

# **Science and Technology Project on Integrated Disease Vector Control**

## **Annual Report 2013–14**



### **National Institute of Malaria Research (Indian Council of Medical Research)**

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## An Overview of the Activities

Under the aegis of the Integrated Disease Vector Control (IDVC) project, studies on basic, applied and operational research were continued and carried out during the year 2013–14. These studies are related to Epidemiology, Vector Biology, Vector Control and Parasite Biology. New tools for vector control and testing of plant products for antimalarial and antivector activities and therapeutic efficacy of new antimalarials have been evaluated. Technical support to the State Health Departments and National Vector Borne Disease Control Programme (NVBDCP) is being carried out as routine on priority.

Studies on Vector Biology and Vector Bionomics were undertaken in different topographical areas to understand the disease vectors in the changing ecological context. The study in Bengaluru shows that the villages situated on the banks of the river with sandy beds have very high vector abundance throughout the year compared to the non-reverine areas. Regular monitoring of densities of different malaria vectors was carried out in all the field units. Seasonal investigations on the abundance of dengue vector *Aedes (Stegomyia) albopictus* and *Aedes (Stegomyia) aegypti* in Guwahati metropolis and sub-urban settlements revealed that these species are widely abundant in the region, and breeding habitats were recognised. A door-to-door survey in Hardwar and BHEL township carried out by the Hardwar Field Unit helped the District Authorities to control the dengue cases. At Nadiad, Dengue vectors were found in high density in containers under command area of Sardar Sarovar Dam resulting in increase of dengue and chikungunya cases in that area. In Sundergarh district of Odisha, an *Aedes* mosquitoes survey in arboviral epidemic prone areas was undertaken. In Bengaluru also a dengue outbreak was investigated in villages Salundi and Dhalundi, CHC Jaipura, Mysore Taluka in June 2013.

At Kondagaon district in southern Chhattisgarh, study on the impact of insecticide resistance in malaria vector on the effectiveness of combination of indoor residual spraying (IRS) and long-lasting insecticidal nets (LLINs) was continued. Monitoring of insecticide susceptibility status of malaria vectors was undertaken in 11 districts of selected malaria endemic areas.

Monitoring of the epidemiological impact of ratio of insecticide for IRS was done. The study compared the effectiveness of IRS with DDT on multi-resistance vector population and malaria incidence in comparison to IRS with synthetic pyrethroid. Also a study on new insecticide molecule chlorfenapyr against multi-resistant malaria vectors was done as a new tool to effectively manage the problem of insecticide resistance.

At Panaji (Goa), larvicidal and pupicidal activity of methanol and chloroform extracts of dried leaves of plant G/C (code name) against urban malaria vector *An. stephensi* Liston, filariasis vector *Cx. quinquefasciatus* Say and dengue and chikungunya vector *Ae. aegypti* Linn with promising results and new fractionation of active compound is being done. At Panaji, studies were carried out on proteome of *An. stephensi* and annotation of

genes responsible for these proteins involved in *Plasmodium* infection. Eight putative proteins were identified from the mid gut of female *An. stephensi*. Characterization of proteome of *Ae. aegypti*, was also carried out. In all 1944 and 1912 proteins were identified from the salivary gland and mid gut, respectively.

At Bengaluru, lambda-cyhalothrin 10CS (ICON 10CS) was evaluated in Karnataka which showed extended efficacy up to 12 weeks post-spray. LLIN NetProtect (impregnated with deltamethrin) was evaluated in Saharanpur district of Uttar Pradesh. More than 80% mortality was recorded after 20 washes and drastic reduction in vector density was recorded and community acceptance was high.

WHOPES Phase-III evaluation to compare insecticidal efficacy and community acceptance of long-lasting insecticidal net DuraNet with conventional insecticide-treated net has been completed. Studies are being continued on the field evaluation of NetProtect LLIN (impregnated with deltamethrin) and PermaNet 3.0, a combination LLIN against malaria vectors and impact on malaria incidence. A Phase-III evaluation of new candidate long-lasting insecticidal net DawaPlus 2.0 is being continued for its bio-efficacy, durability and community acceptability. WHOPES sponsored small-scale field testing and evaluation to compare residual efficacy of Pirimiphos-methyl CS (Actellic 30% CS) with Pirimiphos-methyl 50% EC by IRS on different local indoor surfaces are being conducted. A project on remote sensing was undertaken in Uttarakhand to prepare cost-effective model of malaria control, based on the results extracted through Remote Sensing and Geographic Information System. DDT residues in human milk, human blood and adipose tissue from DDT-sprayed and non-sprayed areas were compared. Also, DDT residues in human blood and human milk were determined in pregnancy and after pregnancy to correlate lactation and pregnancy outcome. Study on therapeutic efficacy of ACT (AS + SP) was undertaken along Indo-Bangladesh border in Mizoram (Lunglei district) and Tripura (Gomti district). Further, a randomized controlled trial of Artesunate + Sulphadoxine-Pyrimethamine (ASP) vs ASP + Primaquine for decreasing malaria transmission was completed in the Jharkhand state. A Phase-II/III randomized clinical trial on the efficacy and safety of Artesunate + Sulphadoxine- pyrimethamine and Artesunate + Mefloquine to treat uncomplicated falciparum malaria in pregnancy is ongoing in the Jharkhand state. Comprehensive malaria case management is continuing in low, medium, high and hyperendemic malaria transmission areas. A multi-centric study on the impact of new tools and changing epidemiological patterns of malaria is continuing in Odisha and Tamil Nadu states. The study on the evaluation of the effectiveness of intervention measures on malaria prevalence is being carried out in two tribal districts as Translational Research projects in collaboration with the Govt. of Madhya Pradesh.

Study on heme biosynthesis in malaria parasite was carried out in Bengaluru field unit to study knockouts (KOs) for the first enzyme, δ-aminolevulinate synthase (ALAS), and the last enzyme, ferrochelatase (FC), in the heme-biosynthetic pathway of *P. berghei* (*Pb*). Study on therapeutic efficacy of chloroquine was undertaken in *P. vivax* malaria in Rameswaram Island, Tamil Nadu. Elsewhere, study on the identification of molecular marker(s) for relapse malaria in *P. vivax* to determine the genotype for differentiation between relapse and re-infection was also undertaken.

Two NIH sponsored projects are operational under the International Centre of Excellence for Malaria Research at NIMR Field Units. NIH funded CSCMi project is ongoing in highly endemic districts of Odisha at Rourkela, Chennai and Nadiad field units to study vector bionomics, ecoepidemiology, mapping of malaria cases, and to compare various diagnostic methods. Study on the impact of *P. falciparum* and *P. vivax* ratio on malaria burden is being carried out at Nadiad in Gujarat as a part of multi-centric study on eco-

epidemiology and transmission of complex malaria. In Panaji (Goa), the Field Unit has initiated studies on seasonal vector abundance and prevalence of *Plasmodium* infection in the vectors under NIH sponsored MESA project to study the epidemiology and malaria evolution in South Asia. In addition to these research activities, IDVC Field Units provide technical inputs to strengthen malaria control activities, viz. health education and capacity building measures, mass propagation and distribution of larvivorous fishes, epidemic investigations, epidemiological surveillance, cross-checking of blood smears, and providing technical expertise on LLINs.

I take this opportunity to thank all the Officers Incharge and staff of the Field Units for their contribution and technical support to the National Programme. I also wish to thank ICMR and Ministry of Health and Family Welfare, Govt. of India for providing financial support for this project. I express my sincere thanks to Dr Arun Sharma, Scientist 'F' of the Institute for compiling the report and Publication Division for production of the report.

Neena Valecha  
Director



## Introduction

### Background

Integrated Disease Vector Control (IDVC) project is one of the eight mission mode Science and Technology projects in various fields identified in 1983 by the Scientific Advisor to the then Prime Minister. This project was assigned to the Malaria Research Centre (renamed as National Institute of Malaria Research on 4 November 2005), Delhi (for malaria) and Vector Control Research Centre, Puducherry (for filariasis) in 1985. The IDVC project is being funded jointly by the Indian Council of Medical Research (ICMR) and the Ministry of Health and Family Welfare (MoHFW), Govt. of India. Between 1986 and 1992, Malaria Research Centre (MRC) opened 13 field units at various locations in the country in consultation with the National Anti Malaria Programme, now National Vector Borne Disease Control Programme (NVBDCP). The project was extended up to the eighth plan by the recommendation of the Scientific Advisor to the Prime Minister. Its continuation beyond March 1997 has been done on yearly basis with the approval and financial support from the MoHFW, Govt. of India.

The project successfully demonstrated malaria control in rural, urban, industrial, forest, tribal and coastal areas of the country. In addition to this, the project also successfully demonstrated malaria control through the primary health care system in the country which received appreciation from various agencies. In addition to annual reviews by the Research Advisory Committee (RAC) of IDVC project, Scientific Advisory Committee (SAC) of NIMR, Scientific Advisory Group, Scientific Advisory Board and Governing Body of ICMR, various committees, namely Nitya Anand (1985), Harcharan (1993), Pattanayak (1995) and Rudrappa (1996) periodically reviewed the IDVC project, and made scientific recommendations in view of

the increasing malaria cases in the country and recurrence of epidemics. There is need for continued research to develop and test new technologies for malaria control to be used by NVBDCP. The Scientific Advisory Committee of the MRC which reviewed the annual progress repeatedly also underscored the importance of the regularisation and re-organisation of the IDVC project and made recommendations to effect that from time-to-time. To implement these recommendations, approval of the 52nd Scientific Advisory Board (2000) of the ICMR was taken which endorsed the recommendations made by the Rudrappa Committee on the permanency of long-term extramural projects. Subsequently, the Governing Body of ICMR, New Delhi in 1999 and 2000 recommended that 12 field units located outside Delhi should continue as a permanent activity of MRC and Delhi field unit be closed. And it was further stated that the field units could be shifted as per the needs of the malaria control programme. In continuation, a meeting comprising of the representatives of NAMP/MRC/ICMR under the Chairmanship of Dr S. Pattanayak in the year 2001 discussed the issue of re-organisation of the IDVC project threadbare and recommended the same. The committee also highlighted the areas of applied field research of immediate importance to control malaria in the country.

The Standing Finance Committee of the MoHFW recommended re-organisation of the IDVC project in its meeting on 10 August 2005. Following the re-organisation, 10 field units are functioning under this project in different states of the country. Keeping in view the recommendations of various committees and continued need for research support to malaria programme in the country, NVBDCP and NIMR have jointly identified priority areas to realise the gains in malaria control through research.

## Objectives

All the field units have a common research programme of providing support to NVBDCP. However, depending on the local malaria situation, research interest of the field unit and recommendations of the Scientific Advisory Committees of the field unit/NIMR, each field unit has a specific research programme.

The research programmes have been planned up to the end of tenth five year plan. Further planning of research beyond the 10th plan period will be done in consultation with NVBDCP, State Health Departments, Scientific and Research Advisory Committees of NIMR and considering the prevailing malaria situation at a given period.

## Common research programme

- (i) Basic and applied research on vector biology and transmission dynamics of malaria in different ecosystems in order to provide inputs to malaria control programme.
- (ii) Technical support in malaria control activities of the state health programme with reference to the following activities:
  - Preparation of annual malaria action plan
  - Insecticide resistance monitoring
  - Evaluation of insecticide spray quality
  - Therapeutic efficacy of antimalarials
  - Capacity building in the field of malaria entomology, microscopy and surveillance
  - Evaluation of strategies (ITN, LN, larvivorous fish, blister pack, etc.) as and when required by NVBDCP.
- (iii) Evaluation of new products (insecticides, drugs, diagnostic, etc.)
- (iv) Epidemic investigations for rapid response and management.

## Nodal department and collaborating agency

Indian Council of Medical Research (ICMR), V. Ramalingaswami Bhawan, Ansari Nagar, New Delhi-110 029.

## Institute

National Institute of Malaria Research (ICMR), Sector 8, Dwarka, New Delhi-110 077.

## Collaborating organisation

National Vector Borne Disease Control Programme (NVBDCP), 22 Sham Nath Marg, Delhi-110 054.

## Budget

Grant received from ICMR during the financial year 2013–14 was ₹ 112.5 million and expenditure had been shown in Fig. 1.

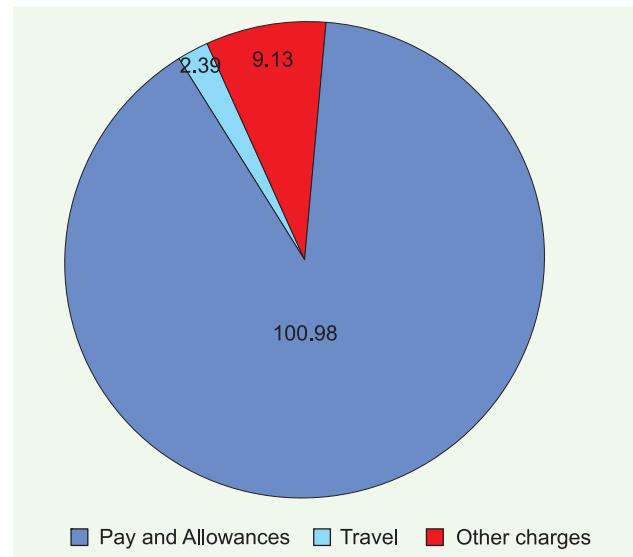


Fig. 1: Expenditure incurred during 2013–14 (Pay & Allowances: ₹ 100.98 million; Travel: ₹ 2.39 million; Other charges: ₹ 9.13 million).



## Highlights of the Research Activities

### 2.1 Bengaluru (Karnataka)

- Vector biology and bionomics study in the Upper Krishna project area showed that the villages situated on the banks of the river with sandy beds were having high vector abundance throughout the year as compared with non-riverine villages.
- Field trial of lambda-cyhalothrin 10CS (ICON 10CS) in Karnataka showed extended efficacy up to 12 weeks post-spray.
- The larvivorous fish, *Gambusia affinis* and *Poecilia reticulata* were found effective in containing JE vectors in Gorakhpur district (Uttar Pradesh). Here rice fields support vector breeding only during monsoon.
- Field trial of two formulations of alpha-cypermethrin against *An. stephensi* showed superior efficacy of WG formulation over WP formulation on all the surfaces tested.
- Phase II household surveys were carried out in six districts of Karnataka for understanding the functioning of the programme implementation.
- $\delta$ -aminolevulinate synthase (ALAS) is an important enzyme for heme synthesis. This would be an important candidate for drug and vaccine candidate.
- Transcriptome analysis is being done to understand how cerebral malaria occurs in *Pf* cases.
- Two larvivorous fish, *Poecilia reticulata* and *Gambusia affinis* are being used in malaria control in Karnataka and the study has been extended to the northern districts of Karnataka.
- Transmission assessment survey was carried out in Udupi district. ICT result indicated that filariasis transmission in this district is very low and qualify for elimination.
- One dengue and one malaria outbreak

investigations were carried out. Dengue outbreak was investigated in Villages Salundi and Dhalundi, CHC Jaipura, Mysore Taluka in June 2013.

### 2.2 Chennai (Tamil Nadu)

- *In vivo* studies on the therapeutic efficacy of chloroquine against *P. vivax* malaria were undertaken in Rameswaram Island, Tamil Nadu.
- Studies were undertaken on the impact of new control tools and changing patterns in two PHCs and two Zones in Ramanathapuram district and Corporation of Chennai, respectively.
- Assessment of malaria gametocytaemia with duration of symptoms: A potential programme monitoring tool for delay in seeking treatment was carried out among malaria patients attending the clinic.
- Field studies on transmission of *An. stephensi* in potential breeding habitats; recording of micro environmental temperature and humidity of the breeding/resting habitats besides, eco-epidemiology of malaria comprising: (a) community study to determine the incidence/prevalence rate of malaria including asymptomatic malaria; and (b) clinic study to investigate the impact of complex malaria on disease outcome in symptomatic individuals were undertaken as part of the NIH project entitled, 'Centre for the Study of Complex Malaria in India (CSCMi)'.
- Identification of the molecular marker(s) for relapse malaria in *P. vivax* to genotype for differentiation between relapse and re-infection was undertaken.
- Technical support was provided to various institutes/colleges/Govt. agencies and

collaborative research studies were also undertaken with NIMR, New Delhi.

- Malaria clinic continued to function, catering to the needs of the public by providing early diagnosis and prompt treatment.

### **2.3 Guwahati (Assam)**

- Dengue as emerging arboviral infection in northeast India, investigations on seasonal abundance of *Aedes (Stegomyia) albopictus* and *Aedes (Stegomyia) aegypti* in Guwahati metropolis and suburban settlements revealed that both these mosquito species—the implicated vectors are widely abundant in the region and breed in a variety of habitats including discarded tyres, cement tanks, used battery boxes, etc.
- To track emergence of artemisinin resistance, study on therapeutic efficacy of artesunate + SP was undertaken along the Indo-Bangladesh border in Mizoram (Lunglei district) and Tripura (Gomti district). Based on extended follow up study, > 20% treatment failure cases to ACT (AS+SP) were recorded in both the study locations, but there was no evidence of reduced sensitivity to artemisinin, and failure was attributed to SP component. Accordingly, the drug policy has now been changed to artemether + lumefantrine combination in northeast India.
- DDT is being used ever since inception of the malaria control programme in northeast India. A controlled study of possible adverse effects of DDT on human reproductive health was undertaken with special reference to lactation and pregnancy outcome.
- There is an increasing evidence for significant differences in concentrations of DDT residues between population groups in DDT-sprayed and unsprayed areas.
- With rapid population explosion and associated development projects in north-eastern states of India, the mosquito fauna surveys in varied ecological conditions revealed that populations of *An. minimus* are diminishing, and the niche is being occupied by *An. culicifacies*, the emerging vector resistant to most of the insecticides.
- Other activities included technical inputs to strengthen the malaria control activities specific to northeastern region, viz. health

education and capacity building measures, mass propagation and distribution of larvivorous fish (Guppy and *Gambusia*) in town areas, and in providing technical expertise on long-lasting insecticidal nets.

### **2.4 Hardwar (Uttarakhand)**

- A study is being carried out for assessment of the efficiency of ASHA workers in delivery of health services. A questionnaire was prepared and survey has been initiated in Laksar PHC of District Hardwar.
- A Remote Sensing and Geographical Information System (GIS) based approach for mapping, monitoring, prediction of mosquitogenic potential and probable determinants of malaria in District Hardwar of Uttarakhand state: Using LISS IV satellite image of various landscape features and statistics of four malarious and two non-malarious areas data were generated. Spot maps of two villages have been prepared for ground truth verification. Compilation and analysis of data of malarious and non malarious villages are in progress.
- During a focal outbreak of dengue in Hardwar during 2013, a door-to-door survey was carried out in houses and peridomestic areas in Hardwar and BHEL township to find out breeding sites of dengue vector. During 2013, a total of 1310 cases of dengue were recorded by rapid immuno-chromatographic test kit, of which 401 cases were confirmed by ELISA test by the district authorities. Four deaths due to dengue were reported.
- Field Unit is working on industrial malaria control since 1986 and successfully controlled malaria in BHEL complex. During April 2013 to March 2014, a total of 1641 blood slides were examined, out of which 38 slides were found positive for *P. vivax*, SPR being 2.3. One fish stock of *Poecilia reticulata* has been established in BHEL township.
- Evaluated NetProtect LLIN (impregnated with deltamethrin) against malaria vector in district Saharanpur of Uttar Pradesh. More than 80% mortality was recorded in mosquitoes exposed to NetProtect LLIN washed up to 20 times. Reduction in vector anopheline density was recorded in all the villages where NetProtect nets were distributed. Community acceptance

of NetProtect LLIN was high.

- Technical support to the programme was also provided. Cross-checking of blood slides received from District Hardwar for malaria parasites was performed. Entomological surveillance of dengue vector in District Hardwar was carried out.

## **2.5 Jabalpur (Madhya Pradesh)**

- The assessment of durability of LifeNet in comparison with NetProtect and PermaNet 2.0 LNs/long-lasting insecticide-treated nets was carried out in CHC Kundam of Jabalpur, Madhya Pradesh. Baseline chemical assay for insecticidal activity carried out before distribution showed that the deltamethrin content in all the three LINs are within WHO specification tolerance limits. After distribution of all the three types of nets, the adverse events reported by the net users on week 1 was 17–47% mainly skin itching, facial burning, and eye irritation. Susceptibility tests revealed >97% mortality of *An. culicifacies* against deltamethrin 0.5%.
- The study on evaluation of the effectiveness of intensive intervention measures on malaria prevalence was carried out in two tribal districts, Dindori and Balaghat as translational research project funded by ICMR in collaboration with the Govt. of Madhya Pradesh. While in Dindori there was a sharp decline in malaria prevalence from 2011 onwards and in Balaghat the decreasing trend was observed only in 2013 in villages where Zero Vector Durable Lining (ZVDL) was installed.
- Study on bionomics of malaria vectors was initiated in the month of October 2013 in two districts of Chhattisgarh state. Korea district showed relatively higher man hour density of *An. fluviatilis* than Bastar. *An. culicifacies* in Bastar district is resistant to DDT, malathion and synthetic pyrethroid while, in Korea *An. culicifacies* is susceptible to alpha-cypermethrin.
- On the request of Govt. of Madhya Pradesh, malaria outbreak investigation was carried out in Katni and Anooppur districts of M.P.
- Two training workshops on malaria and other vector borne diseases were organized for Medical Officers from various districts of

Madhya Pradesh during the year.

## **2.6 Nadiad (Gujarat)**

- The mosquito breeding in containers was found very high in Phase II districts under command area of Sardar Sarovar Dam as a result dengue and chikungunya cases have increased.
- A pilot epidemiological study indicated no significant difference between microscopy and PCR. Seasonal transmission, infections possibly are relapsing vivax in summer possibly once in past 12 months.
- Considering persistence, efficacy and impact on the elements of vectorial potential of both formulations, pirimiphos-methyl CS was found more effective than pirimiphos-methyl EC.
- In order to know the current status of intervention coverage and challenges faced by the malaria control programme at community level household surveys in four districts of Gujarat have been completed.
- Assessed disease prevalence and control activities undertaken by VBD teams in different areas and organized trainings of medical and nursing students and laboratory technicians.

