physicians, laboratory staff and other study staff in all four sites use the same standardized, easy to use data forms to capture the data critical for ANISA to achieve its objectives.
ANISA data system

Systems have been designed to transform the paper-based data into electronic data in the form of clean databases accessible to country study teams and also to the ANISA Coordination Team so that analysis can be conducted in real time and progress monitored. The use of paper to record data in the laboratory and to track specimens has been minimized to both avoid errors and ensure that laboratory results are accessible as they become available.

Figure: Development and implementation of data management system coordinated by Data Coordinating centre (CHRF)
Harmonization of clinical and laboratory skills, and interpretation of algorithm and laboratory results are pivotal to generate an appropriate set of data from a geographical and culturally diverse population. To achieve these, ANISA team organized centralized training for CHWs, physicians and laboratory staffs.

**Centralized Training**

**SMS System**

Data collectors and study physicians use text messages over mobile phones to update the databases on select group key information essential for tracking progress in study enrollment, selection of controls and tracking of referral of sick newborns.
Similar to centralized training, use of same diagnostic methods, reagents and consumables is also important to generate interpretable results. Keeping that consideration in mind, ANISA ensured similar diagnostic methods at all the sites, and arranged central procurement and shipping of capital equipments and reagents to the sites.
ANISA Study Profile

- Pregnancy Surveillance
  - 2 monthly home visits By CHW
  - Consent given
  - Pregnancy Not Identified
    - Demographic data
    - Enrolled
    - Visit 1
- Birthassessment
  - Pregnancy Identified
  - Within 24 hrs
  - Basic antenatal intervention package
  - Pregnancy Not Identified
    - Consent given
  - Enrolled
  - Visit 1
  - Pregnancy Identified
    - Consent given
    - Enrolled
- Postnatal follow up visits
  - 2 6 13
  - 20 27 34
  - 41 48 59
- Sepsis screening
- Suspected infection
  - Eligibility criteria Assessed by doctor
  - Eligible
  - Consent taken
  - Specimen collected
  - Treatment
- Blood
  - Blood Culture
  - Cytology
  - Antigen test
  - PCR
  - Bio-banking
- Respiratory specimen
  - Blood Culture
  - PCR
  - Bio-banking
- CSF
  - Blood Culture
  - Antigen test
  - PCR
  - Bio-banking
  - Consent taken
  - Specimen collected
  - Treatment
- Specimen collected
- Treatment

*Will be only collected at Hospital
**Coordination Team**

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The Child Health Research Foundation (CHRF) was initiated by a group of child health researchers of Bangladesh, working with a mission to improve child health in Bangladesh and other countries through research and educational efforts to facilitate the decision making on appropriate child health policy. The idea for setting up CHRF came from the realization of the founding group of researchers that developing countries like Bangladesh often lack the evidence needed for rational and timely policy decisions with the consequent inadequate and improper use of limited resources. Scarcity of evidence, in turn, discourages the government and donors to invest in child health. CHRF has established collaboration with different national and international organizations like USAID, WHO, Johns Hopkins University, ICDDR,B, International Vaccine Institute (IVI), Novartis Vaccine Institute of Global Health and others.

In addition to research, the foundation is also making efforts to work with other non-profit hospitals to facilitate low cost diagnostic services for the poor patients with a vision to i) improve child health, and ii) generate evidences for policy decisions.

Through an association with the Department of Microbiology at Dhaka Shishu Hospital and through a consortium of urban and rural hospitals, it has access to research facilities and is able to inform and improve public health approaches and contribute to scientific and medical knowledge and procedures for application in South Asia.

Through these activities Child Health Research Foundation aims to break the vicious cycle that limits the potential of children in Bangladesh and convert it to a virtuous cycle by making appropriate use of limited resources, produce evidence, facilitate evidence-based policy decisions, shorten the time lag between evidence and implementation and generate interest for increased and sustained investments in child health. Finally, CHRF dreams to contribute in reducing child mortality by creating awareness for appropriate care and care-seeking, ensuring rational treatments, and evidence-based preventive programmes.

### Methods development

#### Preparatory phase

#### Cases and controls enrollment

#### Final analysis and report writing