## **2.7 Panaji (Goa)**

- Studies were carried out on proteome of *An. stephensi* and annotation of genes responsible for these proteins with particular focus on those involved in *Plasmodium* infection. Eight putative proteins were identified from the mid gut of female *An. stephensi*, which are involved in vector-parasite interactions.
- Studies were carried out on the characterization of proteome of *Ae. aegypti*. In all, 944 and 1912 proteins were identified from the salivary gland and mid gut, respectively using in-gel digestion approach. About 628 proteins were found common in both salivary gland and mid gut. Further, analysis is in progress.
- Under NIH sponsored MESA project, the field unit has initiated studies on seasonal vector abundance and prevalence of *Plasmodium* infection in vectors. Cyclic colony of *An. stephensi* (type form) has been established for vector competence studies.

- Larvicidal and pupicidal activity of methanol and chloroform extracts of dried leaves of plant code named GIC was evaluated against urban malaria vector *An. stephensi* Liston, filariasis vector *Cx. quinquefasciatus* Say and dengue and chikungunya vector *Ae. aegypti* Linn. with promising results. Fractionation of active compounds is in progress.
  - Bioassays with LLINs revealed that up to 21 months, there was absolute mortality in the vectors after washing the nets which after 24 months of usage and 7 washes declined to 73.5% in the laboratory bred *An. stephensi*, 57.5 and 87.5% in the case of *Cx. quinquefasciatus* and *Ae. aegypti*, respectively. Without washing the LLINs, the percent mortalities remained at 95, 97.5 and 97.5% in *An. stephensi*, *Cx. quinquefasciatus* and *Ae. aegypti*, respectively.
  - Insecticide susceptibility studies with DDT, malathion and deltamethrin revealed that mortality in *An. stephensi* females was 53.2, 73 and 92% respectively; in *Cx. quinquefasciatus* mortality was 25, 40 and 93% respectively; in *Ae. aegypti* it was 87.2, 83.7 and 100% respectively; whereas, in *Ae. albopictus* mortality was 15.95% against DDT and 80% against malathion.
  - Vector monitoring and evaluation is being carried out in six high risk PHCs/UHCs in Goa and based on which action is being taken to control malaria transmission by the state health services.
  - Field Unit undertakes monitoring of vector populations at seaport and international airport in Vasco-da-Gama, Goa as per International Health in Ports of entry and ground crossings and in 400 m perimeter of these establishments. Reports were submitted for interventions to the Port Health Officer and state health authorities on regular basis.
  - Twelve training programmes were organized for medical officers, students, laboratory and field staff.
- 2.8 Raipur (Chhattisgarh)**
- Study on the implications of insecticide resistance on the effectiveness of combination of indoor residual spraying (IRS) and long-lasting insecticidal nets (LLINs) is being carried out. The study is exploring appropriate strategy to manage insecticide resistance for effective malaria control.
  - Monitoring the epidemiological impact of rotation of insecticides for indoor residual spraying (IRS) in malaria endemic areas was done. The study compared the effectiveness of IRS with DDT on multi-resistance vector population and malaria incidence in comparison to IRS with synthetic pyrethroid.
  - Monitoring of resistance in malaria vectors to various insecticides in 11 districts of the state was done to help the state government to plan and rationalize the insecticide spraying strategy.
  - Evaluation of a new insecticide molecule chlorfenapyr with novel mode of action against multi-resistant malaria vectors was done to use it as a tool to effectively manage the problem of insecticide resistance and malaria control in tribal forested areas.
  - Large-scale, community based, household randomized trial of long-lasting nets treated with alpha-cypermethrin, deltamethrin and permethrin was carried out.
  - Induction and refresher training was organized for the malaria laboratory technicians from various health facilities all over the state.

## 2.9 Ranchi (Jharkhand)

- Mosquito fauna survey was undertaken with particular reference to anophelines in West Singhbhum district of Jharkhand state.
- Field evaluation of NetProtect LLIN (impregnated with deltamethrin) against malaria vectors and its impact on malaria incidence was completed in Jharkhand state.
- A randomized controlled trial of artesunate + sulphadoxine-pyrimethamine (ASP) vs ASP + primaquine for decreasing malaria transmission was completed in Jharkhand state.
- Filariasis survey was carried out in the tribal population of Santhal Pargana (Dumka, Godda, Pakur, Sahibganj and Jamtara districts) of Jharkhand state.
- MDA assessment as per NVBDCP guidelines for elimination of lymphatic filariasis was conducted in Deoghar and Godda districts of Jharkhand state.
- Monitoring of insecticide resistance in malaria vectors was undertaken in different ecotypes of different districts of Jharkhand state and the

results were provided to the SPO.

- Technical support was provided to NVBDCP in the areas of malaria microscopic surveillance and capacity building on entomological aspects.
- Diagnostic and treatment services were provided to malaria and filarial patients attending the field unit clinic.

## 2.10 Rourkela (Odisha)

- The MMV funded Comprehensive Case Management Programme was launched in collaboration with the Government of Odisha, in four districts, namely Bolangir, Dhenkanal, Angul and Kandhamal categorized as low, moderate, high and hyper-endemic to malaria, respectively.
- Studies on eco-epidemiology and transmission of complex malaria were continued in highly endemic *P. falciparum* dominated Sundergarh district of Odisha as a part of multi-centric

study being carried out simultaneously in other centres under NIH funded CSCMi project.

- The Phase-II/III randomized clinical trial on the efficacy and safety of artesunate + sulphadoxine-pyrimethamine and artesunate + mefloquine to treat uncomplicated falciparum malaria in pregnancy was continued.
- Study on the assessment of treatment seeking behaviour, LLIN usage and IRS acceptance by tribal communities of Odisha was continued.
- Study on the impact of new tools and changing patterns in the control of malaria in Odisha was continued.
- A study on *Aedes* mosquitoes in arboviral epidemic prone areas of Sundergarh district, Odisha was undertaken.
- A study on insecticide susceptibility status of malaria vectors in six western districts of Odisha was undertaken.





# 3

## Detailed Progress of the Work Done



- Bengaluru (Karnataka)
- Chennai (Tamil Nadu)
- Guwahati (Assam)
- Hardwar (Uttarakhand)
- Jabalpur (Madhya Pradesh)

- Nadiad (Gujarat)
- Panaji (Goa)
- Raipur (Chhattisgarh)
- Ranchi (Jharkhand)
- Rourkela (Odisha)





## 3.1

### Bengaluru (Karnataka)

#### Delineation of malaria vector(s) in Upper Krishna Project area, Karnataka for effective malaria control

Upper Krishna Project (UKP) area in Karnataka is endemic for malaria. The present study was carried out to find out the vector abundance, feeding, resting and breeding habitats, sibling species composition, and insecticide susceptibility status of malaria vectors to evolve a suitable vector control strategy. The study was initiated in August 2012. Three villages, i.e. riverine stony/sandy having high malaria, riverine muddy having low malaria and non-riverine with high malaria transmission were selected for monthly monitoring. Three vector species *Anopheles culicifacies*, *An. stephensi* and *An. fluviatilis* were recorded. *Anopheles culicifacies* was the most predominant species followed by *An. stephensi* and *An. fluviatilis*. In the riverine high malaria village abundance of *An. culicifacies* was maintained almost throughout the year while in the muddy village peak density was noticed in September. In non-riverine high malaria village the peak density was found only in September. The parity rate of *An. culicifacies* was more in riverine and non-riverine high malaria villages than riverine low malaria village. *Anopheles culicifacies* was mainly endophilic and zoophagic. The peak landing of *An. culicifacies* in riverine and non-riverine high malaria villages was in November and September, respectively between 0200–0300 hrs. However, in riverine low malaria village there was no clear peak and intermittent landing was observed mainly from June to November in second half of the night. The main breeding sources of *An. culicifacies* in riverine high malaria village were sandy/stony river and seepages, in riverine low malaria village muddy river with floating vegetation, seasonal streams and seepages and in non-riverine high malaria village irrigation tank, seasonal stream, seepage and wells.

Maximum contribution of *An. culicifacies* was from sandy/stony river, followed by seasonal streams, seepages, irrigation tank, river muddy and wells. *Anopheles culicifacies* was found resistant to DDT 4%, malathion 5% and lambda-cyhalothrin 0.05% in the area. The densities of *An. culicifacies* did not decline after indoor spray of lambda-cyhalothrin in either of the study villages.

The peak densities of *An. stephensi* were more in riverine high malaria and non-riverine high malaria villages than riverine low malaria village. During the study period *An. stephensi* was prevalent in 4–6 months in riverine high malaria (August, November, December, February, April, June) and non-riverine high malaria (August, September, October, July) villages, whereas in three months in riverine low malaria village (November, June, July). *Anopheles stephensi* was mainly endophilic and zoophagic. The maximum landing of *An. stephensi* on animal bait was observed in second half of the night in May in riverine high malaria and riverine low malaria villages and in September in non-riverine high malaria village, with the peak in between 0400–0600 hrs in riverine high malaria village and 1200–0200 hrs in riverine low malaria and non-riverine high malaria villages. The main contribution of *An. stephensi* was from sandy/stony river followed by wells and muddy river. Egg morphometrics of *An. stephensi* revealed that it is mysoriensis, a non-vector rural form in the area.

The peak man hour density of *An. fluviatilis* in riverine villages was more than non-riverine village. Further, among riverine villages the density was high in riverine low malaria village than riverine high malaria village. *Anopheles fluviatilis* was prevalent only in November in riverine high malaria village, whereas in riverine low malaria village from October to February and in non-riverine village from September to November. *Anopheles fluviatilis*

was mainly endophilic and zoophagic. The maximum landing of *An. fluviatilis* was observed between 0300 and 0500 hrs in riverine high and low malaria villages. The peak landings in riverine high malaria village was in November whereas, in riverine low malaria village peak landings were observed in December.

A total of 231 blood smears in riverine high malaria, 273 in riverine low malaria and 230 in non-riverine high malaria villages were collected and examined for malaria parasites for the reporting period. Out of these, only one in riverine high malaria and two in non-riverine high malaria villages were found positive for *Plasmodium vivax* during April 2013; and September 2012 and July 2013, respectively. All the positive cases were migratory in nature.

This study showed that malaria is more abundant in the villages located adjacent to the dam having stone margins. We have recommended that cemented walls may be fenced along the border of the dam so that mosquito breeding can be prevented. In this area, fish have been released and found very effective.

#### **Phase-III trial for evaluation of Lambda-cyhalothrin 10CS indoor residual spraying for malaria vector control in Karnataka**

The study was continued in two areas of Karnataka. In Upper Krishna Project (UKP) area, the comparator insecticide was lambda-cyhalothrin 2.5% WP, whereas in Gulbarga district DDT is the comparator insecticide.

#### **Upper Krishna Project (UKP) area**

Villages are situated in the command area of UKP. Three villages, namely Madlingnal, Kureknal and Kureknal Thanda covering 2986 population were selected as experimental and Davergaddi, Mallengaddi and Rodal Banda camp covering 2950 population as control villages. In this area, ICON 10% WP was taken as comparator insecticide. Pre-intervention per man hour and per structure densities of malaria vector *An. culicifacies* monitored in August 2012 in the experimental area were 18.5 and 41 and in the control area 5.5 and 6, respectively. In the experimental area, I round of lambda-cyhalothrin 10CS indoor spray was carried out in November 2012 with 66.7% room coverage and II round in June 2013 with 65% room coverage. In control villages, two rounds of lambda-

cyhalothrin 10%WP spray were carried out with 59.5% room coverage in I round and 57% in II round. After lambda-cyhalothrin 10CS spray the densities of malaria vector *An. culicifacies* remained low throughout the study period as compared to the baseline densities. After I round of spray per man hour and per structure densities of *An. culicifacies* declined gradually up to five months. The densities increased after six months of I round spray and again after II round the densities declined for one month and increased in the third month, but below baseline. After lambda-cyhalothrin 10% WP spray the densities remained low up to 5 months of I round spray. The densities increased above baseline densities in the sixth month of I round of spray. After second round, the densities declined up to one month and increased more than baseline in the third month. Cone bioassays on lambda-cyhalothrin 10CS sprayed walls revealed >80% mortality up to 2 months of I round of spray. In third and fourth months, the mortality declined to 66.7 and 60%, respectively. After II round of spray the mortality was >80% up to one month and dropped to 75% in the third month. However, cone bioassays on lambda-cyhalothrin 10% WP sprayed walls revealed >80% mortality up to one month after I and II rounds of spray. The mortality gradually declined to 50% after four months. The parity rate of malaria vector *An. culicifacies* before spray was 83.3% which came down to 10% after lambda-cyhalothrin 10CS spray. However, in lambda-cyhalothrin 10% WP sprayed area, the parity rate before spray was 69.2 which came down to 25% after spray.

#### **Gulbarga district**

This area is semi-arid. In Javargi taluka of Gulbarga district, Sakapur and Auarad villages having 3649 population were selected as experimental villages and Ravnoor and Koodi villages having 3075 population as control villages. Pre-intervention man hour density of malaria vector *An. culicifacies* monitored in August 2012 in experimental and control villages was 17.5 and 19, respectively. Lambda-cyhalothrin 10CS was sprayed in Sakapur and Auarad in November 2012 as I round with 65.9%, and in June 2013, as II round with 66.6% room coverage. In Ravnoor and Koodi villages, DDT 50% was sprayed as comparator. In Ravnoor village DDT was sprayed by the state health staff, whereas in Koodi DDT was sprayed

under the supervision of NIMR staff with 71.7% room coverage in I round and 50.4% in II round. Per man hour and per structure densities of *An. culicifacies* remained low up to five months after I round and up to one month after II round of lambda-cyhalothrin 10CS spray. After three months of II round of lambda-cyhalothrin 10CS spray the densities increased above the baseline densities. In DDT sprayed area, the per structure density of *An. culicifacies* declined after one month of I and II rounds of spray. However, after 2–3 months the densities increased above baseline density. Cone bioassays on lambda-cyhalothrin 10CS sprayed walls revealed >80% mortality up to three months of I round of spray. Cone bioassays on DDT sprayed walls revealed only 5–40% mortality after I and II rounds of spray. The parity rate of malaria vector *An. culicifacies* before spray was similar in lambda-cyhalothrin 10CS and DDT sprayed areas. However, after spray the parity rate was lesser in lambda-cyhalothrin 10CS sprayed area than DDT sprayed area.

This study showed that ICON 10CS is effective up to 12 weeks.

#### **Larvivorous fish in vector control in JE prone areas in Gorakhpur district, eastern Uttar Pradesh.**

This challenging project has been undertaken to address the long problem of JE-related AES cases and related deaths mostly among children. In July 2012, about 15000 *Gambusia* were brought to Gorakhpur from Bengaluru. The fish multiplied and established in a pond at Gorakhnath Temple. From July 2013 release of this fish has been completed in 27 tolas in six villages and 30 ponds in 19 blocks in the district. About 255,000 *Gambusia* were released in 58 ponds, rice-fields and borrow pits. About 5300 Guppies have been released in 68 wells.

The entomological data revealed that the vector mosquito *Cx. tritaeniorhynchus* and *Cx. pseudovishuni* were found breeding in wells, rice-fields, ponds and burrow pits. In July, *Cx. epidesmus* was found in light-trap collection. This area is represented by extreme weather conditions. In the monsoon season rice-fields provide additional breeding grounds for the vectors. Falling winter rice-fields are replaced with wheat cultivation and in the summer nearly 60% ponds and 70% wells get dry. Road side burrow pits also get dry. Here ponds and wells are the main breeding grounds

for the vectors. Most of the wells are covered with garbage and support the breeding of *Cx. quinquefasciatus*. It is suggested to cover the wells. We are targeting these two habitats with fish which are found to be very effective.

#### **Small-scale evaluation of the efficacy and residual activity of alpha-cypermethrin WG (250 g a.i./kg) for indoor spraying in comparison with alpha-cypermethrin WP (50 g a.i./kg) in India**

A study on indoor residual spraying with alpha-cypermethrin WG (250 g a.i./kg) formulation in comparison with WP formulation (50 g a.i./kg) was undertaken at Balepura village, Devanhalli Taluka, Bengaluru, India. Two dosages of the two formulations, i.e. 20 and 30 mg a.i./m<sup>2</sup> were used in the present study and the efficacy and duration of effectiveness was assessed on alpha-cypermethrin susceptible population of *An. stephensi*. Four types of surfaces were selected for assessing the efficacy of the formulations, namely cement wall with distemper coating, cement wall with lime coating, mud wall with lime coating, and brick wall unpainted. Spraying was carried out with Hudson sprayer fitted with control flow valve. Bioassays were carried out at weekly and then fortnightly intervals. Chemical analysis of filter paper samples collected from the sprayed surfaces were done at Walloon Agricultural Research Institute for the target concentration of the alpha-cypermethrin content. The duration of effective residual action (>80% mortality) of alpha-cypermethrin WG 30 mg/m<sup>2</sup> applied on local surfaces was 16 weeks for cement walls and mud walls, whereas on brick walls unpainted it was 13 weeks; while on WG 20 mg/m<sup>2</sup> applied surfaces the efficacy was 15 weeks on cement wall and distemper (a paint containing water, chalk and glue/resin) coated surfaces, while cement wall + lime coated, mud wall + lime coated and brick wall unpainted surfaces, it was 13 weeks. The duration of effective residual action of alpha-cypermethrin WP formulation sprayed at 30 mg/m<sup>2</sup> was 14 weeks on cement wall surfaces, and 11 weeks on mud wall and brick wall surfaces, whereas on 20 mg/m<sup>2</sup> sprayed surfaces were 15 weeks on cement wall + distemper coated surface and mud wall + lime coated surface and 11 weeks on cement wall + lime coated and brick wall surfaces (Table 1). No adverse events were reported either by the spraymen or the household inhabitants during and after

**Table 1. Duration of effective residual action of different dosages and formulations of alpha-cypermethrin against *Anopheles stephensi***

S. No.	Formulation Test surface and Dose	Duration of effectiveness in weeks post-spray (>80% mortality)
1.	WG-30 Cement wall + distemper coated	16
	Cement wall + lime coated	16
	Mud wall + lime coated	16
	Brick wall unpainted	13
2.	Cement wall + distemper coated	15
	Cement wall + lime coated	13
	Mud wall + lime coated	13
	Brick wall unpainted	13
3.	Cement wall + distemper coated	14
	Cement wall + lime coated	14
	Mud wall + lime coated	11
	Brick wall unpainted	11
4.	Cement wall + distemper coated	15
	Cement wall + lime coated	11
	Mud wall + lime coated	15
	Brick wall unpainted	11

the spray operations. Sneezing and headache reported by two persons was transient in nature and subsided shortly without requiring medical treatment.

The results suggest that the dose of WG 30 mg/m<sup>2</sup> gave longer effective residual action against malaria vector (16 weeks) on most common indoor surfaces and could be used for effective control of *Anopheles* mosquitoes. The WG formulation was found to be easy to handle, no smell was reported during the spray and was found to be operationally acceptable for indoor residual spraying.

#### **Phase-II baseline household survey in World Bank districts of Maharashtra, Gujarat, West Bengal and Karnataka**

This is a World Bank assisted project. In Karnataka, 10 PHCs in six districts, namely Gulbarga, Raichur, Bagalkot, Bijapur, Koppal and Tumkur were included in the study. Eight villages were selected from each PHC, thus 80 villages were selected based on malaria endemic status. Again, 22 households from each village were selected based on random number formula. Ten tools mainly questionnaire survey-based were applied for the survey.

Local staff were selected and underwent training at two workshops at Raichur and Tumkur. Ten teams were formed consisting of five members each. In all these villages, slides were prepared from

recent fever cases and none found positive for malaria. This indicated that malaria is on the verge of elimination in this area.

#### **Malaria parasite biology: An avenue to discover new drug targets. Study on heme-biosynthesis in malaria parasite and development of new antimalarial drug targets**

This is a collaborative study between Prof. G Padmanaban, Department of Biochemistry, IISc, Bengaluru and NIMR, Field Unit, Bengaluru. Heme is required for the biogenesis of cytochromes—the essential components of parasite electron transport chain. In the intraerythrocytic stages, the parasite acquires heme from host hemoglobin and detoxifies it into hemozoin. Malaria parasite is capable of synthesizing heme *de novo*. To address the role of dual sources of heme in parasite growth and development, knockouts (KOs) were generated for the first enzyme, δ-aminolevulinate synthase (ALAS), and the last enzyme, ferrochelatase (FC), in the heme-biosynthetic pathway of *P. berghei* (*Pb*). The intra-erythrocytic stages of both *Pb*ALAS and *Pb*FCKOs grow and multiply normally in mice. *In vitro* radiolabeling studies carried out with the *Pb*KOs and *P. falciparum* wild type parasites have revealed that the malaria parasite can incorporate both hemoglobin-heme and *de novo* heme into mitochondrial cytochromes and hemozoin. The identical fates of the two sources of heme indicate that they serve as back up mechanisms to provide heme in the intraerythrocytic stages.

Interestingly, the *de novo* heme becomes indispensable in sexual and liver stages, where the parasites do not have access to the host hemoglobin. *Pb*KO parasites show a drastic decrease in the formation of oocysts and no sporozoite could be detected in the salivary glands of mosquitoes. The basis for this absolute requirement of heme needs to be investigated. In *Pb*ALASKO parasites, this could be restored by supplementing the mosquitoes with ALA. Further, ALA supplementation to the animal is required for these sporozoites to undergo liver stage development in mice.

#### **Analysis of *in vivo* transcriptome of *Plasmodium falciparum* from Indian patients suffering from cerebral malaria and its comparison with that from patients infected with severe malaria (with MOD symptoms)**

This is a collaborative study between Prof.

Namita Surolia, Jawaharlal Nehru Centre for Advanced Scientific Research, Bengaluru and NIMR Field Unit, Bengaluru. Cerebral malaria caused by *P. falciparum* infection is an ongoing challenge to understand the process of such events. This study has been undertaken for comparative reasons. Severe and cerebral malaria cases will be taken from Jabalpur, whereas non-severe cases will be taken from Mangalore City as comparator. Details of patient history, clinical and biochemical outcome will be recorded. RNA will be isolated for transcriptome analysis.

### Proteome analysis of *in vitro* gametocytogenesis in *Plasmodium falciparum*

This is a collaborative study between Prof. Utpal Tatu, Department of Biochemistry, IISc, Bengaluru and NIMR, Field Unit, Bengaluru. Gametocyte in malaria parasite is an important stage. This stage is targeted for transmission blocking. We have developed an *in vitro* technique where a good amount of *P. falciparum* gametocytes can be harvested. Different stages of chloroquine resistant and sensitive gametocytes have been successfully established. We did not find any difference between the two strains as per proteomic analysis.

### Technical Support to the Programme

#### Investigation of dengue outbreak

In 2013, outbreaks of dengue were reported from different parts of Karnataka. With the request from the State Health Department, an investigation was carried out in two villages Salundi and Dhalundi, CHC Jaipura, Mysore Taluka in June 2013. Entomological surveillance indicated high larval index of Aedes sp (Breteau index > 70), even after antilarval measures taken by the local health department. This information was shared with the state government and proper action was taken to eliminate the Aedes breeding and thus contained the outbreak. Besides Ae. aegypti, high abundance of Ae. vitatus (16%) was recorded.

#### Investigation of malaria-related deaths in Dakshina Kannada district

In January 2014, two malaria deaths were reported in village Shishila, PHC Hathyadka, Belthangady Taluka, Dakshina Kannada district. We conducted entomological investigations from February to May 2014, which could not establish

indigenous transmission of malaria in the village. *Anopheles jeyporiensis* was recorded in this area.

#### Transmission assessment survey of lymphatic filariasis in Udupi district

TAS was carried out in Udupi district from 18 to 20 March 2014. This was to find out whether this district will qualify for elimination of lymphatic human filariasis. A total of 1556 standard I and II school children (6 to 8 years age) from 53 schools in Udupi district were tested for filariasis using BinaxNOW—Filariasis antigen ICT kits (Figs. 1 and 2). ICT results on school children indicated that District Udupi qualify for ELF.



Fig. 1: Screening for filarial antigen detection using BinaxNOW Filariasis ICT kit in a school in Udupi district.



Fig. 2: Results of screening shown by a filarial antigen test.

#### Fish based malaria control in Karnataka

The fish-based malaria control has been extended to most malaria endemic areas of Karnataka, mostly in districts of northern Karnataka. Data revealed so far that malaria cases have come down to 50% compared to 2012 and almost half of malaria cases are being contributed from Mangalore and Udupi cities. Action is being taken to bring down malaria cases in these cities.

Besides this programme, entomological training to the local Entomologists has been provided. Training on larvivorous fish release, handling, monitoring, etc was also given to the local health staff.

### Participation in Conferences/Scientific meetings

#### Dr SK Ghosh

1. Attended the video conference on Dengue in Vikas Sadan on 9 July 2013.
2. Attended the XII International conference on vectors and vector borne diseases' at Udaipur, India from 16 to 18 September 2013.
3. Attended the 62nd Annual Meeting of the American Society of Tropical Medicine and Hygiene held at Washington DC from 13 to

17 November 2013.

4. Attended a meeting with the Chief Executive Officer, Zilla Parishad, Dakshina Kannada district at Mangalore on 20 March 2014.

#### Training/Workshops/Seminar organized

1. Organized Transmission Assessment Survey for ELF Programme for Karnataka, Madhya Pradesh and Uttar Pradesh from 30 September to 1 October 2013. This survey was supported by NVBDCP, Delhi.
2. Organized two workshops on Household Surveys in Karnataka from 26–30 August 2013; and 2–3 September 2013 in Raichur and Tumkur districts, respectively. About 80 participants attended these two workshops.

□

## 3.2

### Chennai (Tamil Nadu)

#### **Monitoring of antimalarial resistance to *Plasmodium vivax* and *Plasmodium falciparum* malaria in endemic areas of Tamil Nadu, India**

The study was designed to find out the therapeutic efficacy of chloroquine to vivax malaria in Rameswaram Island based on the request from the Directorate of Public Health & Preventive Medicine, Govt. of Tamil Nadu. The study has been carried out in Thangachimadam and Pamban PHCs of Rameswaram Island, Ramanathapuram district, Tamil Nadu in September 2013 following the protocol of NIMR, New Delhi. Out of 217 blood smears examined at Thangachimadam PHC, 12 were found to be positive for malaria with 9 Pv, 2 Pf and 1 with mixed infection. In Pamban PHC, 142 blood smears were examined and, all 9 were found to be positive for Pv. A total of 9 vivax malaria patients (5 from Thangachimadam and 4 from Pamban) were enrolled in the study based on the inclusion/exclusion criteria and informed consent was obtained. Among them, 3 were males and 6 were females. Six patients were above 15 years of age and the remaining three were children below 10 years. The hemoglobin level of patients at the time of enrolment ranged from 10.2 to 12.6 g/dl and the minimum and maximum parasitaemia was 760 and 14, 240 parasites/ $\mu$ l respectively.

All the patients were followed up on Days 1, 2, 3, 7, 14, 21 and 28 and on any other day they reported for fever. Results indicated adequate clinical and parasitological response (ACPR). Complete parasite clearance was observed in 3 patients each by 24, 48 and within 72 h. Further, enrolment and followup is planned to enroll more patients when more number of malaria cases are reported.

#### **Impact of new control tools and changing patterns**

The objective of the study is to describe the

changing trends of malaria incidence at micro-level (PHCs) between 2005 and 2011; to find out the coverage achieved by new malaria control tools (RDGs, ACT & LLINs) for each micro-level and to determine the association between new policies and intervention coverage at the micro-level with changes in malaria incidence. A total of 48 PHCs in different districts of Tamil Nadu have been randomly selected for the study. During the reporting period, the survey was undertaken in the Primary Health Centre's of R.S. Mangalam and Mandapam in Ramanathapuram district and malaria clinics in Zones VI and VIII (Old) of Corporation of Chennai.

Preliminary observations in both the PHCs of Ramanathapuram district indicated surveillance mechanism for case detection to be highest by passive case detection in OPD (56.08%) and in sub-centres (23.88%) followed by MPW (13.86%), mass/contact surveys (5.78%) and fever treatment depots (0.4%). However, in Chennai surveillance for case detection was predominant by passive case detection in malaria clinics (95.28%) followed by mass/contact surveys (3.08%) and by active surveillance (1.64%). Rapid diagnostic kits and LLINs have not been made available in the PHCs. ACT treatment for Pf cases is being carried out directly under the supervision of District Malaria Officer in Ramanathapuram district. *Plasmodium vivax* cases are treated with chloroquine and primaquine. Presumptive treatment has been withdrawn and 14 days treatment with primaquine initiated since 2010.

#### **Assessment of malaria gametocytaemia with duration of symptoms: A potential programme monitoring tool for delay in seeking treatment**

The study has been planned to determine the validity of the proportion of patients with

gametocytes as a monitoring tool for delayed access to health care and treatment. During the period, a total of 34 malaria patients, 32 *Pv* and 2 *Pf* cases were enrolled in the malaria clinic, functional at Field Unit, Chennai. All the malaria patients were above 15 years of age and the number of males and females were 24 (70.6%) and 10 (29.4%), respectively. Previous history of malaria and treatment seeking period of patients were recorded and 35.3% of the patients informed that they had received treatment elsewhere for malaria, but it could not be confirmed whether the treatment was complete or incomplete. Patients got their blood diagnosed for malaria on Days 3, 4, 6, 7, 9, 10, 13 and 14 after the onset of fever. Majority of the patients (44.1%) sought diagnosis and treatment on Day 4 after the initial onset of fever indicating seeking diagnosis and treatment almost after 2 bouts of fever episode while 88.2% of the patients visited the clinic for treatment within a week after the onset of fever and 11.8% between week 1 and 2. In patients with vivax malaria, gametocytes were observed in all the patients. The minimum and maximum gametocytaemia was 200 and 5040/ $\mu$ l, respectively. The mean gametocytaemia in patients who reported within and after a week of onset of fever were 1077 and 970/ $\mu$ l, respectively. The proportion of gametocytes to asexual stages was proportionally high in 10 out of 34 patients enrolled.

#### **Centre for the Study of Complex Malaria in India (CSCMi)**

The community and clinic study, the two major arms of eco-epidemiology project was initiated in Besant Nagar, Chennai from December 2012. Enrolment for community (quarterly) and clinic study (weekly) was carried out and all the positive cases were followed on Days 2, 7, 14, 21, 28 and 42 in the clinic study, where 658 individuals were enrolled for cross-sectional study to estimate the prevalence of asymptomatic malaria and 407 (61.9%) were females and 251 (38.1%) were males. The prevalence of malaria was 7 (1.1%) by microscopy and 21 (3.3%) by PCR out of 636 samples processed. However, the prevalence of asymptomatic malaria was very low, 2 (0.3%) by microscopy and 18 (2.8%) by PCR. Among the malaria positives detected by microscopy, 6 (85.7%) were *Pv* and 1 (14.3%) *Pf*. Out of 21 positives detected by PCR, *Pv* accounts for 12 (57.1%), 8 (38.1%) *Pf* and 1 (4.8%) with mixed infection.

In all, 309 fever patients reported to in the clinic were screened to find out the prevalence of symptomatic malaria, where 53 (17.2%) were found positive by microscopy and 59(20.7%) by PCR, out of 285 samples processed (Fig. 1). Species prevalence indicated 51 (96.2%) as *Pv*, 1 (1.9%) *Pf* and 1 (1.9%) with mixed infection on microscopic examination. However, 49 (83.1%) *Pv* and 10 (16.9%) *Pf* were detected by PCR. Of the 53 malaria positive cases, 41 (77.4%) were males and 12 (22.6%) were females, while 28 (52.8%) enrolled subjects were followed on Days 2, 7, 14, 21, 28 and 42. Of which 9 (32.1%) were microscopy positive on Day 2 and all were negative on subsequent visits. In all, 139 individuals from 34 households enrolled in longitudinal study were followed once in three months to find out the occurrence of malaria. PCR detected 8 malaria positives, out of which 3 were *Pv*, 4 *Pf* and 1 mixed infection. The subjects were then followed up every 15 days for the presence of any febrile/illness related to malaria and none reported symptoms related to/diagnosed as malaria (Fig.2). Subsequently, in January 2013 reactive case detection surveillance was initiated immediately in response to positive case reported at malaria clinic involving screening of all the household members of the index case. Individuals living in close proximity (< 50 m radius) to passively detected case and additionally, households within 100 m radius were surveyed for the presence of fever besides, randomly selected control households without any history of fever in previous two weeks were screened for the presence of malaria parasites by microscopy, RDT and PCR.

Regular field sampling in wells, overhead tanks



Fig. 1: Laboratory staff attending a patient at CSCMi Clinic at Rajaji Bhawan, Chennai.



Fig. 2: Follow up of patients at the field site.

and cisterns was carried out (fortnightly basis) to check the larval abundance and co-habitation of anophelines with other mosquito species. The mean larval density ranged from 0.42 to 7.53 in wells, 0.5 to 10.9 in OHTs and 0.06 to 9.98 in cisterns (Fig. 3).

Fortnightly adult collections of *An. stephensi* in cattlesheds were carried out to check the vector density and seasonal fluctuations. The average density (MHD) ranged from 3 (January) to 73.85 (June). The collected adult mosquitoes were identified to the species level and abdominal

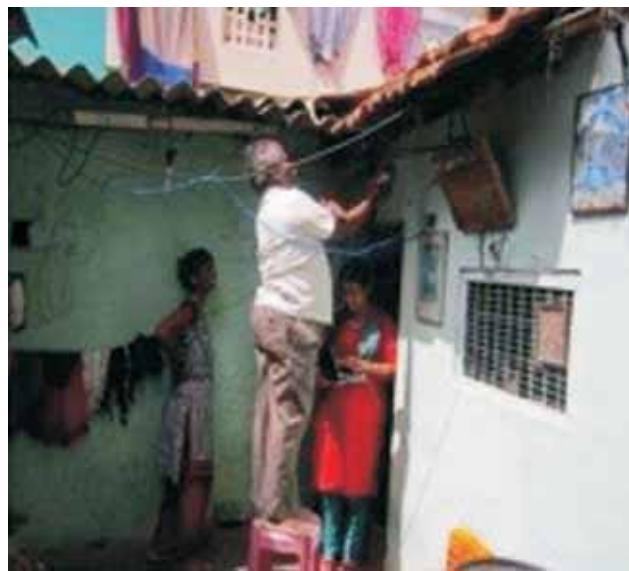


Fig. 4: Placement of 'Hobo' for recording temperature and humidity.

conditions were categorized (Fig. 4). All were dissected for the presence/absence of sporozoites and oocysts of *Plasmodium* and were found negative. Besides, blood meal analysis performed on 362 samples, 14 (3.86%) were found to be of human, 13 (3.59%) mixed, 2 (0.55%) with unidentified results and the rest (91.99%) were of bovine blood. Micro climatic ambient temperature ( $^{\circ}\text{C}$ ) and relative humidity (%) recorded (hourly basis) in different structure types/habitats observed

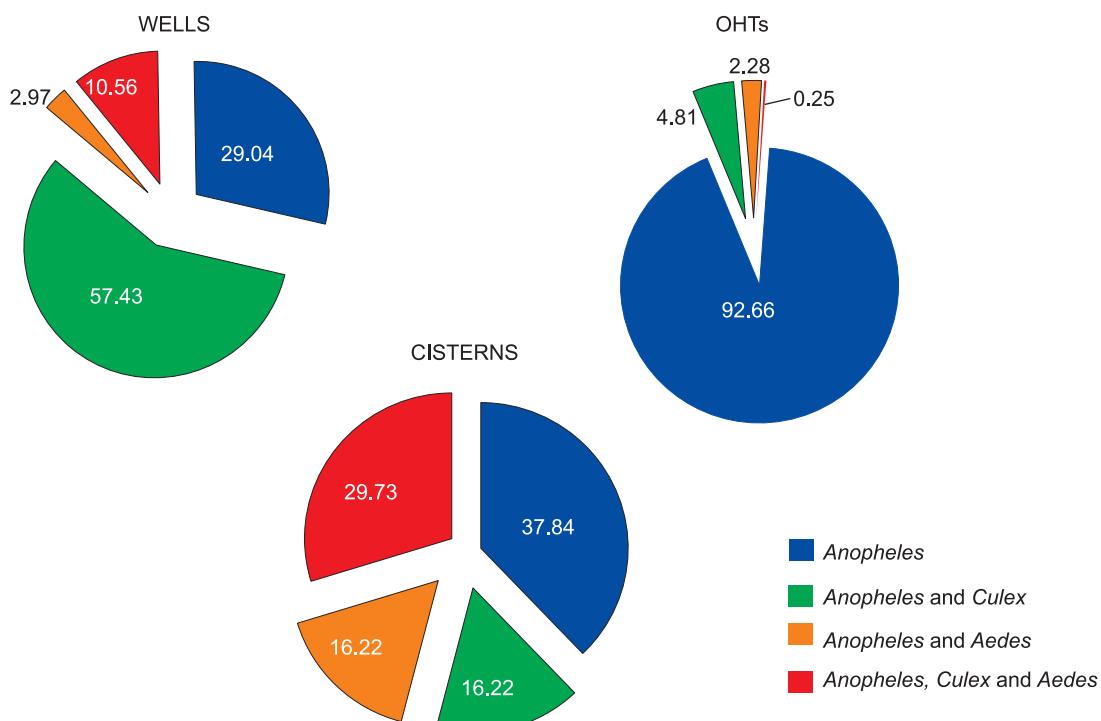


Fig. 3: Percent contribution of different mosquito species in wells, over head tanks (OHTs) and cisterns in Chennai City.

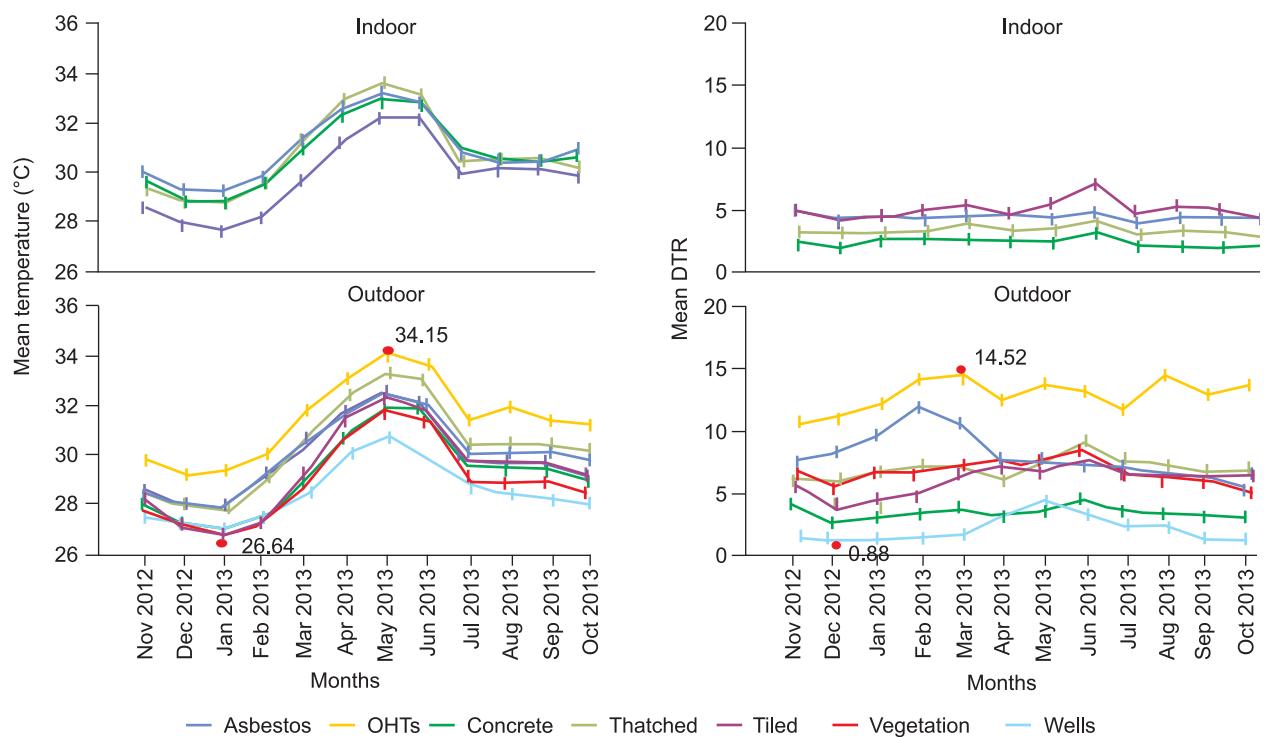


Fig. 5: Mean temperature and mean DTR in different resting habitats of *An. stephensi* in Chennai City.

minimum mean temperature of  $26.64 \pm 0.44$  (January) in outdoor thatched structure and the maximum mean temperature of  $34.15 \pm 2.29$

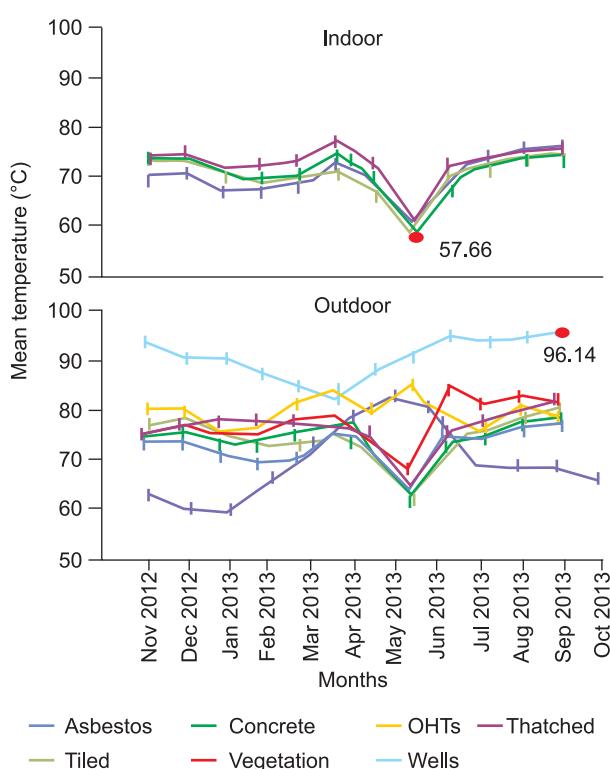


Fig. 6: Monthly mean relative humidity in different resting habitats (indoor and outdoor) of *An. stephensi* in Chennai City.

(May) in outdoor structure (OHT) (Fig. 5). Similarly, the minimum mean relative humidity of  $57.65 \pm 8.01$  (June) in indoor concrete structure, whereas the maximum mean relative humidity of  $96.14 \pm 3.44$  (October) was recorded in wells (Fig. 6). Physico-chemical parameters of *An. stephensi* breeding habitats (wells and OHTs) analyzed indicated that the mean sulphate ( $\text{SO}_4$ ) was significantly more in wells with breeding ( $p < 0.05$ ).

#### Identifying the molecular marker(s) for relapse malaria in *Plasmodium vivax*

Malaria patients attending Central Malaria Laboratory (clinic) of George Town, Chennai were screened for *P. vivax* genotyping in order to differentiate between relapse and reinfection. In all, 160 patients who had attended the clinic had been screened to find out the prevalence of malaria where there were 125 (78.12%) males and 35 (21.87%) females and the prevalence of malaria was only 3.75% on microscopic examination. Of the 6 malaria positives, 3 were *Pv*, 1 *Pf* and 2 with mixed infection and 3 patients selected on date based on the inclusion/exclusion criteria and history recorded. Parasitaemia recorded on Day 0 ranged from 1080 to 11,520 parasites/ $\mu\text{l}$ . Blood spots of *Pv* patients were collected on sterile Whatmann filter paper on Day 0 and after the completion of

treatment/subsequent episodes of malaria for genotyping of *P. vivax*. Further screening of *P. vivax* patients, recording of parasitaemia and collection of blood spots is in progress.

### Technical and training support to the programme

#### Diagnosis and treatment of malaria patients

The malaria clinic continued to function at the Field Unit premises. A total of 269 febrile patients from nearby areas, namely Ayapakkam, Anna Nagar, Mogappair, Padi Pudu Nagar and Ambattur, came to the clinic were diagnosed for malaria by conventional microscopy. Among them, 32 and 2 patients were suffering from vivax and falciparum

malaria, respectively. All the malaria patients were treated as per NVBDCP drug schedule/policy. Clinic data on malaria cases were provided to the Health Department, Corporation of Chennai for further follow-up and control measures.

#### Health Education/Awareness on Malaria

A radio-talk in Tamil by Dr K John Ravindran, ARS on malaria and its prevention was broadcasted as part of the programme Arogya Bharatham, Health serial sponsored by the Ministry of Health & Family Welfare, Govt. of India (September 2013). The recording and broadcasting was organized by the All India Radio (AIR), Chennai.



### 3.3

## Guwahati (Assam)

### Seasonal abundance of *Aedes (Stegomyia) albopictus* and *Aedes (Stegomyia) aegypti* in Guwahati metropolis and suburban settlements, Assam, northeast India

Dengue is fast establishing in northeast India and spreading on account of rapid urbanization and population movement with reported morbidity and attributable deaths. The present study is aimed to determine seasonal abundance of *Aedes (Stegomyia) albopictus* and *Ae. (Stegomyia) aegypti* in Guwahati metropolis and suburban settlements, and to characterize the breeding sources and ascertain susceptibility status to adulticides and larvicides. Mosquito larval surveys were made in different localities both in Guwahati City and adjoining suburbs during January till December 2013 to determine seasonal abundance of disease vectors and their breeding preferences (Fig. 1). Insecticide susceptibility status against mosquito adults as well as larval populations of both *Ae. aegypti* and *Ae. albopictus* was ascertained using WHO standard diagnostic concentrations and test procedures.

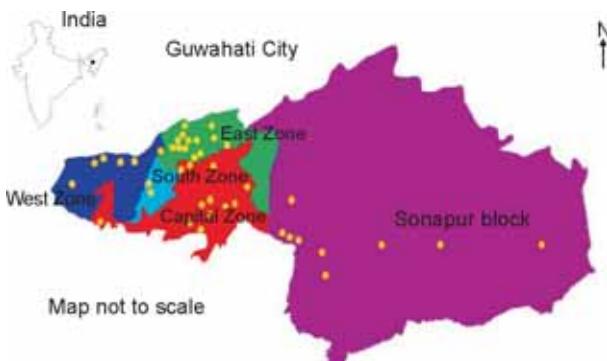


Fig. 1: Diagrammatic sketch map of Guwahati City and adjoining suburban Sonapur block, showing mosquito larval survey locations (yellow circles) in different zones. *Aedes aegypti* is the predominant mosquito species in the city (total cases = 4121) and *Ae. albopictus* in the suburban Sonapur block (total cases = 2). Inset is the map of India showing location of the study site, and Table 1 giving distribution of dengue cases in different zones and corresponding entomological indices of disease vectors.

The present study revealed that both *Ae. aegypti* and *Ae. albopictus* are widely abundant in Guwahati City and suburbs (Tables 1 and 2), and observed breeding in wide variety of resources (Fig. 2). *Aedes albopictus*, however, was the

**Table 1. Monitoring seasonal abundance of *Aedes albopictus* and *Aedes aegypti* mosquito breeding, dengue vector mosquito species in Guwahati metropolis and suburbs, Assam**

City Zone	No. of dengue cases	Mean mosquito breeding indices (range)		
		HI	CI	BI
West Zone	238	13.66 (0.04–30)	34.76 (5.19–61.5)	55.31 (8–160)
East Zone	2863	12–36 (3.22–28.5)	36–67 (13.43–80)	31.34 (20.19–57.1)
Capital Zone	819	14.40 (2.17–27.5)	23.39 (1.07–38.4)	40.43 (2.17–62.5)
South Zone	201	No data	No data	No data
Sonapur Block	2	20.41 (4.34–40)	26.39 (2.85–60)	62.06 (4.34–120)

HI: House/premise index; CI: Container index; BI: Breteau index.



Fig. 2: Breeding habitats of *Ae. aegypti* and *Ae. albopictus* in Guwahati City and adjoining suburbs, Assam, India: (a) A discarded tyre, a preferred breeding source for *Ae. aegypti* in urban areas; (b) A cut-bamboo stump, a breeding source for *Ae. albopictus* in rural areas; (c) A leaf axil, breeding source for *Ae. albopictus*; and (d) A flower vase, a preferred breeding habitat for *Ae. albopictus*.

## GUWAHATI (ASSAM)

**Table 2. Seasonal container positivity of *Ae. albopictus* and *Ae. aegypti*, dengue vector mosquito species in Guwahati metropolis and adjoining suburbs, Assam, India (2013)**

Month	Total rainfall (mm)	Total No. of wet containers searched for mosquito breeding	No. (%) containers positive for mosquito breeding	No. (%) of container (+) ve for species			No. of reported dengue cases*
				<i>Ae. albopictus</i>	<i>Ae. aegypti</i>	Mixed breeding	
Jan 2013	0	97	0	0	0	0	0
Feb	11.4	172	0	0	0	0	0
Mar	21.7	263	0	0	0	0	0
Apr	94.7	205	6 (2.92)	6 (2.92)	0	0	0
May	464.3	192	65 (33.85)	54 (28.12)	1 (0.52)	10 (5.20)	1
Jun	336.5	140	42 (30)	12 (8.57)	30 (21.43)	0	0
Jul	260.6	285	65 (22.80)	45 (15.78)	17 (5.96)	3 (1.05)	2
Aug	135.4	226	40 (17.69)	25 (11.06)	15 (6.63)	0	75
Sep	122.7	237	60 (25.31)	12 (5.06)	27 (11.39)	21 (8.86)	1635
Oct	214.6	587	77 (13.1)	10 (1.70)	41 (6.98)	26 (4.42)	1395
Nov	0	75	25 (33.3)	5 (6.67)	20 (26.67)	0	827
Dec	0	92	28 (30.43)	3 (3.26)	16 (17.39)	9 (9.78)	188
Total	1661.9	2571	408 (15.9)	172 (6.7)	167 (6.5)	69 (2.7)	4123

\*Data based on reported cases in the study area (Source: State Health Directorate of Assam).

**Table 3. Major breeding habitats and relative abundance of *Ae. albopictus* and *Ae. aegypti* in Guwahati metropolis and adjoining suburbs, Assam, India (April–December 2013)**

Breeding habitat	Ecotype	No. of wet containers searched for mosquito breeding	No. (%) of containers positive for mosquito breeding	No. of containers (+) ve for species (Container Index)		
				<i>Ae. albopictus</i>	<i>Ae. aegypti</i>	Mixed breeding
Tyres	Urban	799	172 (21.5)	17 (2.1)	122 (15.3)	33 (4.1)
	Semi-urban	194	72 (37.1)	21 (10.8)	25 (12.9)	26 (13.4)
Bamboo stumps	Urban	75	13 (17.3)	11 (14.7)	0	2 (2.7)
	Semi-urban	132	37 (28)	35 (26.5)	0	2 (1.5)
Tin/Plastic containers	Urban	334	46 (13.8)	29 (8.7)	13 (3.9)	5 (1.5)
	Semi-urban	50	10 (20)	10 (20)	0	0
Flower vases	Urban	154	14 (9.1)	14 (9.1)	0	0
	Semi-urban	202	26 (12.9)	26 (12.9)	0	0
Leaf axils	Urban	35	4 (11.4)	4 (11.4)	0	0
	Semi-urban	7	2 (28.6)	2 (28.6)	0	0
Cement tanks	Urban	16	7 (43.7)	0	7 (43.7)	0
	Semi-urban	2	0	0	0	0
Battery boxes	Urban	28	3 (10.7)	0	4 (14.3)	0
	Semi-urban	1	0	0	0	0
Coconut shells	Urban	7	1 (14.3)	0	0	1 (14.3)
	Semi-urban	3	1 (33.3)	1 (33.3)	0	0

predominant mosquito species in suburbs breeding preferentially in flower vases, cut-bamboo stumps and leaf axils. *Aedes aegypti* was the most common in city breeding predominantly in discarded tyres, cement tanks and used battery boxes (Table 3). Both *Ae. aegypti* and *Ae. albopictus* were resistant to DDT (4%), but susceptible to malathion (5%), and exhibited varied response to pyrethroids (Table 4). However, larval populations of both these mosquito species were susceptible to larvicides including

malathion (1.0 mg/l), temephos (0.02 mg/l) and fenthion (0.05 mg/l) at much lower dosages than diagnostic concentrations (Table 5). Given the seasonal abundance and case incidence in city areas, it is highly probable that *Ae. aegypti* is the predominant mosquito vector transmitting dengue virus. The study results have direct relevance for benefit of the state dengue control programme in targeting interventions and averting disease outbreaks and spread.

**Table 4. Insecticide susceptibility status of *Ae. aegypti* and *Ae. albopictus*, dengue mosquito vector species in Assam, India (July–October 2013)**

Insecticide (Concen.)	Mosquito species	No. (Replicates) of mosquitoes exposed*	No. (%) of mosquitoes knockdown in 60 min	No. (%) of mosquitoes dead post 24 h exposure	Susceptibility status**
DDT (4%)	<i>Ae. aegypti</i>	70 (5)	1 (1)	2 (2)	R
	<i>Ae. albopictus</i>	60 (5)	7 (12)	8 (13)	R
Malathion (5%)	<i>Ae. aegypti</i>	70 (5)	69 (99)	70 (100)	S
	<i>Ae. albopictus</i>	80 (6)	78 (98)	80 (100)	S
Deltamethrin (0.05%)	<i>Ae. aegypti</i>	101 (8)	96 (95)	81 (80)	R
	<i>Ae. albopictus</i>	100 (7)	100 (100)	100 (100)	S
Permethrin (0.75%)	<i>Ae. aegypti</i>	110 (6)	28 (25)	24 (22)	R
	<i>Ae. albopictus</i>	101 (8)	88 (87)	97 (96)	VR

\*Mortality in control replicates was <5%; \*\*R = Resistant (mortality <80%); VR = Verification required (mortality 81–97%); and S=Susceptible (mortality 98–100%).

**Table 5. Summary statistics of susceptibility test results of mosquito larvae of *Ae. aegypti* and *Ae. albopictus* to various larvicides in Assam, India (June–July 2013)**

Larvicide (concn. in mg/l)	Mosquito species	No. of larvae assayed	Regression equation	Pearson $\chi^2$ goodness of fit	LC <sub>50</sub> (95% CI)*	LC <sub>95</sub> (95% CI)*
Malathion (1.0)	<i>Ae. aegypti</i>	280	$y = 4.91 + 2.85x$	0.029	0.0199 (0.0138–0.0271)	0.0707 (0.0474–0.1562)
	<i>Ae. albopictus</i>	160	$y = 5.48 + 3.1x$	0.410	0.01747 (0.01292–0.02108)	0.0518 (0.0415–0.0764)
Temephos (0.02)	<i>Ae. aegypti</i>	200	$y = 9.67 + 2.28x$	0.010	0.000059 (0.000018–0.000183)	0.000317 (0.000128–0.5883)
	<i>Ae. albopictus</i>	120	$y = 20.52 + 4.5x$	0.427	0.000027 (0.000023–0.000031)	0.000058 (0.000047–0.000083)
Fenthion (0.05)	<i>Ae. aegypti</i>	160	$y = 28.76 + 10.91x$	0.204	0.00226 (0.00216–0.00283)	0.00294 (0.00272–0.00342)
	<i>Ae. albopictus</i>	160	$y = 17.61 + 7.27x$	0.192	0.00374 (0.00340–0.00404)	0.00579 (0.00519–0.00699)

\*LC<sub>50</sub> and LC<sub>95</sub> values are expressed in mg/l.

### A multicentric trial to detect *in vivo* resistance of *Plasmodium falciparum* to artesunate in patients with uncomplicated malaria (TRAC)

*Plasmodium falciparum* is the predominant infection in northeast India with large concentration of cases in tribal population groups located along international border with Bangladesh (Fig. 3). The border is highly porous and it is strongly believed that population movement serve as carriers of drug resistant malaria for which northeast is the corridor for spread in rest of India. To arrest the development and spread of drug resistant strains there is an imperative need to monitor the therapeutic efficacy of present drug regimen (artesunate + SP), artesunate component in particular for possible revision of drug policy for effective treatment. Keeping this objective in mind, the study was undertaken along Indo-Bangla bordering PHCs in



Fig. 3: High risk tribal population located along international border with Bangladesh, Mizoram, India.

Tlabung (Lunglei district) of Mizoram during May–August 2013, and beginning July 2013 the study was initiated and presently continuing in

Silachari PHC (Gomti district) of Tripura in collaboration with the state health authorities. In Tlabung, of total 1962 blood smears screened for malaria parasite, 394 (20%) were positive for malaria parasite, of which *P. falciparum* was the majority infection (83%). A total of 68 cases were enrolled for follow up investigations which comprised all age groups of both sexes (Table 6). In Silachari, of total 1224 blood-smears examined, 126 (10%) were malaria positive of which 80% were *P. falciparum* cases (Table 7). As of October 2013, a total of 31 cases were enrolled at the site for follow up investigations which comprised all the age groups of both sexes.

**Table 6. Prevalence of malaria in Tlabung, Lunglei district of Mizoram (Indo-Bangla border) and subjects enrolled (May–August 2013)**

Age group (yrs)	No. of fever cases examined	Malaria (+)ve cases	<i>P. falciparum</i> cases	No. of subjects enrolled
0–< 1	80	7	5	1
1–4	304	73	58	05
5–15	465	89	76	25
>15	1113	225	188	37
Total	1962	394	327	68

**Table 7. Prevalence of malaria in Silachari, Gomti district of Tripura (Indo-Bangla border) and subjects enrolled (July–October 2013)**

Month	No. of fever cases examined	Malaria (+)ve cases	<i>P. falciparum</i> cases
Jul	107	21	20
Aug	495	61	54
Sep	359	33	22
Oct	263	11	5
Total	1224	126	101

The study results revealed that even though >20% of failure was recorded to artesunate + SP therapy, there was no decrease in sensitivity to artesunate component but instead it is due to SP. Based on these research findings, the drug policy has now been changed to artemether + lumefantrine (AL) for the treatment of *P. falciparum* malaria specific to northeast India. It is now proposed to establish baseline therapeutic efficacy of AL in these border areas.

#### Controlled study of possible adverse effects of DDT, used for indoor residual spraying on human reproductive health with particular reference to lactation and pregnancy outcome

In northeastern states of India, ever since

inception of the malaria control programme, DDT is the main residual insecticide in use against malaria transmitting mosquitoes. To estimate the DDT residue and possible adverse effects of DDT on human reproductive health with particular reference to lactation and pregnancy outcome, sampling of blood samples of follow up pregnant mothers was done keeping pregnant mothers of non-sprayed areas as control group for comparative purposes. GC-MS method for determination of DDT and its metabolites in human milk has been standardized and validated. Preliminary investigations revealed significant differences in concentrations of DDT residues in two population groups. The study has been concluded and data are being analyzed.

#### Ecological succession of anophelines and other mosquitoes in northeastern states of India

The project relates to monitoring, vector abundance in target sites in districts of Assam, Meghalaya, Manipur and Sikkim states of northeast. Systematic mosquito samplings including indoor resting populations, CDC traps and all night mosquito landing catches, breeding resources survey were done in Kamrup and Goalpara districts of Assam and that of East and West Khasi Hills of Meghalaya, and Sikkim during the months of pre-monsoon (March–April) and monsoon (August–September). There are clear indications that populations of *An. minimus* are diminishing and *An. culicifacies* are establishing foothold in northeast states. Data are being analyzed in relation to true prevalence of disease vectors based on GIS/RS technologies. The study has been concluded.

#### Malaria Clinic

It is an ongoing activity in Sonapur Block PHC of Kamrup district much to ensure early case detection and prompt treatment to reduce morbidity and mortality attributable to malaria. During April 2013–March 2014, as many 5829 fever cases were examined for malaria parasite, of which 40 (0.69%) were positive (Table 8). Of these, 38 (95%) were *P. falciparum* cases and the remaining were *P. vivax* infections. All positive cases were administered radical treatment as per the national drug policy, i.e. artesunate + SP combination therapy. It was observed that cases were few and far and disease transmission trends were truly declining owing to multiple interventions

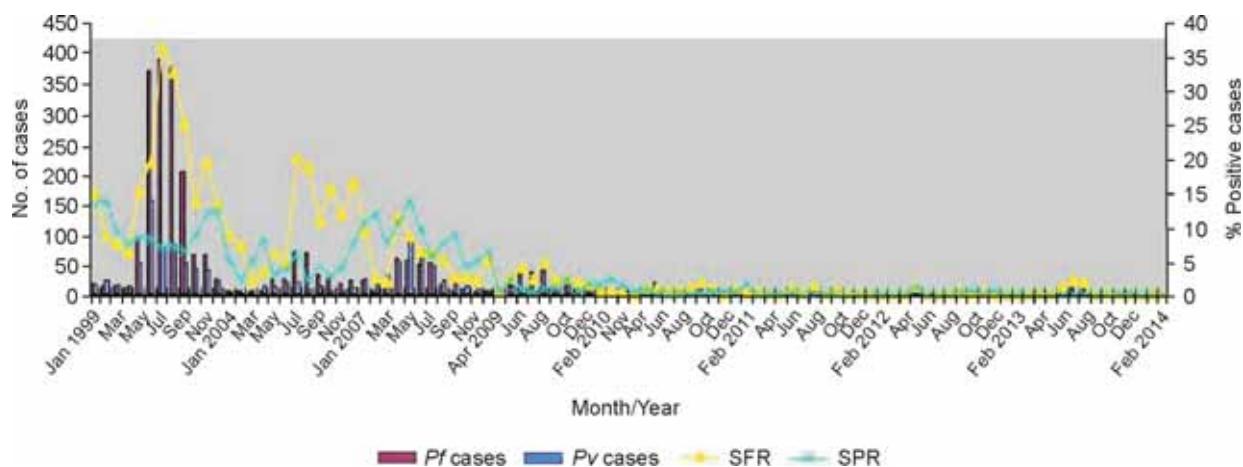


Fig. 4: Showing decline trend of malaria after introduction of ACT and LLINs in Sonapur PHC from 1999 to 2013.

**Table 8. Monthly detection of cases based on passive surveillance (malaria clinic) in the Sonapur Primary Health Centre, Kamrup, Assam**

Age group (yr)	No. of fever cases examined	Malaria (+)ve cases	Pf cases	No. of subjects enrolled
Apr 2013	509	1	1	0.196
May	658	1	1	0
Jun	1223	14	14	0
Jul	495	11	11	0
Aug	436	8	8	0
Sep	506	0	0	0
Oct	441	1	1	0
Nov	490	2	1	0.41
Dec	356	1	0	0.28
Jan 2014	184	0	0	0
Feb	201	0	0	0
Mar	330	1	1	0.30
Total	5829	40	38	0.69

including early case detection and effective chemotherapy and concurrent impregnation of community-owned mosquito nets/ mass scale distribution of long-lasting insecticidal nets for control of disease vectors (Fig. 4). The community compliance is overwhelming and forthcoming.

### Technical support to the Control Programme

- Stocks of both Guppy and Gambusia fish are being maintained in Guwahati metropolis and adjoin suburb of Sualkuchi, an activity supported by NRHM, Assam. These larvivorous fishes were provided to endemic districts of Kokrajhar and Sibsagar, and to various other establishments including defence services as well as adjoining northeastern states. It has also been proposed to extend mass distribution of Guppy fish in Guwahati metropolis and other major

- townships to combat mosquito menace.
- Post MDA assessment for filariasis elimination was undertaken in districts of Darrang and Udalguri during 5–14 June 2013. It was observed that even though coverage was 100% of the eligible population, only 542/622 (87%) admitted for consuming drugs (survey based on 30 houses); of these only 139/542 (25%) consumed drugs right in presence of drug distributors. Only 32/542 (6%) reported side effects which included nausea (13/542), headache (7/542), fever (9/542), and some even (3/542) reported fear of adverse reactions.
  - Served as resource institute for training programme for batches of VBD consultants from northeastern states in May, August and September 2013 and January 2014 where delivered lecture on "Insecticide impregnated nets for control of malaria", "Biological control of mosquito breeding" and Vector bionomics.
  - Participated in an exhibition on "Dengue awareness" in collaboration with the State Health Directorate of Assam held at Guwahati City on 24 September 2013. Displayed dengue mosquito larval breeding habitats and adult mosquitoes for public awareness and prevention and control of dengue.
  - Arranged demo for benefit of nursing student trainees from Down Town Hospital on Blood smear making and staining procedures for malaria microscopy.
  - Served as resource person at Regional review meeting on malaria (IMCP-II) held at Guwahati

- and delivered a lectures on Quality assurance of RTD and Therapeutic efficacy study in Tripura and Mizoram.
7. A batch of ASHA (184 participants) were given demo on Blood smear preparation and drug regimen in different age groups at NIMR clinic maintained at Sonapur PHC.
- Participation in National & International Conferences/Meetings/Workshops**
- Dr Vas Dev**
1. Participated in the 'Brain storming conference on dengue scenario in India: Disease burden, surveillance and control' held at Madurai from 25–26 July 2013, and presented a paper on "Dengue, an emerging arbovirus infection in Assam, northeast India".
  2. Participated in the 'Millennium alliance conference" held at Guwahati on 22 October 2013 organized by FICCI/USAID, and in the FICCI workshop held at Guwahati on 26 December 2013 for DST-Lockhead Martin, Innovation Growth Programme (by invitation).
  3. Participated in the NASI conference held at Goa from 5–7 December 2013 and presented a paper on Epidemiology of malaria transmission in Tripura, northeast India and also served as a member on the panel for judgment of best poster in the student category for the award '*Swarna Jayanti Puruskars*'.
  4. Participated in the International Conference on Entomology held at Panjabi University, Patiala from 21–23 February 2014, and presented a paper on "The dominant mosquito vectors of human malaria in India" and also chaired a session on medical entomology.
  5. Participated in the 6th International Conference on Tropical Medicine and Parasitology held at Kuala Lumpur and presented a paper on "Seasonal abundance of *Aedes (Stegomyia) albopictus* and *Aedes (Stegomyia) aegypti* in Guwahati metropolis and suburban settlements, northeast India" funded by ICMR, New Delhi. Dr Dev also co-chaired a session on 'Forensic science, mosquitoes and vector borne diseases' on 7 March 2014.
  6. Participated in the workshop on "Delaying artemisinin resistance in India" held at New Delhi from 24–25 March 2014 jointly organized by NIMR and Public Health Foundation of India (PHFI).

□

## 3.4

### Hardwar (Uttarakhand)

#### Epidemiological and entomological investigations of dengue infection in Uttarakhand state

Dengue was absent from Uttarakhand state till 2007 and started from Dehradun district with a total of 140 confirmed dengue cases in 2008 and later spread to other districts of the state. Maximum number of dengue cases were recorded in 2010 with a total of 4140 and Dehradun district contributed 2889 (69.9%) of the total. Analysis of all the districts (dengue cases) revealed that only three districts, namely Dehradun, Nainital and Hardwar contributed >95% of total dengue cases reported from Uttarakhand state. An analysis of the data from Himalayan Institute of Medical Sciences, Dehradun revealed that >50% of the dengue cases recorded in hospital were from nearby districts of Uttarakhand State. Maximum number of cases were recorded in 2010, similar to the data reported by the State Health Department. It is to point out that dengue cases recorded from Himalayan Institute of Medical Sciences, Dehradun, were not included in the data report obtained from the State Health Department. Entomological surveys also confirmed high breeding of Aedes mosquitoes. Results of the adult susceptibility test revealed that Ae. aegypti was resistant to DDT while susceptible to malathion and deltamethrin insecticides in Dehradun, Nainital and Hardwar districts of Uttarakhand state. Therefore, it is appropriate to strengthen intervention measures like breeding source survey, antilarval application, use of effective insecticide like malathion/pyrethroids, health education supported by dengue case detection and treatment in Dehradun, Nainital and Hardwar before start of the monsoon (June) itself to check the spread of dengue cases in entire area of Uttarakhand state.

#### Assessment of the efficiency of ASHA workers in delivery of health services in District Hardwar

In spite of the progress made a high proportion of the population especially in rural areas continue to suffer and die from preventable diseases. The Govt. of India launched the NRHM in 2005 under which many innovations have been introduced in the states to deliver health care services in an effective manner to the population. One of the core strategies proposed in this mission was to create a village level social activist designated as ASHA (Accredited Social Health Activists) for every village with 1000 population.

Therefore, a study has been planned for the assessment of the efficiency of ASHA workers in delivery of health services and what are the problems they are facing and to further suggest measures for optimum utilization of their services. In the initial stage, a survey has been carried out and block-wise position of ASHA's has been obtained (Table 1). Out of a total of 1431 posts of ASHA sanctioned in District Hardwar, 1257 ASHAs are presently working.

A questionnaire was prepared and survey had been initiated in Laksar PHC of District Hardwar.

Table 1. Block-wise posting of ASHA in District Hardwar

S.No.	Block	Total number of ASHA sanctioned	Total ASHA in place	Vacant
1.	Narsen	294	292	02
2.	Laksar	150	146	04
3.	Khanpur	40	40	0
4.	Bhagwanpur	179	211	-32
5.	Imlikheda	173	163	10
6.	Bahadarabad	259	253	06
7.	Hardwar (Urban)	222	89	133
8.	Roorkee (Urban)	114	63	51
Total		1431	1257	174

**Table 2. Preliminary data collection**

S.No.	Respondents	No. of respondents	Method of collection
1.	ASHA	12	Interview
2.	ANM	3	Interview
3.	AWW	4	Interview
4.	Community member	7	Interview

Interviews of ASHA workers and the members of the community were conducted (Table 2). Most of the ASHAs interviewed had the profile of young age and better education level. Most of them were at least 8th pass. The most important factor motivating ASHAs for the job was to earn some money. The second most important factor was that the job gave them an opportunity to serve the community. Most of the ASHAs had received training at various levels. All the ASHAs felt that the community considered their job useful. The study is in progress.

### A remote sensing (RS) and geographical information system (GIS) based approach for mapping, monitoring, prediction of mosquito potential and probable determinants of malaria in District Hardwar of Uttarakhand state

Recent studies have shown that satellite images, digitized land use maps and geographic information system are promising for predicting changes in habitats of mosquito vectors. The technique of remote sensing incorporated with GIS will not only overcome the problem of inaccessibility, but will also provide valuable information on spatial and temporal changes in the environment which are introduced mainly, but not exclusively by climate variability. Large areas can be explored in a single time frame and data on land cover/ground conditions conducive for mosquito breeding can be easily obtained. Therefore, a project has been undertaken to prepare cost-effective models for malaria control based on the results extracted

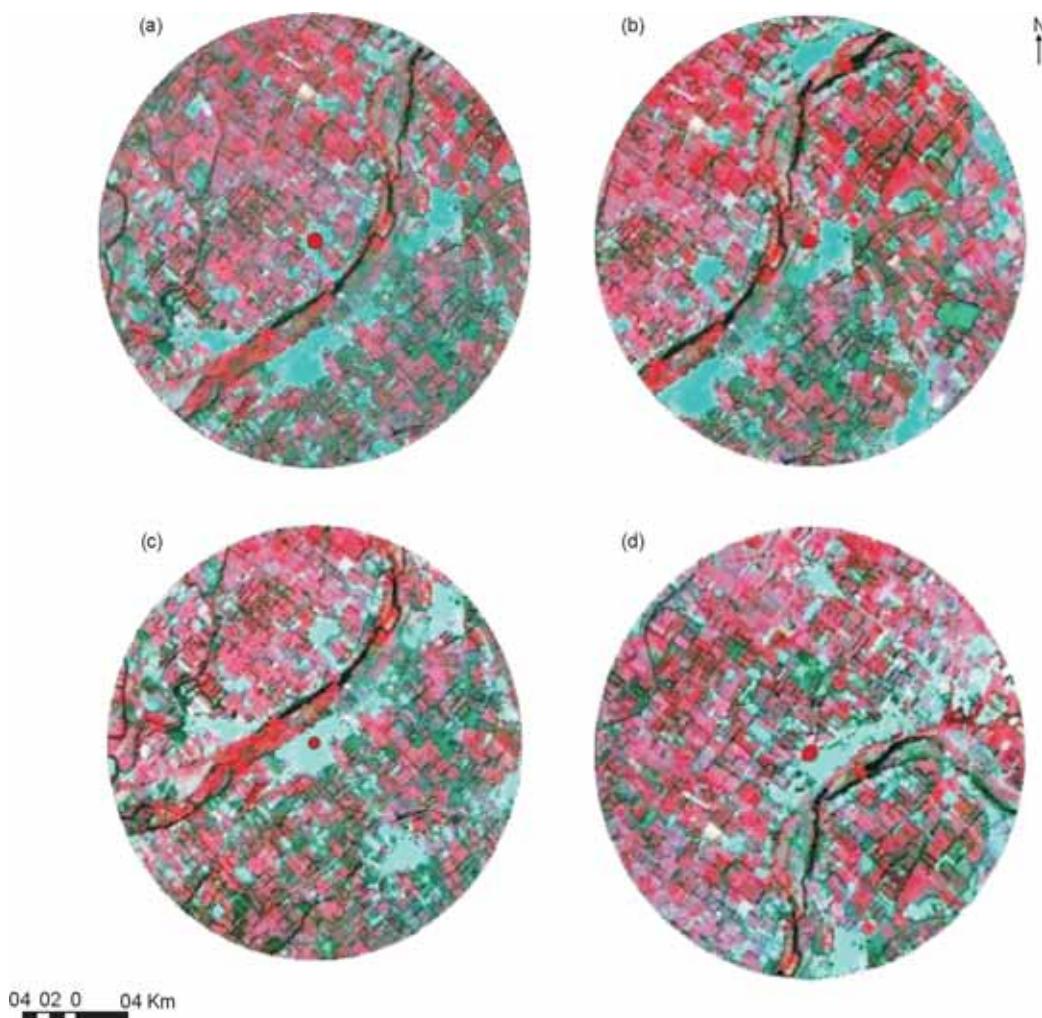


Fig. 1: False colour composite (FCC) in four malarious villages in Laksar CHC: (a) Village Ospur; (b) Purwala; (c) Dargahpur; and (d) Ismilepur.

through RS and GIS in Laksar block of Hardwar district.

During the initial phase, toposheets from Survey of India and Laksar block map were scanned and geo-referenced using Arc GIS 9.3 and ERDAS Imagine 9.1 software. Epidemiological and meteorological data of Laksar PHC for past five years have been collected and tabulated. LISS IV satellite image of Hardwar district for the month of October 2012 was obtained. False colour composite (FCC) of Laksar CHC was subsetted from

LISS IV satellite image (5.8 m resolution) using ERDAS Imagine 9.3 software. Land use/ land cover of laksar CHC was prepared using unsupervised classification.

Based on malaria incidence data of Laksar CHC, four malarious villages, viz. Ospur, Purwala, Dargahpur and Ismilepur (Fig. 1 a, b, c, d) and two non-malarious villages, viz. Dabki Kala and Bhikampur (Fig. 2 a & b) were selected. Area within 1.5 km radius of the malarious and non-malarious villages was extracted by mask using Arc GIS 9.3

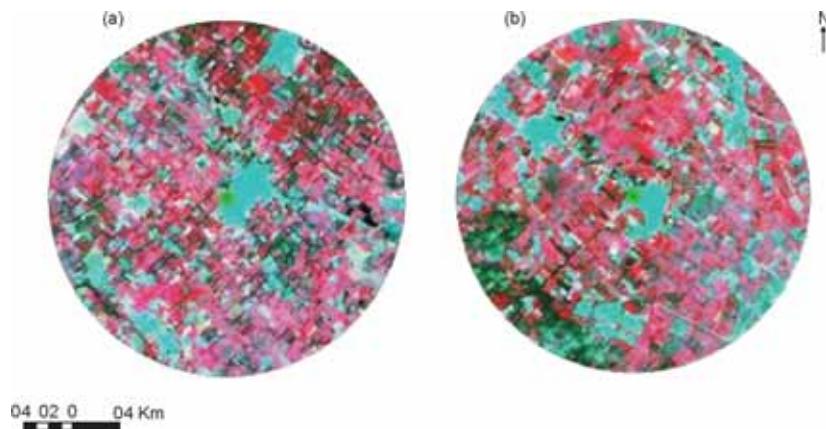


Fig. 2: False colour composite in two non-malarious villages in Laksar CHC: (a) Dabki Kala; and (b) Bhikampur.

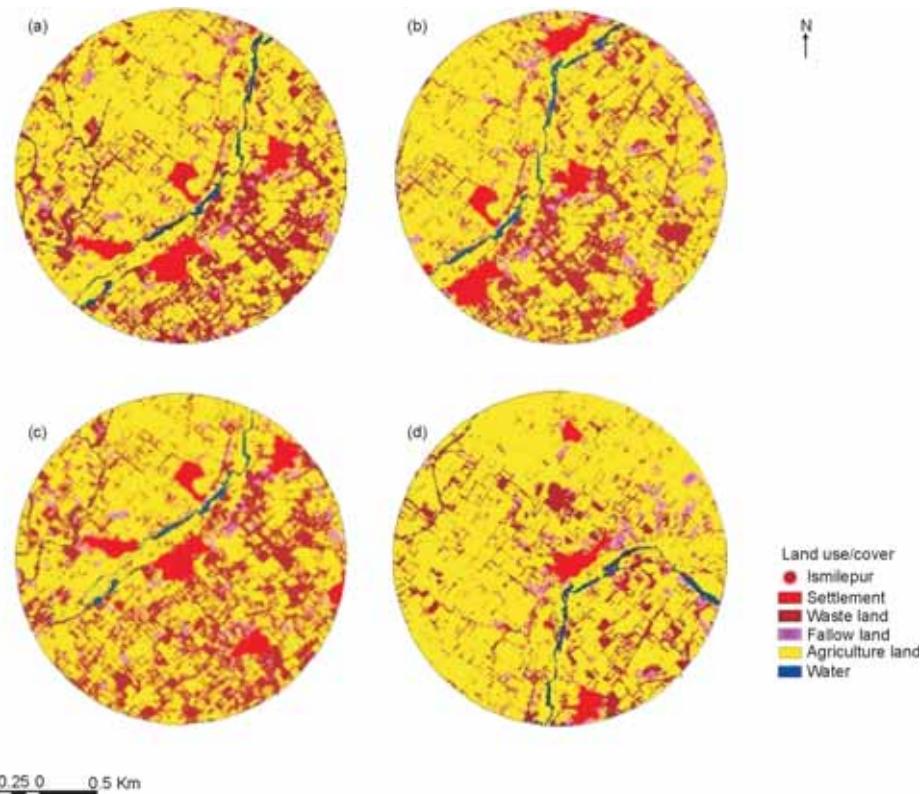


Fig. 3: Landscape use/cover in four malarious villages in Laksar CHC: (a) Village Ospur; (b) Purwala; (c) Dargahpur; and (d) Ismilepur.

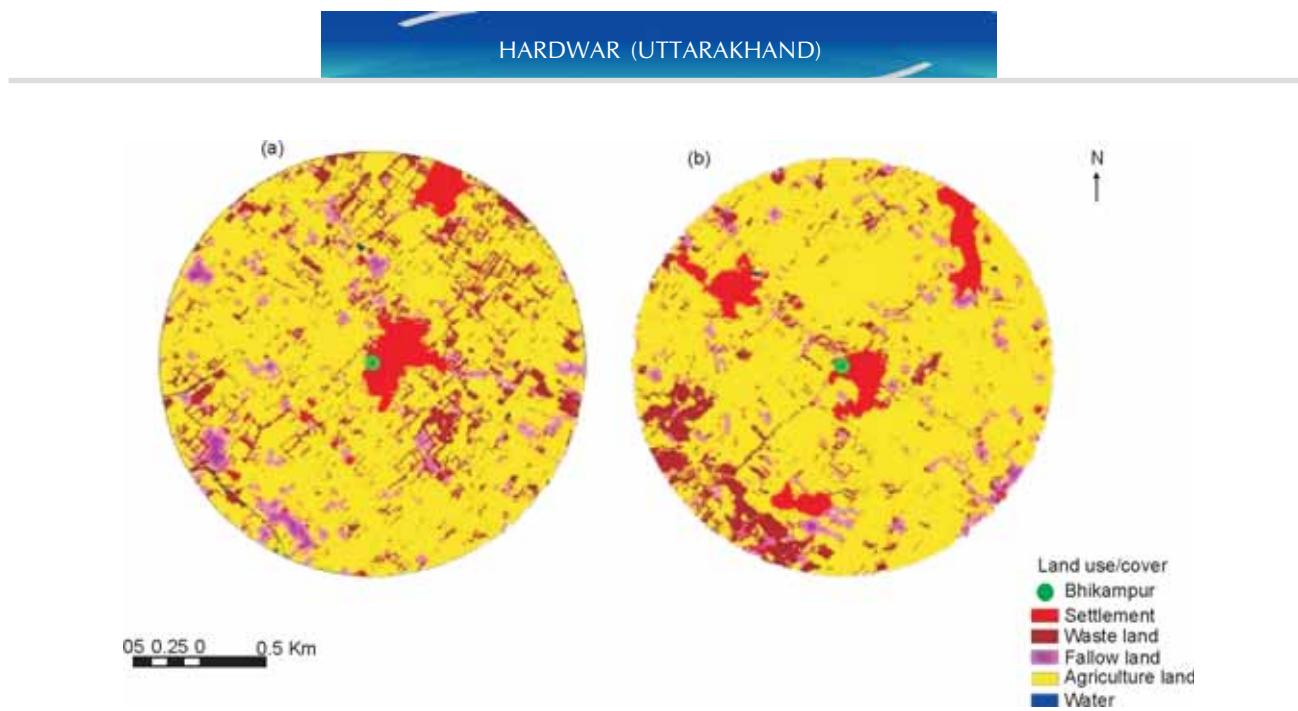


Fig. 4: Landscape use/cover in two non-malarious villages in Laksar CHC: (a) Dabki Kala; and (b) Bhikampur.

software. Unsupervised classification was used for landscape features and classification of malarious (Fig. 3 a, b, c & d) and non-malarious (Fig. 4 a & b) areas with statistics. Using LISS IV satellite image we have been able to generate data of various landscape features and statistics of four malarious and two non-malarious areas. Spot maps of two villages have been prepared for ground truth verification. Preliminary analysis indicates no discernible landscape features in malarious and non-malarious villages. Compilation and analysis of data are in progress.

#### Entomological survey in dengue affected area in Hardwar district

Focal outbreak of dengue was reported in Hardwar district of Uttarakhand state during 2013. Therefore, a survey was undertaken in Hardwar and BHEL township. A door-to-door survey was carried out in houses and peridomestic areas to find out breeding sites of dengue vector. Larval collections of *Aedes* were made from various breeding sites

and brought to the laboratory for species identification. Insecticide susceptibility of dengue vector was carried out against different groups of insecticides. During September/October, a total of 251 houses were surveyed, out of them 175 houses were found positive for *Aedes* breeding. Out of 614 containers searched, 258 containers were found positive for *Aedes* breeding. House, container, breteau and pupal indices were 69.7, 42.0, 102.8 and 41.8, respectively (Table 3). Breeding of *Aedes* at different sites revealed that coolers were major breeding sites in both the areas and >45% coolers were found positive for *Aedes* breeding (Table 4).

Five species of *Aedes*, namely *Ae. aegypti*, *Ae. albopictus*, *Ae. vittatus*, *Ae. pseudotaueniatu*s and *Ae. thomsoni* were identified. In Hardwar, *Ae. aegypti* was most prevalent and percent species composition was >95%. Highest prevalence of *Ae. albopictus* (97.8%) was recorded in the BHEL township. Overall prevalence of *Ae. aegypti* was highest in dengue affected areas (Table 5). *Aedes*

Table 3. Breeding prevalence of *Aedes* in District Hardwar

Area	Houses searched	Houses positive	Containers searched	Containers positive	Positive for pupae	Percent			
						HI	CI	BI	PI
Hardwar	118	101	370	108	42	85.6	29.2	91.5	35.6
BHEL	133	74	244	150	63	55.6	61.5	112.8	47.4
Total	251	175	614	258	105	69.7	42.0	102.8	41.8

HI – House index; CI – Container index; BI – Breteau index; PI – Pupal index.

**Table 4. Breeding of *Aedes* in different breeding habitats in District Hardwar during September–October 2013**

Breeding sites	Hardwar			BHEL		
	No. surveyed	Positive for Aedes	% Positivity	No. surveyed	Positive for Aedes	% Positivity
OHT	53	13	24.5	14	4	28.6
Mud pot	14	7	50	7	6	85.7
Bucket	13	6	46.2	7	6	85.7
Tyre	3	0	0	11	7	63.6
Tank	42	7	16.7	7	4	57.1
Cistern	35	4	11.4	1	1	100
Cooler	88	40	45.5	109	76	69.7
Flower pot	69	20	29	19	9	47.4
Refrigerator	44	5	11.4	23	3	13
Container	4	2	50	36	25	69.4
Drum	1	1	100	5	4	80
Any others	4	3	75	5	5	100
Total	370	108	29.2	244	150	61.5

**Table 5. Species composition of *Aedes* larvae collected from BHEL township and adjoining areas of Hardwar during September–October 2013**

Area	No. of emerged adults identified	Species composition				
		<i>Ae. aegypti</i>	<i>Ae. albopictus</i>	<i>Ae. vittatus</i>	<i>Ae. pseudotaeniatus</i>	<i>Ae. thomsoni</i>
Hardwar	380	96.1	2.6	0.5	0.8	0
BHEL Township	257	1.9	95.3	1.6	0	1.2
Total	637	58.1	40	0.9	0.5	0.5

**Table 6. Insecticide susceptibility of *Aedes aegypti* collected from Hardwar (October 2013)**

Insecticides	No. of mosquitoes exposed	Knockdown in 1 hour exposure period	% mortality
DDT (4%)	40	3	7.5
Malathion (5%)	40	40	100
Deltamethrin (0.05%)	40	40	100

*aegypti* was resistant to DDT as only 7.5% mortality was observed. Exposure of *Ae. aegypti* to malathion and deltamethrin induced 100% mortality indicating that species was susceptible to malathion and deltamethrin (Table 6).

During 2013, a total of 1310 cases of dengue were recorded by rapid Immuno-chromatographic test kit, of which 401 cases were confirmed by ELISA test by the district authorities. Four deaths were reported from dengue (Table 7).

**Table 7. Dengue cases reported in District Hardwar**

Year	Suspected cases	Confirmed cases *	Deaths
2011	144	64	0
2012	368	59	0
2013	1310	401	4

\*ELISA method; (Source: CMO Office)

### Industrial malaria control of BHEL Hardwar

Field Unit, Hardwar is working on industrial malaria control since 1986 and successfully controlled malaria in BHEL, Hardwar. During April 2013 to March 2014, a total of 1641 blood slides were examined, out of which 38 slides were found positive for *P. vivax*, SPR being 2.3 (Table 8).

A total of five anopheles species, namely *An. culicifacies*, *An. fluviatilis*, *An. subpictus*, *An. nigerrimus* and *An. stephensi* were collected during the month of October 2013. MHD of *An. culicifacies* and *An. fluviatilis* was recorded 3 and 0.5, respectively (Table 9). Larval breeding survey

**Table 8. Malaria cases recorded at BHEL, Hardwar**

Year/Month	BS collected	Total positive	Pv	Pf	SPR
Apr 2013	68	—	—	—	—
May	94	2	2	—	2.1
Jun	150	5	5	—	3.3
Jul	178	3	3	—	1.7
Aug	188	7	7	—	3.7
Sep	282	18	18	—	6.4
Oct	393	3	3	—	0.8
Nov	126	—	—	—	—
Dec	52	—	—	—	—
Jan 2014	27	—	—	—	—
Feb	38	—	—	—	—
Mar	45	—	—	—	—
Total	1641	38	38	—	2.3

**Table 9. Man hour density of anophelines in BHEL complex Hardwar (October 2013)**

S.No.	Species	MHD
1.	<i>An. culicifacies</i>	3
2.	<i>An. fluviatilis</i>	0.5
3.	<i>An. subpictus</i>	11.5
4.	<i>An. nigerrimus</i>	1
5.	<i>An. stephensi</i>	0.5

was carried out and fish *P. reticulata* were released into various breeding sites. One fish stock of *P. reticulata* has been established in BHEL township.

#### Evaluation of NetProtect LLIN (impregnated with deltamethrin) against malaria vector in District Saharanpur of Uttar Pradesh

More than 80% mortality was recorded in *An. culicifacies* exposed to NetProtect LLIN washed up to 20 times. Reduction in vector anopheline density was recorded in all the villages where NetProtect was distributed as compared to plain net and no net villages. Community acceptance of NetProtect LLIN was high.

#### Development of plant-based immersion oil for microscopy

In microscopic examination of malaria parasites, oil immersion is a technique used to increase the resolution of a microscope. This is achieved by immersing both the objective lens and the specimen in a transparent oil of high refractive index, thereby increasing the numerical aperture of the objective lens. At the time of using an oil objective, immersion oil plays an important role. In the absence of immersion oil, flat images with little contrast may be observed. Immersion oils are transparent oils that have specific optical and viscosity characteristics and are widely used in the optical system of microscopes, optical measurements and other related fields. Conventional immersion oils typically contain polychlorinated biphenyls (PCBs) which when blended with mineral oil and viscosity adjusting compounds provide generally useful immersion oil having many of the ideal characteristics. In recent years, however, PCBs have been discovered to be carcinogenic, a hazard to the human environment, and are generally regarded as toxic.

One of the criteria of the immersion oil is that its refractive index must match with that of the glass. The refractive index of glass is varied according to

its type and may be summarized as follows. Refractive Index of various glass materials range from 1.472–1.805, whereas pure castor oil (*Oleum ricini*) has 0.953–0.964. Cedar wood oil has a refractive index of 1.495–1.510. The mounting medium normally used are also have the same refractive index. DPX mountant 1.5240 and Canada balsam 1.5250.

The project has been started in June 2012 initially with the specific objectives: (i) Development of cost effective and biodegradable plant-based immersion oil; (ii) The immersion oil obtained from sustainable source (not petroleum based) should be non-toxic to the environment and human beings; (iii) The immersion oil must have a viscosity below 15 cps and preferably around 10–12 cps so that it gives a better image; (iv) The density should be above 0.8 g/cm<sup>3</sup> and preferably approaching 1.0 g/cm<sup>3</sup>; (v) The developed immersion oil must be non-reactive, biodegradable and should not damage the immersion oil objective with its frequent use; and (vi) The immersion oil thus developed must have a potential to be scaled-up.

A total of 30 plants were selected on the basis of physical and chemical properties required for immersion oil. Oil from these plants was extracted, identified as per earlier literature. Briefly, seeds were collected from FRI, Dehradun. The seeds were dried under shade for sometime before use. Seeds of the plants were cracked and the shells were carefully removed. The kernels were grounded by using mechanical method (Mortar and pestle). The oils were extracted in petroleum ether by using Sox let apparatus for 3–4 h. The solvent of the extract was removed by using vacuum evaporator. Seed oils were stored at 4°C for subsequent physiochemical analysis. These oils were tested as immersion oil for the identification of malaria parasite (*P. falciparum*/*P. vivax*).

Results revealed that oils coded—AYR-1, AYR-2, AYR-3 and AYR-4 have shown very good properties as immersion oils. Further studies were carried out only for these four oils. Seven combinations of above four plant oils with different proportions were prepared based on microscopic results examined as immersion oils for microscopic examination of malaria parasites. Results revealed that fractions code AR\_NIMR1, AR\_NIMR2, AR\_NIMR3 and AR\_NIMR7 showed the best oil to be used for microscopic examination of malaria parasite (Fig. 5). Studies were carried out to

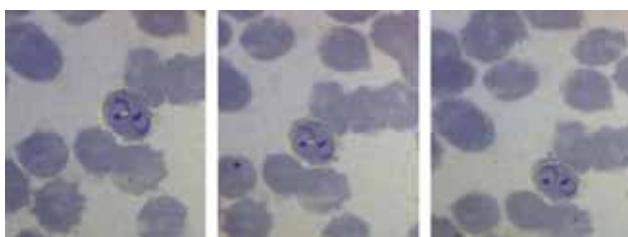


Fig. 5: Comparison of immersion oil with AR\_NIMR1 and AR\_NIMR7 for microscopic identification of *P. falciparum* ring.

investigate stability of oils with time for all four oils and gave similar magnitude of visibility up to five weeks of observation period. Refractive index and density of all oils/blended mixtures were examined by refractometer and relative density bottle (Picnometer). The value of the refractive index and density of oils/blended mixture were in the prescribed range 1.495–1.515 and 0.8–1g/cm<sup>3</sup>, respectively. Experiments are in progress to investigate acid value, viscosity, GC-MS analysis and field evaluation and validation of the developed product in different laboratories.

#### **Synthesis, pKa determination and *in vivo* toxicity of new promising antimalarial 6-methoxy-5,8-di-(4-amino-1-methylbutyl-amino)-quinoline**

6-Methoxy-5,8-di-(4-amino-1-methylbutyl-amino)-quinoline [Indian Patent No. 189970—3280/DEL/98] isolated by peroxydisulfate oxidation of primaquine has been found to possess good gametocytocidal activity against *P. yoelli* infected mice. Project was started in July 2013. Work was mainly divided into two sections, viz. synthesis and *in vivo* toxicity study.

Primary target was to design synthetic scheme for the promising antimalarial “6-methoxy-5-8-di-(4-amino-1)”. Various synthetic strategies were proposed and finally, 7-step synthetic scheme was adopted. Out of 7 synthetic steps, 4 are successfully optimized. Synthesis for the side chain has been obtained. Traced quantity of the 5th step product was confirmed via mass spectrometry, but yet reaction needs to be optimized to get good yield.

On the *in vivo* toxicity front, acute oral toxicity test was performed as per OECD-423 guidelines for primaquine as a standard drug. All the animals were randomly distributed into one control group and primaquine, containing five animals per group. Rats were orally administered single dose of 200, 500 and 1000 mg/kg of suspension of standard drug primaquine. Mortality, signs of toxicity (physical parameter), body weight, food and water

consumption, biochemical, physical and behaviour parameters were observed on 0, 7 and 14 days post-treatment of primaquine. No significant differences were noticed in the body and organ weights and physical and behaviour parameters between the control and 100/200 mg dose primaquine treated groups while 500 mg/kg primaquine treatment group showed some change in physical behaviour pattern like lethargy and tremors. Body weight of all the treated animals increased in a dose dependent fashion with reference to the initial weight as compared to control group, however, in biochemical parameters, increase level of lipid MDA, SGOT, SGPT and AKT in blood serum were recorded but no significant change in blood was found in level of glucose, total cholesterol, triglyceride in blood serum and glucose-6-phosphate dehydrogenase could be observed.

#### **Development of botanical insecticide formulation of essential oils extracted from *Lantana camara* and *Valeriana jatamansi* and *Psoralea corylifolia* for the control of mosquitoes**

The project under top priority projects of ICMR and DRDO, New Delhi has financially supported the project for ₹ 48,97,440 only in collaboration with Dr. Vijay Veer, Director DRL, Tejpur, Assam (COPI) in September 2013. Experimental work was started in October 2013.

From the leaves of *Lantana camara* plant 100 ml oil was extracted, and from roots of *Valeriana jatamansi* plants by using hydro distillation and solvent (n-hexane) extraction process. LD<sub>50</sub>, LD<sub>90</sub>, KD<sub>50</sub> and KD<sub>90</sub> values of *Lantana camara* and *Valeriana jatamansi* plants were reconfirmed by WHO tube adulticide bioassay method with female *An. stephensi*. LD<sub>50</sub> and LD<sub>90</sub> values of *L. camara* were 0.06 mg/cm<sup>2</sup> and 0.10 mg/cm<sup>2</sup> while LD<sub>50</sub> and LD<sub>90</sub> values of *V. jatamansi* were 0.14 mg/cm<sup>2</sup> and 0.24 mg/cm<sup>2</sup>, respectively KDT<sub>50</sub> and KDT<sub>90</sub> values of essential oil of *L. camara* with 0.208 mg/cm<sup>2</sup> (1.5% w/v) impregnated papers were 14 and 25 min, respectively while KDT<sub>50</sub> and KDT<sub>90</sub> values of essential oil of *V. jatamansi* were 17 and 30 min, respectively with 0.28 mg/cm<sup>2</sup> (2.016% w/v) impregnated papers. It is to note that these values were similar to the values reported in our filed patents.

Attempts were made to find out the impact of DEPA (N, N-diethyl phenyl acetamide) a synthetic mosquito repellent developed by DRDO on the

KDT<sub>50</sub> and KDT<sub>90</sub> values on the oils *L. camara* and *V. jatamansi* against *An. stephensi*. A combination of (1.5%) of *L. camara* oil with 2% DEPA in acetone was applied on Whatman filter paper (0.208 mg/cm<sup>2</sup>) and tested against *An. stephensi* as per WHO method. Results revealed that the combination of DEPA in Lantana oil reduced the KDT<sub>90</sub> from 25 to 20 min. Similarly, a combination of (2%) of *V. jatamansi* oil with 2% DEPA in acetone on Whatman filter paper (0.28 mg/cm<sup>2</sup>) reduced the KDT<sub>90</sub> from 30 to 25 min against *An. stephensi*. Experiments against *Ae. albopictus* and *Cx. quinquefasciatus* also revealed that addition of DEPA in oils of *L. camara* and *V. jatamansi* resulted in the reduction of KDT values, thus enhanced the insecticidal activity. It is to point out that DEPA did not show any adulticidal activity against mosquitoes. Attempts are being made to prepare a suitable formulation with the composition consisted of: (i) DEPA plus Lanatana oil in isoamyl alcohol with pressurized gas; (ii) DEPA plus Valeriana oil in isoamyl alcohol with pressurized gas; (iii) Lantana oil in isoamyl alcohol with pressurized gas; and (iv) Valeriana oil in isoamyl alcohol with pressurized gas.

#### **Controlled study of possible adverse effects of DDT, used for indoor residual spraying on human reproductive health with particular reference to lactation and pregnancy outcome**

A project has been undertaken to determine DDT and its metabolites in women who had pre-term delivery and women who had delivered at normal terms from two study areas as exposed to DDT as per National Vector Borne Disease Control Programme, Delhi and without use of DDT or other insecticides for vector control. A total of 678 samples including 243 human blood samples from sprayed area (Sonapur PHC) and 170 human blood samples from non-sprayed area (Satgoan PHC Guwahati) and 165 human milk samples from sprayed and 100 human milk samples from non-sprayed areas were collected during December to January 2011–13. From sprayed area 60 human blood samples and from non-sprayed areas 55 samples while from sprayed and non-sprayed areas 40 human milk samples each were analysed for DDT residues. The mean concentrations of DDT in whole blood from women in pre-term delivery from sprayed and non-sprayed areas were 23.70 and 12.88 µg/l, respectively and the difference was

found statistically significant ( $t=3.065, p<0.01$ ), while the DDT residues in whole blood from post-term delivery subjects were 20.62 and 11.87 µg/l, respectively and also found significantly different ( $t=2.99; p < 0.01$ ). It is to note that the difference of DDT residues in whole blood between pre-term and post-delivery was statistically insignificant. The mean concentrations of DDT in human milk of lactating mothers from sprayed and non-sprayed areas were 15.41 and 10.35 mg/kg, respectively and was found significantly different statistically ( $t=4.11; p < 0.001$ ).

#### **Evaluation of NetProtect LLIN (impregnated with Deltamethrin) against malaria vectors in Uttar Pradesh, Haryana and Jharkhand states**

The present study was proposed in high malarious areas of three states, namely Chhattisgarh, Karnataka and Uttar Pradesh and the observations on different aspects during the intervention and its acceptability by community will yield useful information for its use in disease control. We [Dr VK Dua, PI] have field evaluated through multicentric trials of LLIN incorporated with deltamethrin against malaria vectors in India for two years. Now on the request of Best Net Insect Control it is proposed to monitor the field efficacy of NetProtect for one more year during 2013 for the assessment of net status in relation to time. Field trials were started in May 2011 for LLIN impregnated with deltamethrin against malaria vectors in three villages, namely Chaupura, Bannukheri, and Vazeerpur of Gangoh PHC, Uttar Pradesh. Susceptibility status of *An. culicifacies* revealed that *An. culicifacies* was 100% resistance to DDT while, 95% susceptible to malathion and 100% susceptible to deltamethrin. Results of residual efficacy studies showed >80% mortality of *An. culicifacies* after 3 min exposure and 24 h holding in the intervention period from April 2012 to December 2013. In ring net bioassay, median knockdown time was 16–22 min from April 2012 to December 2013 in the field conditions.

Average per room density of *An. culicifacies* and total anophelines during intervention phase in NetProtect LLIN, plain net and no net villages was 4.6, 9.5 and 8.2, respectively. Reduction of *An. culicifacies* density was observed in LLIN area as compared to plain net area. During pre-intervention period, slide positivity rate was 26.1, 25.5 and 11.4 in NetProtect LLIN, plain net and no net areas,

respectively. SPR during intervention phase was 7, 16.7 and 13.3, respectively, while malaria parasite incidence (MPI) was 3.45, 6.73 and 1.21 during pre-intervention period and 0.38, 1.92 and 0.96 during intervention phase in NetProtect LLIN, plain net and no net areas, respectively. Social acceptability and perceived side effects of NetProtect LLINs were also studied. NetProtect LLINs were well accepted by the communities. Field work at Uttar Pradesh and Jharkhand has been completed while the work at Haryana will complete in 2015.

#### **Technical support provided to the programme**

1. Cross-checking of blood slides for malaria parasites in District Hardwar.
2. Entomological surveillance of dengue vector in District Hardwar.

#### **Other Activities**

##### **Guidance for Ph.D. Thesis**

- Sandeep Kumar (Working, Jiwaji University Gwalior). "Organochlorine residues in soil, water, whole blood and major local food products from low and high malaria endemic areas from Assam" (Supervisors: Dr VK Dua and Dr Rekha Bhaduria).

##### **Patent granted**

- A new plant based insecticide for mosquito control by Dua VK, Alam F, Pandey AC and Dash AP. Indian Patent granted in 2013 (Patent No. 2344279).

##### **Scientific meeting attended**

Dr T Sharma held discussion with the Director General of Health Services, Dehradun in connection with malaria and dengue cases on 9 April 2013.

Dr A Gupta, Dr AC Pandey and Dr NC Gupta discussed with the Chief Medical Officer, BHEL in connection with the ongoing vector control measures in the township area on 16 August and 20 September 2013.

Dr T Sharma discussed with the Chief Medical Officer Saharanpur on 10 September 2013 and Dr NC Gupta with DMO, Saharanpur on 14 March 2013 in connection with the data collection of malaria, dengue and J.E.

Dr NC Gupta made on spot survey in different blocks of BHEL factory from 19 August to 22 September, 2013 and discussed with the concerned AGM's for immediate dengue larval control measures.

Dr A Gupta, Dr AC Pandey, Dr T Sharma and Dr NC Gupta with the Chief Medical Officer, Hardwar in connection with dengue in Hardwar on 20 September 2013.

Dr A Gupta discussed with Township & Estate Incharge in connection with the establishment of larvivorous fish hatchery on 29 October 2013.

Dr A Gupta, Dr AC Pandey and Dr NC Gupta discussed with the Chief Medical Officer, BHEL in connection with the establishment of larvivorous fish hatchery on 7 March 2014.

##### **Technical Reports submitted**

1. Evaluation of PermaNet® 3.0 LLIN against malaria vectors resistant to pyrethroids in Madhya Pradesh, Chhattisgarh and Jharkhand states.
2. Phase-III evaluation of 2% tablet and 2% granule formulations of diflubenzuron (Bi-Larv®), an insect growth regulator against larvae of mosquito vectors.
3. DDT residues in human milk, whole blood and adipose tissues from DDT sprayed areas and areas where DDT not sprayed.



## 3.5

### Jabalpur (Madhya Pradesh)

#### Assessment of durability of LifeNet® Long-lasting insecticidal nets in a phase-III study in Madhya Pradesh, India

The general purpose of the study is to undertake WHOPEs Phase-III evaluation of LifeNet® long-lasting nets (LN) according to standard WHO guidelines and procedures under three years of continuous use in field conditions with the specific objectives: To determine and compare durability (survivorship and fabric integrity) of LifeNet® LN to a WHOPEs fully recommended LN (i.e. PermaNet 2.0) and to a deltamethrin incorporated into polyethylene LN (i.e. NetProtect) as positive controls; and to assess insecticidal activity of LifeNet® LNs compared with two positive controls over 3-years as these are routinely used by people in their local settings.

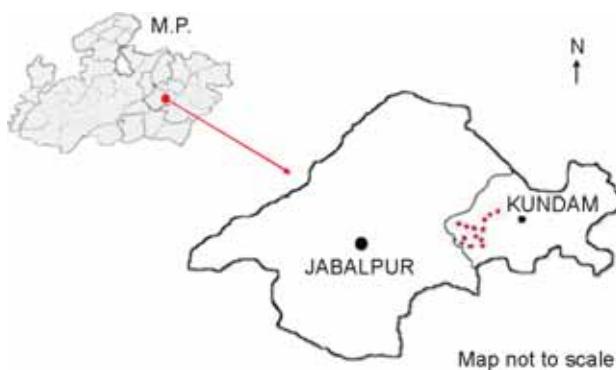


Fig. 1: Map of Madhya Pradesh showing study villages in Kundam, CHC Jabalpur.

The household (HH) census of 13 villages in Kundam CHC, Jabalpur (Fig. 1) covering 1530 households and 6701 population was completed in October 2013. These HH are mainly farmers (71%) and tribals (79%) having 1 or 2 sleeping rooms in their houses. A total of 92% HH know that bednets are used to avoid mosquito bites. Only 5% HH had their own nets before distribution of

study nets.

A total of 10 meetings with the villagers and Sarpanch of all the 13 villages and with health staff (health worker, malaria inspector, block medical officer of Kundam CHC and district malaria officer of Jabalpur district) were organized. They were informed about the study objectives and methods.

For insecticidal activity, 30 each of three types of nets (LifeNet, NetProtect and PermaNet 2.0) were sampled and sent to Gambloux, Belgium for chemical analysis. Reports showed that deltamethrin content in the 3 LNs are within WHO specification tolerance limits.

All the nets were given code numbers (Fig. 2) once with wash resistant ink and a spot with water soluble ink was made for assessment of washing. A random list of HH was made to allocate to LifeNet, PermaNet 2.0 and NetProtect for distribution of different purposes, i.e. bioassay, chemical assay, durability and fabric integrity. A total of 3568 LNs (1202 LifeNet, 1192 NetProtect and 1174 PermaNet 2.0) were distributed in 1530 houses (Fig. 3).



Fig. 2: Coding of long-lasting insecticide-treated nets before distribution.



Fig 3: Distribution of long-lasting insecticide-treated nets to the villagers.

Monitoring of adverse effects at week 1 and 1 month after net distribution was done using questionnaire. Users of 30 randomly selected coded nets of each type were selected for this purpose in each survey. The adverse event reported by the net users at week 1 was higher in LifeNet LNs (47%) as compared to NetProtect (20%;  $p=0.027$ ) and PermaNet 2.0 (17%;  $p=0.013$ ). Users mainly complained ( $>70\%$ ) about skin itching, facial burning, eye irritation and bad smell. The adverse effects reported at month 1 after net distribution were almost nil.

Insecticide susceptibility tests were carried out to assess the susceptibility of targeted disease vector *An. culicifacies* of study area against deltamethrin 0.05%, using the WHO kit and method. Results of 30 replicates revealed 98% corrected mortality of *An. culicifacies* in 24 h with 88% knockdown (Table 1). It shows the susceptible status of *An. culicifacies* in this area.

### Translational research activities

#### To determine the effectiveness of intensive intervention measures on malaria prevalence in tribal district, Dindori, Madhya Pradesh

Investigations were carried out in villages of

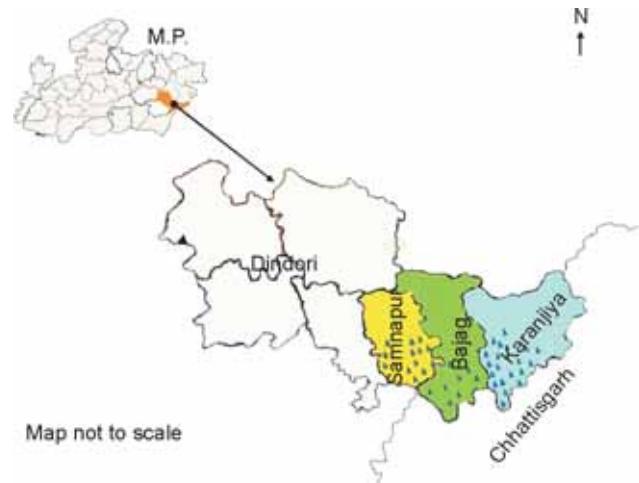


Fig. 4: Map of Madhya Pradesh showing study area: Baiga Chak of District Dindori.

Baiga Chak area, District Dindori of Madhya Pradesh (Fig. 4) to evaluate new intervention measures for developing a suitable model for forest malaria control in similar terrain in collaboration with the State Vector Borne Disease Control Programme.

Rapid fever surveys carried out in the year 2013–14 revealed a total of 71 malaria positive cases (62 Pf, 8 Pv and 1 mixed of Pf and Pv) out of total 2201 fever cases screened. The slide positivity rate (SPR), slide falciparum rate (SFR) and *P. falciparum* percent ( $Pf\%$ ) were 3.2, 2.9 and 88.7%, respectively (Fig. 5). There was 72% decline in SPR and SFR in 2013 when compared with baseline. The spleen rate in children was only 5% in the year 2013.

The anopheline fauna of the villages consisted of mainly *An. culicifacies*, *An. subpictus*, *An. fluviatilis* and *An. annularis* in indoor resting collection, total catch and light trap catches. The mean density of *Anopheles* caught per man hour during the year (Fig. 6) was 10.9, of which the density of *An. culicifacies* and *An. fluviatilis* was 7.7 (70.6%). In total catch, the numbers per catch of total anophelines was 11.3, of which *An. culicifacies* and *An. fluviatilis* was 8.0 (70.8%). During light trap catches the per trap catch of total anophelines was 10.1 in indoors and 11.1 in outdoors.

Table 1. Insecticide susceptibility status of adult *An. culicifacies* in Kundam CHC villages

Insecticide	No. of replicates	No. of mosquitoes exposed*	Knockdown in 1 h	No. dead in 24 h	% mortality	Corrected % mortality
Deltamethrin (0.05%)	30	450	397	440	97.8	97.7
Control	12	180	0	9	5	

\*15 mosquitoes were exposed for 1 h in each replicate.

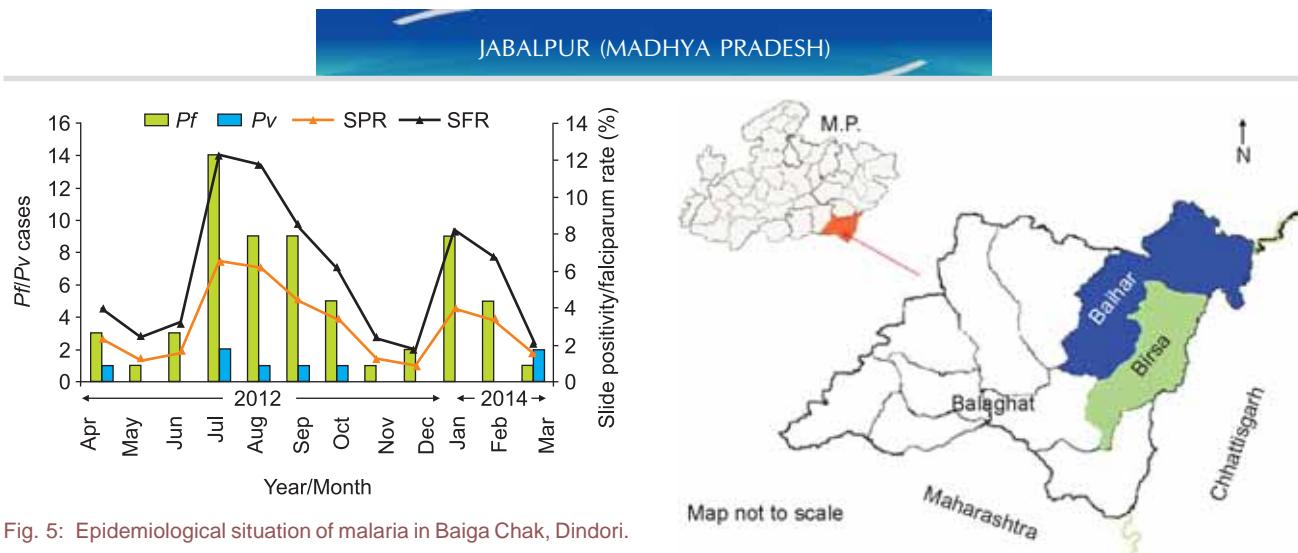


Fig. 5: Epidemiological situation of malaria in Baiga Chak, Dindori.

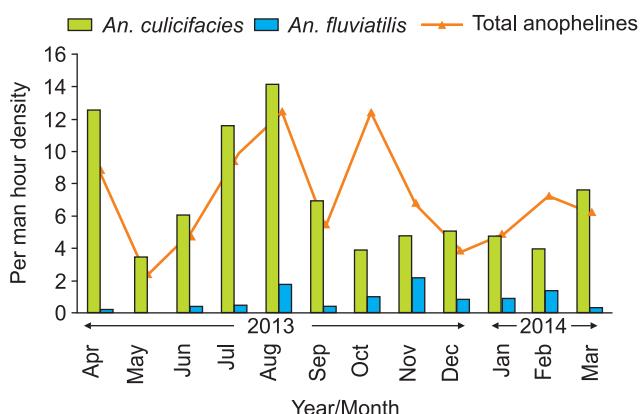


Fig. 6: Per man hour density of mosquitoes in Baiga Chak, Dindori (April 2013–March 2014).

During this year, 1102 *An. culicifacies* and 108 *An. fluviatilis* were tested for vector incrimination by ELISA, of which none was reactive to CS protein of *Pf* or *Pv*.

#### **Assessment of the effectiveness of intensive intervention measures on malaria control programme in tribal district, Balaghat, Madhya Pradesh**

This study was undertaken in collaboration with the State Vector Borne Disease Control Programme in Balaghat district (Fig. 7). Durable well-lining (DWL) were fixed in January 2013 in six villages covering 3300 population (provided by Vestergaard Frandsen). This is deltamethrin incorporated plastic sheet fixed on the inside wall of the houses. Eight villages with a population of 3500 were kept as control. Fever surveys carried out during the year 2013-14 revealed that a total of 816 fever cases were screened, of which 130 were found positive for malaria (120 *Pf* and 10 *Pv*). The SPR, SFR and *Pf*% were 15.9, 14.7 and 92.3%, respectively. However, in control villages (CTL) where no DWL was installed, malaria prevalence was higher



Fig. 7: Map of Madhya Pradesh showing study area in District Balaghat.

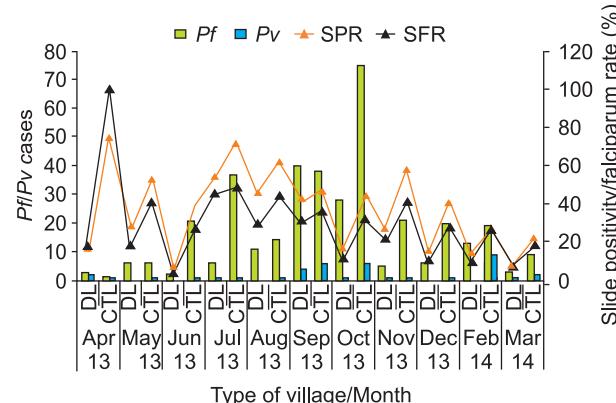


Fig. 8: Epidemiological situation of malaria in Balaghat after distribution of durable lining.

( $p < 0.0001$ ). A total of 293 malaria positive cases (261 *Pf*, 30 *Pv* and 2 mixed of *Pf* and *Pv*) were found out of 885 screened. The SPR, SFR and *Pf*% were 33.1, 29.7 and 89.8 %, respectively (Fig. 8).

During the year, in DWL villages, 435 *An. culicifacies* and 10 *An. fluviatilis* were tested for vector incrimination by ELISA, of which none was detected sporozoite positive. However, in control villages, four *An. culicifacies* were found sporozoite positive (2 *Pf* and 2 *Pv*) out of 1399 assayed (sporozoite rate 0.28%).

#### **Studies under Vector Science Forum**

#### **Bionomics of malaria vectors and their sibling species, and to establish their role in malaria transmission in Chhattisgarh, India**

The study was initiated in October 2013 in two malarious districts Bastar and Koria of Chhattisgarh state (Fig. 9). Two CHCs in the district and 4 villages in each CHC were selected for this study. The

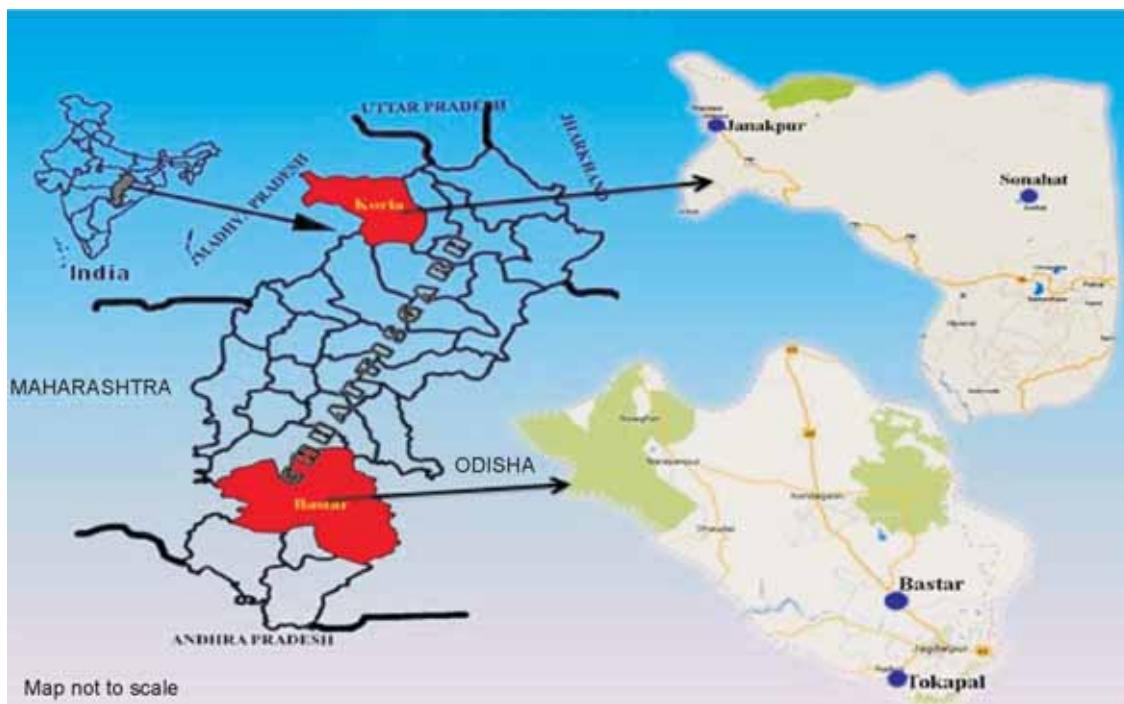


Fig. 9: Map of Chhattisgarh showing study sites of Districts Korea and Bastar.

overall objective is to study the bionomics of prevalent malaria vectors and their role in malaria transmission for development of evidence-based sustainable malaria control strategy with special reference to vector control.

Susceptibility of *An. culicifacies* to diagnostic dose of DDT (4%), malathion (5%), deltamethrin (0.05%) and alpha-cypermethrin (0.1%) was studied in both the districts. The corrected mortality in Korea was <15% to DDT (R), 75% to malathion (R), around 90% to deltamethrin tolerant (VR) and 100% to alpha-cypermethrin in different localities. In Bastar district, the corrected mortality was <10% to DDT, about 75% to malathion, <90% to

deltamethrin and <90% to alpha-cypermethrin (Table 2). These results indicate that the species is resistant to DDT and malathion and tolerant to deltamethrin in Bastar.

Blood meal test was done to determine the host feeding preference. Results revealed almost 100% positive for bovine blood except two samples of Bastar district which showed human blood positive.

The anopheline fauna of the villages in both the districts consisted of mainly *An. culicifacies*, *An. subpictus*, *An. fluviatilis*, *An. annularis* and *An. vagus* in indoor resting collections. The mean density of *Anopheles* caught per man hour during

Table 2. Susceptibility status of *An. culicifacies* against various insecticides in different ecotypes

District	Insecticide	No. of mosquitoes		% Mortality		% Corrected mortality	
		Forest	Foothill	Forest	Foothill	Forest	Foothill
Korea	Alpha-cypermethrin (0.1%)	120	120	100	100	100	100
	Deltamethrin (0.05%)	120	120	91.2	94.2	91.2	94.2
	Malathion (5%)	120	120	71.7	76.7	71.7	75.8
	DDT (4%)	120	120	12.5	13.3	12.5	13.3
Bastar	Alpha-cypermethrin (0.1%)	120	120	87.5	89.2	87.1	89.2
	Deltamethrin (0.05%)	120	120	86.7	86.7	86.2	86.7
	Malathion (5%)	120	120	74.2	79.2	73.3	79.2
	DDT (4%)	120	120	7.5	5.8	7.5	5.8

\*15 mosquitoes were exposed for 1 h in each replicate.

## JABALPUR (MADHYA PRADESH)

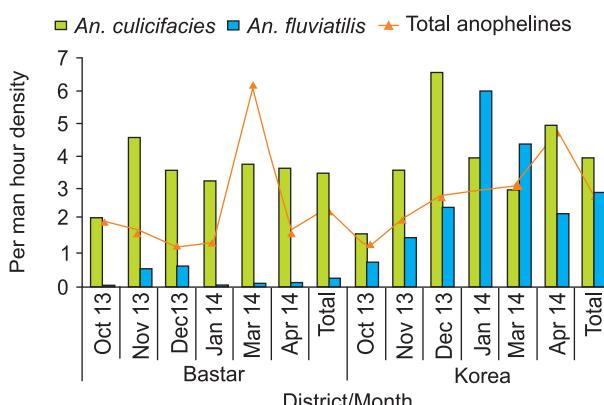


Fig. 10: Per man hour density of mosquitoes in Chhattisgarh.

the year (Fig.10) was 13.7 in Bastar district and 16.1 in Korea, whereas the density of *An. culicifacies* and *An. fluviatilis* was 27.4% in Bastar and 42.2% in Korea.

Rapid fever surveys carried out during the period revealed a total of 30 malaria positive cases (26 *Pf* and 4 *Pv*) out of total 477 fever cases screened from both the districts. The SPR, SFR and *Pf* % were 6.3, 5.5 and 86.7%, respectively. District-wise data show more malaria positive cases in Korea (SPR 10.5) as compared to Bastar (SPR 1.0).

Species-specific breeding site surveys revealed high breeding of anopheline mosquitoes in streams and seepage water in Korea district from which mainly five species, viz. *An. culicifacies*, *An. subpictus*, *An. fluviatilis*, *An. theobaldi* and *An. jeyporiensis* emerged. The percent emergence of *An. culicifacies* and *An. fluviatilis* was >22%.

### Technical support to the programme

#### Malaria outbreak epidemic investigation in District Katni, Madhya Pradesh

A research team investigated malaria outbreak in six villages of Dhimarkheda CHC of Katni district in the year 2013 on the request of the State Government. Rapid fever surveys, spleen surveys and mosquito collections were done in the notified area. A total of 270 febrile cases were screened for malaria by bivalent RDT kit of malaria, of which 82 cases were found positive. SPR, SFR and *Pf*% were 30.4, 26.7 and 87.8%, respectively. *Plasmodium falciparum* (72 cases) was main cause of febrile sickness followed by *P. vivax* (10 cases). The area was surrounded by thick deciduous forest. All the affected villages were unreachable and situated about 30 to 40 km away from CHC HQs.

Spleen survey revealed that spleen rate was 20% only. All the malaria positive cases were treated by ACT and chloroquine according to respective malaria species. The report of survey was communicated to CMO&H of District Katni for further perusal. District health administration started organization of medical health camps and focal IRS of synthetic pyrethroid in the affected villages.

#### Nutritional and health related study among Baiga tribe residing in Pushprajgarh CHC of Anuppur district of Madhya Pradesh

During the period, five visits were made in Baiga dominated villages of Pushprajgarh CHC of Anppur district in the months of May, July, August and October. A total of 756 fever cases were screened, of which 157 cases were found positive for malaria (Table 3). *Plasmodium falciparum* (146 cases) was dominating parasite species followed by *P. vivax* (10 cases) and only one case was mixed infection of *Pf* and *Pv*. SPR, SFR and *Pf*% were 20.8, 19.4 and 93.6%, respectively. It was reported to BMO Pushprajgarh for strengthening of active surveillance and treatment in malaria affected areas by distribution of RDT and ACT among peripheral health workers (ANMs, MPWs and ASHAs). The DMO and CMO were also intimated for further vector control measures.

Table 3. Report of rapid fever sueveys, conducted in Pushprajgarh CHC of Anuppur district during the year 2013-14

Months	BSE	Pos	Pv	Pf	SPR	SFR	Pf%
May	203	64	4	60	31.50	29.60	93.80
Jul	199	22	4	18	11.10	9	81.80
Aug	35	2	0	2	5.70	0	0
Oct	127	32*	1	30	25.20	24.40	96.90
Jan	192	37	1	36	19.20	18.75	97.29
Total	756	157	10	146	20.45	16.65	83.11

\*One mixed (*Pf* + *Pv*).

#### Meetings/Workshops/Conferences/Trainings/Symposia attended/organized

##### Dr Neeru Singh

- Organized a Tribal Health Research Forum quarterly meeting on 15 April 2013 as Coordinator, THRF at VCRC, Puducherry.
- Attended Brain-storming meeting under Vector Science Fourm at ICMR HQs, New Delhi on 1 May 2013.

3. Attended Malaria Group meeting at NIMR, New Delhi on 3 May 2013.
4. Attended Expert Group meeting on Insecticides at NIMR, New Delhi on 8 May 2013.
5. Attended Task Force meeting to review protocols received under Task Force on Insecticide resistance monitoring in different disease vectors at NIMR, New Delhi on 10 May 2013.
6. Attended second Malaria RTAG meeting as Temporary Adviser to the Regional Director WHO SEARO at New Delhi from 14–18 May 2013.
7. Attended a Task Force meeting on Biology and bionomics of malaria and dengue/chikungunya vectors under Vector Science Forum at ICMR, New Delhi on 20 May 2013.
8. Attended third Iron and Malaria Research Review Committee (RRC) meeting at Rockville, Maryland, University of Toronto, Canada from 13–14 June 2013. Delivered a lecture and discussion on Indo-Canada collaborative project from 15–18 June 2013 at Rockville, Maryland.
9. Attended Project Review Committee meeting on Tribal Health Research (ECD) at Bengaluru from 24–25 June 2013.
10. Attended Vector Science Forum meeting at ICMR HQs, New Delhi on 24 July 2013.
11. Attended project Review Committee meeting of Tribal Health (ECD) at ICMR, HQs, New Delhi on 25 July 2013.
12. Attended 4th Annual Meeting of THRF at DMRC, Jodhpur from 8–11 August 2013.
13. Attended meeting regarding Regulatory check for issuing commercial licence of Malaria Rapid Diagnostic Tests at Central Drugs Standard Control Organization, New Delhi from 9–11 September 2013.
14. Attended meeting on Priority areas of research for malaria, leishmaniasis, filariasis, dengue, chikungunya and Japanese encephalitis under Vector Borne Disease Science Forum at ICMR HQs, New Delhi from 8–10 October 2013.
15. Attended meetings at Geneva, Switzerland: (i) World Health Organization (WHO) Global Technical Strategy (GTS) for Malaria Control and Elimination 2016–2025 Country Typology meeting from 14–15 October 2013; (ii) Roll Back Malaria Partnership Work Plan (PWP) Dialogue meeting on 16 October 2013; and (iii) Operational Research Planning meeting from 17–18 October 2013.
16. Attended PRC meeting of Tribal Subplan at ICMR, New Delhi on 5 November 2013.
17. Attended Task Force meeting on Biology and Bionomics of Vectors at MoHFW, Gol, Consultant, NRHM on 21 November 2013.
18. Attended meeting of Need for regulatory check for issuing commercial license to manufacturers of Malaria Rapid Diagnostic Tests" at ICMR, New Delhi on 18 December 2013.
19. Attended Expert Group meeting to review proposals on insecticide at ICMR, New Delhi on 20 December 2013.
20. Attended brainstorming meeting on diseases in elimination phase at Tanda, HP from 27–28 December 2013.
21. Attended annual review meeting at ICMR HQs, New Delhi to review the progress of the VDLs at RMRIMS, Patna, RIMS, Ranchi, KIPM, Chennai, SMS, Jaipur and RMRC, Jabalpur on 28 January 2014 and regarding establishment of new laboratory at Raipur and Field Unit visit to District Hospital, Korea and Janakpur from 29–30 January 2014.
22. Attended official meeting with the Principal Secretary, Ministry of Tribal Welfare, Govt. of Madhya Pradesh, Bhopal on 12 February 2014.
23. Attended joint monitoring mission meeting at WHO-SEARO on Vector Borne Diseases in India from 1–10 March 2014.

#### **State workshops conducted**

Two training workshops for 42 Medical Officers of 12 districts from Madhya Pradesh on Vector borne diseases were organized in the months of December 2013 and March 2014 at Jabalpur. The workshops were organized jointly by RMRCT, NIMR Field Unit, Jabalpur and Directorate of Health Services, Bhopal under Enhanced Vector Borne Disease Control Programme (EVBDPC). Professors of Medical College, Jabalpur, Scientists of NIMR, RMRCT and State Health Officers imparted training



Fig. 9: Training workshop on malaria and other vector borne diseases for Medical Doctors at NIMR Field Unit, Jabalpur.

on different aspects of malaria and other vector borne diseases to Medical Officers.

#### **Guidance to Ph.D.**

Mr Mrigendra Pal Singh, Technical Assistant, has been registered for Ph.D. under the guidance of Dr KB Saha, Scientist 'D', RMRCT, Jabalpur (Topic: Economic burden of malaria in low and high malaria transmission areas of Madhya Pradesh: A microeconomic approach) in the Department of Economics (Faculty of Social Science), Rani Durgavati University, Jabalpur, Madhya Pradesh.

□

# 3.6

## Nadiad (Gujarat)

### Health impact assessment of development project: Impact of Sardar Sarovar project on vector borne diseases in Gujarat

This study was carried out in Phase-II Command area of Sardar Sarovar project in Kheda, Surendranagar and Patan districts. The activities in districts included mosquito collections, peridomestic larval survey and domestic water container survey for mosquito breeding, host preference and survivorship of malaria vector, cross-section mass blood surveys in sentinel villages and analysis of malaria surveillance data from the districts. Apart from these, assessment of performance of ASHAs in command area was also done.

Malaria cases have declined in 2013 (58,451) in the state as compared to 2012 (76,223) except in three districts. In general, malaria cases in command and non-command areas have declined but the proportion of *P. falciparum* has marginally increased in command area (Figs. 1 and 2). Malaria incidence in districts of Phase-II Command and non-command areas is shown in Fig. 3. In command (8) and non-command (3) sentinel villages, cross-sectional mass blood surveys carried out in pre- and post-monsoon seasons revealed low malaria parasite load in the community. During both periods slide positivity rate was higher in command area. Altogether, 4135 blood smears (command: 2772 and non-command: 1363) were examined for malaria by microscopy (Table 1). Slide positivity

rate was higher in command area in both the seasons. Age-wise distribution of malaria cases in pre-monsoon period showed more malaria

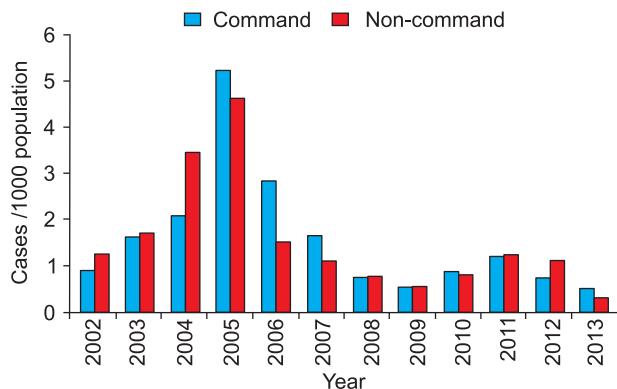


Fig. 1: Secular trend of malaria in command and non-command areas.

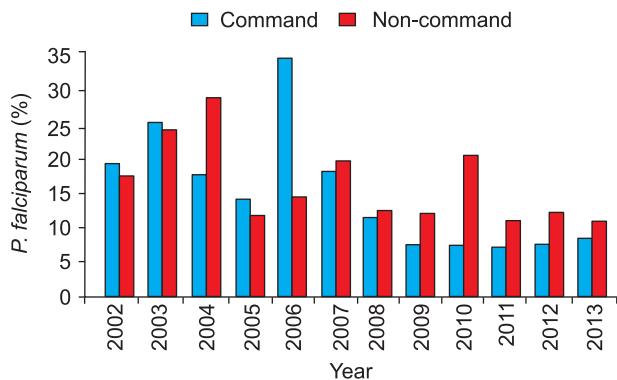


Fig. 2: Proportion of *P. falciparum* in command and non-command areas.

Table 1. Malaria incidence in command and non-command areas

Period	Area	Population	BSE	Cases	Pv	Pf	SPR
Pre-monsoon	Command	11000	1916	8	8	0	0.42
	Non-command	6868	894	2	1	1	0.22
Post-monsoon	Command	11000	856	3	2	1	0.35
	Non-command	6868	469	1	1	0	0.21

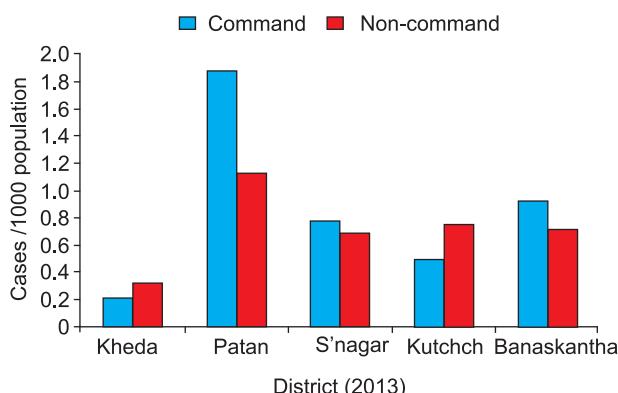
BSE – Blood smears examined; Pv – *P. vivax*; Pf – *P. falciparum*.

## NADIAD (GUJARAT)

**Table 2. Age-wise distribution of malaria cases in command and non-command areas (pre-monsoon)**

Age group (yr)	Non-command				Command			
	BSE	Pv	Pf	SPR	BSE	Pv	Pf	SPR
<1	5	0	0	0	6	0	0	0
1–4	33	0	0	0	85	0	0	0
5–9	38	0	0	0	140	0	0	0
10–14	118	0	0	0	312	2	0	0.64
≥15	700	1	1	0.28	1373	6	0	0.43
Total	894	1	1	0.22	1916	8	0	0.42

BSE – Blood smears examined; Pv – *P. vivax*; Pf – *P. falciparum*.



**Fig. 3: Annual parasite incidence in certain districts in SSP Phase-II area.**

infection in the age group of 10–14 years in the command area (Table 2).

The human filarial infection is confined to only 10–11 districts in coastal area, except small foci in Vadodara. The Gujarat state reported 6272 dengue cases and 15 deaths; and 1157 chikungunya cases in 2013.

An assessment of services of ASHA in command area of 12 villages in three districts was carried out to assess their knowledge and skill (Table 3). It was

observed that all of them were trained at PHC/sub-centre, and collected blood smears from fever patients and sent slides to the PHC. The results showed inadequate supervision by MPHW and other workers. The results are not conveyed in time to them. The fortnight door-to-door visit was done only by 8.3% of ASHAs.

The geographical reconnaissance of mosquito breeding habitats showed that anophelines and culicines were mainly found breeding in the peridomestic breeding habitats. In command area, nine mosquito species and in non-command area eight species were recorded. *Anopheles fluviatilis* spp was found breeding only in command area (Table 4). *Culex vishnui* and *Cx. quinquefasciatus* were predominant species in both the areas followed by *An. subpictus*. Among the vectors of malaria, filariasis and JE, *An. culicifacies*, *An. fluviatilis*, *Cx. quinquefasciatus* and *Cx. vishnui* were found. During intra-domestic surveys in sentinel villages altogether 1014 and 505 houses were examined for mosquito breeding in command and non-command areas, respectively (Table 5). Seasonal prevalence of *An. culicifacies* in

**Table 3. Assessment of performance of ASHA in command area**

S.No	Questions	Response (n = 12 villages)		
		Answer	Nos.	%
1.	Are you provided training by PHC/sub-centre?	Yes	12	100
2.	Are you using RDT?	Yes	11	91.6
3.	Are you collecting blood smears?	Yes	11	91.6
4.	Are you collecting blood smears of all the fever cases?	Yes	12	100
5.	Slides transported to PHC by whom?	Self	12	100
6.	How slide results are conveyed to you (ASHA)?	Self	12	100
7.	What is time lag between BSC and result received (average)?	2–3 days	12	100
8.	Record maintained by ASHA	Satisfactory	12	100
9.	What is frequency of door-to-door surveillance?	Weekly	7	58.3
		Monthly	4	33.3
		Fortnightly	1	8.3
10.	Are you guided and supervised by?	MPHW + MTS + others	4	33.3
		MTS + others	2	16.6
		Others	2	16.6
	Malaria Inspector	1	8.3	

**Table 4. Species composition of mosquitoes based on larval survey**

	Command	Non-command
Larvae collected	5538	2505
Habitats (+)ve for mosquito breeding (%)	59.3	84.7
No of species emerged	9	8
Species composition (%)		
<i>An. annularis</i>	0.6	1.4
<i>An. barbirostris</i>	0.5	1.4
<i>An. culicifacies</i>	7.3	5.7
<i>An. fluviatilis</i>	0.1	0
<i>An. nigerrimus</i>	0.0	0.7
<i>An. pallidus</i>	0.2	0
<i>An. stephensi</i>	0.3	1.4
<i>An. subpictus</i>	12.3	37.9
<i>Cx. quinquefasciatus</i>	70.1	43.6
<i>Cx. vishnui</i>	8.6	7.8

**Table 5. Mosquito breeding in domestic water containers**

Parameters	Command	Non-command
Houses checked	1014	505
House positive	209	117
Containers checked	1063	589
Containers positive	214	119
House index	20.6	23.16
Container index	20.13	20.2
Breteau index	18.34	16.46

command and area is shown in Fig. 4. The density of *An. culicifacies* was highest in March-August in 2013. Out of 866 *An. culicifacies* blood meal samples, 17 mosquito blood meal samples were from human (1.96%; 17/866). The parity rate in *An. culicifacies* was highest in winter season (October-November) and lowest in summer (June).

In the command area of Phase-II district, State Health Department should undertake regular vector

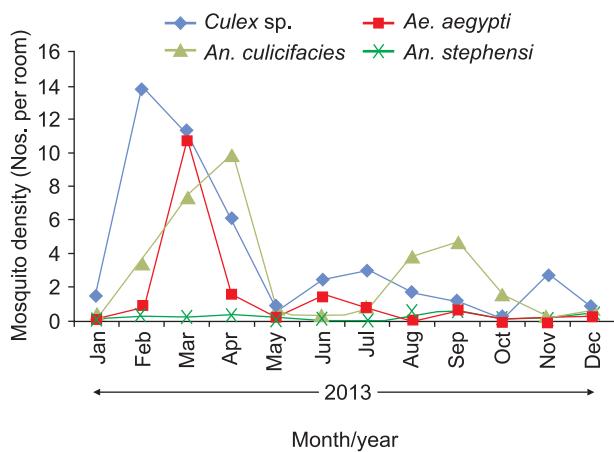


Fig. 4: Seasonal prevalence of mosquitoes in command area.

surveillance to contain the dengue and malaria incidences. The species-specific mosquito breeding sites must be targeted for effective intervention to control population of *Ae. aegypti* and *An. culicifacies*. Establishment of hatcheries of *Aphanius dispar* for scaling up use of larvivorous fish in the command area has been initiated. The study is in progress.

### Centre for Study of Complex Malaria in India

This project has been launched in November 2012 in Gujarat state. Initially, the Nadiad town has been included in the study to determine how different *P. falciparum* and *P. vivax* transmission ratios impact malaria burden. In Nadiad town, census of 1500 houses was done for obtaining general information and location of the houses by GIS. A clinic pilot epidemiological study was done to standardize the malaria diagnostic methods. Clinical and two rounds of cross-sectional studies were carried out in Nadiad town. Two Primary Health Centres of Kheda district and three private laboratories in Nadiad town were also included in the study, as recommended by the CSCMi RAC members.

### Pilot epidemiological study

In clinic, 300 patients were enrolled and their prior consent was obtained before inclusion in the study. Of which 298 cases were analysed, 19 (6%) were positive by microscopy, 17 (5.7%) and 18 (6%) were positive with RDT device "A" and "B", respectively and 18 (6%) were found positive with PCR (Tables 6 and 7). Since RDT device "A" was better in handling in the field and diagnostic

**Table 6. Malaria cases diagnosed by different methods**

BSE	Enrolled	Microscopy (all Pv)	RDT		PCR
			A	B	
1792	300	19 (6.3)	17 (5.7)	18 (6)	18 (6)

Figures in parentheses indicate percentage.

**Table 7. Enrolment and clinical presentation in the pilot study**

	Age (yr)			Gender (%)		Mean temp. (°F)	Weight (kg)	Hb g/dl
	<1	2–14	>15	Male	Female			
	2	89	209	119 (39.6)	181 (66.3)	98.63 (95.8– 106.1)	40.85 (6– 95)	13 <7 <11 80

Hb: Average:11.98 g/dl (3–17.9 g/dl); Anemic: 27.6%.

accuracy was nearly at par with both the devices and device "A" was chosen for the study.

### Clinical study

In Clinic study, 3182 patients were examined and 155 could be enrolled in the study, with 17 positive microscopically, 16 positive with RDT (in 2 cases RDT was not available) and PCR of these enrolled patients showed 16 positive cases (Table 8a). The average haemoglobin level in these patients was 12.43 g/dl. The 29 persons were anemic with haemoglobin 27 for <11 g/dl (Table 8b). All the microscopically positive cases were followed up according to the protocol up to 42 days. But only 5 cases could be followed up to 42 days and remaining could not be completed due to various reasons (Table 9).

**Table 8a. Cases enrolled in a clinical study diagnosed by different methods**

BSE	Enrolled	Microscopy		RDT (A)		PCR	
		Pv	Pf	Pv	Pf	Pv	Pf
3182	155	13	4	12	4	13	3

Two tests could not be done due to non-availability of RDTs.

**Table 8b. Enrolment and clinical presentation in a pilot study**

	Age (yr)			Gender (%)		Mean temp. (°F)	Weight (kg)	Hb g/dl
	<1	2-14	>15	Male	Female			
0	36	79	72	43	43	99.52 (96.1–104.6)	44.84 (11.8–83)	2 27

Hb: Average: 12.43 g/dl (5.5–18.8 g/dl); Anemic; 26.9%.

**Table 9. Follow up of malaria cases in a clinical study**

Cases	Enrolled	Followed up on Day						
		0	2	7	14	21	28	42
Pv	13	13	9	8	6	6	6	5
Pf	4	4	4	2	1	Na	Na	Na
Total	18	18	13	10	7	6	6	5
NA			5	8	11	12	12	13

Na = Not available; NA = Patients not available on the day of follow up.

### Cross-sectional study

In first cross-sectional study (May-June) in Nadiad, 307 patients were enrolled, of which 1 (0.3%) was positive microscopically and RDT device but by PCR two cases were found malaria positive. During second round of cross-sectional study from August

to December, 271 subjects were enrolled and examined and 31 were found positive microscopically and with RDT device (in 6 cases kit was not available) 16, 18 and 2 Pv, Pf and Mix, respectively while with PCR 19 Pv, 10 Pf and Mix 2 were diagnosed (Table 10). Clinical presentation is shown in Table 11. The proportion of anemic subjects was >30% in both the surveys. As per the protocol, 36 cases were followed on Day 7, *P. falciparum* and *P. vivax* on Day 7 and 14. In none of the subjects reappearance of parasitaemia was recorded (Table 12). The study indicates no significant difference between microscopy and PCR, seasonal transmission, infections possibly relapsing vivax in summer, possibly once in past 12 months and large non-cooperation in follow up. The study is in progress.

**Table 10. Cases enrolled and diagnosed in a cross-sectional study**

Period	Enrolled	BSE	Microscopy			RDT (A)			PCR		
			Pv	Pf	Mix	Pv	Pf	Mix	Pv	Pf	Mix
May–	307	307	1	0	0	1	0	0	2	0	0
Jun			(0.3)			(0.3)			(0.65)		
Aug–	271	271	18	11	2	16	18	2	19	10	2
Dec											

Figures in parentheses indicate percentage; 6 cases (Pv) could not be tested with RDT due to non-availability of kits.

**Table 11. Enrolment and clinical presentation of subjects in a cross-sectional study**

Period	Age (yr)			Gender (%)		Mean temp. (°F)	Weight (kg)	Hb g/dl
	<1	2-14	>15	Male	Female			
May–	0	49	258	138	169	97.78	51.94	9
Jun				(44.9)	(55.04)	(93.5)	(10–105.8)	93
Aug–	1	48	222	146	125	98.25	48.93	7
Dec				(53.7)	(46.3)	(94.3–104)	(8–110)	79

May-June— Hb: Average: 11.67 (6.4–16.5 g/dl); Anemic 33.2% and August-December— Hb: Average 11.59 g/dl (2.6–17.2); Anemic 31.73%; Asymptomatic cases— Nil.

**Table 12. Malaria cases confirmed in clinical study and followed**

Study	Cases followed		Followed on Day			
			0	2	7	14
Cross-sectional	Pv	16	16	–	13	15
	Pf	18	18	–	18	–
	Mix	2	2		1	1
Total			36	36	22	16

## A randomised village-scale evaluation to compare the efficacy of pirimiphos-methyl CS (300 g a.i./l) with pirimiphos-methyl EC (500 g a.i./l) by indoor residual spraying for malaria vector control in Gujarat state, India

This trial was undertaken in the villages of Kheda, Vadodara and Panchmahals districts in Gujarat state during September 2012 to August 2013. The objectives were to determine and compare the persistence, efficacy and impact on vectorial potential of pirimiphos-methyl CS (300 g a.i./l) with pirimiphos-methyl EC (500 g a.i./l) @ 1 g/m<sup>2</sup> dose. After three months of baseline study, a single round of spraying of pirimiphos-methyl 300 CS and pirimiphos-methyl 500 EC was undertaken in the last week of December 2012 in 10 villages, i.e. five villages in each arm. The entomological monitoring and cone bioassays were carried out in three villages of each arm.

Indoor spraying was done using hand-operated compression sprayers fitted with new flat fan nozzle with 80° discharges (usually referred to as 8002) according to WHO guidelines. The spray coverage of test insecticide in the villages of both the arms is given in Table 13. Chemical analysis of samples showed that mean value of p-methyl 300 CS content in paper samples drawn from different surfaces varied marginally (Table 14).

All the female mosquitoes 57,956 *Anopheles* spp. and 2000 *Culex* spp. were caught by different methods from the study villages in the baseline period (Table 15). Malaria vector, *An. culicifacies* was 7.27% among all the anopheline species. *Anopheles stephensi* and *An. fluviatilis* were found in low numbers.

**Table 14. Chemical analysis of samples of surfaces sprayed with Pirimiphos-methyl CS and EC**

Surface	No. of samples	Mean	± SD	Range
CS-300 (mg/m <sup>2</sup> )				
Lime coated cement	9	961.93	243.04	625.1–1343.9
Wood	9	880.68	915.86	0–1920.1
Mud plaster	9	988.36	637.52	418.6–2120.7
EC-500 (mg/m <sup>2</sup> )				
Lime coated cement	9	881.38	322.32	606.6–1487.2
Wood	9	864.06	571.27	177.5–1547.2
Mud plaster	9	1586.76	1134.79	402.7–1617.5

**Table 15. Mosquito species collected during the trial period**

Species	No. of mosquitoes collected	Percentage
<i>An. culicifacies</i>	4356	7.27
<i>An. stephensi</i>	299	0.50
<i>An. annularis</i>	1049	1.75
<i>An. fluviatilis</i>	754	1.26
<i>An. subpictus</i>	51399	85.73
Other anophelines	99	0.17
Sub-total	57956	
<i>Culex</i> spp.	2000	3.34
Total	59956	

\* *An. nigerrimus*, *An. barbirostris* and *An. tessellatus*.

### Persistence of insecticidal activity on sprayed surfaces

An assessment of the insecticidal activity was made by cone bioassay tests on common surfaces, i.e. mud-plastered walls, unpainted wood surfaces and lime coated cement surfaces with Pirimiphos-methyl 300 CS and 500 EC formulation until eight months post-spraying. The persistence of both the formulations sprayed on different surfaces

**Table 13. Spray coverage of human dwellings and rooms in both the study arms**

Insecticide sprayed	Village /Hamlet	Human dwellings			Rooms		
		Targeted	Sprayed	Covered %	Targeted	sprayed	Covered %
EC-500	Galteshwar	77	74	96.1	192	185	96.4
	Ramnagar	67	65	97	167	162	97
	Bhatipura	57	54	94.7	142	135	95.1
	Umba	69	68	98.6	175	170	97.1
	Bhadrasa (Juna muada)	48	46	95.8	148	144	97.3
	Total	318	307	96.5	824	796	96.6
CS-300	Varasada (Chandrikanager)	118	116	98.3	295	290	98.3
	Mahisa (Jevana muada)	78	76	97.4	195	190	97.4
	Vamali	69	67	97.1	173	167	96.5
	Balol (Indranagari)	72	68	94.4	182	173	95.1
	Gothada (Mahisagar)	115	106	92.2	373	344	92.2
	Total	452	433	95.8	1218	1164	95.6

## NADIAD (GUJARAT)

**Table 16.** Persistence of both the formulations sprayed on different surfaces

Surface	Effect	Pirimiphos-methyl 300 CS	Pirimiphos-methyl 500 EC
Mud wall	Knockdown*	1 month	1 month 24 h
	Mortality**	4 months	2 months
Wooden surface	Knockdown	2 months	1 month 24 h
	Mortality	6 months	2 months
Lime coated cement	Knockdown	1 month	7 days 24 h
	Mortality	4 months	1 month

\* ≥90%; \*\* ≤80%.

is summarized in Table 16. Considering a cut-off where the residual effect drops below 80%, it is observed that CS formulation was more effective in knockdown and 24 h mortality as compared to EC formulation. On each surface, the effect of CS formulation persisted longer than the EC formulation.

### Impact on vector population

After the spraying in last week of December, there was decline in geometric mean densities of *An. culicifacies* in the villages of both arms sprayed

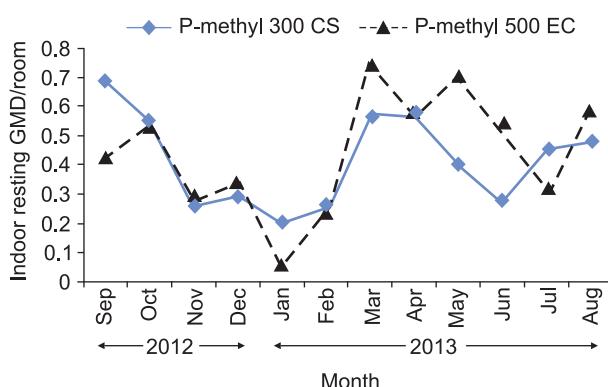


Fig. 5: Impact on indoor resting density of *An. culicifacies*.

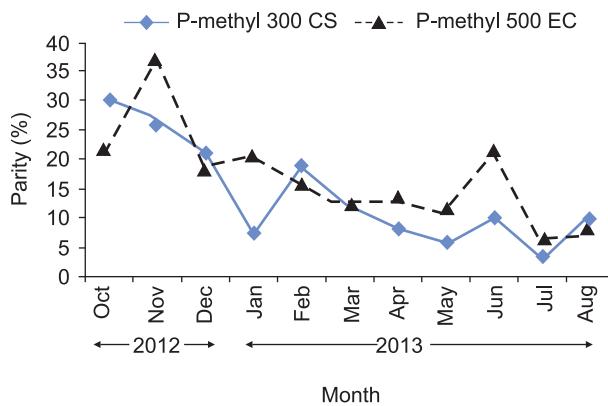


Fig. 6: Impact of p-methyl CS and EC on survivorship of *An. culicifacies*.

**Table 17.** Entomological inoculation rate (EIR) in trial arms

Samples assayed	Assay results				Sporozoite rate (%)	Biting rate	EIR	
	Pf 210	Pv 210	Pv 247	Total				
P-methyl CS	620	2	0	0	2	0.322	1.80	0.579
P-methyl EC	793	2	0	0	2	0.252	1.71	0.430

up to February 2013. The density was lower in villages of CS arm as compared to EC till the end of the trial (Fig. 5). The mean parous rate of *An. culicifacies* in both comparison arms of villages during the baseline period ranged from 23.26 to 37.73% and the rates didn't differ significantly post-spray (Fig. 6). Data on entomological inoculation rate (EIR) using mosquitoes collected off human baits, light trap, pyrethrum spray collection and exit trap are given in Table 17.

Considering the length of the main malaria transmission period in Gujarat, persistence of the insecticidal action on most common sprayed surfaces and effects on the elements of vectorial capacity, two rounds of pirimiphos-methyl 300 CS @ 1g/m<sup>2</sup> dose may be required for effective control of malaria in this area.

### Phase-II baseline household survey in World Bank project in districts of Maharashtra, Gujarat, West Bengal and Karnataka

In Gujarat, this study was undertaken in collaboration with the State Health Department in September 2013 with the objective to find out the current status of intervention coverage and challenges faced by the malaria control programme at community level. For a cross-sectional, questionnaire-based household survey, 80 villages were randomly selected in 13 PHCs of seven



Training of MSWs for household survey.



Household survey in a village.

districts (Godhra, Dahod, Valsad, Surat, Surendranagar, Patan, Kutchch [Bhuj] districts). A set of questionnaires for village information schedule, households schedule, fever today, fever in two weeks, death schedule with 12 months, and information on ASHA, interview tracking, house listing and mapping were used to collect the information related to various indicators. Before commencement of the actual household survey, a 3-day training was organized for 45 students (Master of Social Works) as interviewers/supervisor/NVBDPC Consultant and MTS at Gujarat Vidhyapeeth, Ahmedabad. The surveys in 48 villages of 6 PHCs in four districts (Godhra, Dahod, Surat and Valsad) have been completed. Survey is still in progress.

### **Large-scale (Phase-III) evaluation of efficacy, fabric integrity and community acceptability of PermaNet® 3.0 long-lasting insecticidal nets compared with PermaNet® 2.0 in India**

Study was initiated in the villages of Gujarat state in October 2013 with the objectives: to determine and compare the insecticidal activity and fabric integrity of PermaNet 3.0 LNs with PermaNet 2.0 over three years of use by households under the field conditions, to assess the washing mode and washing habits of LNs by the householders; and to assess the community acceptability of LNs under the field conditions.

The roofing of PermaNet 3.0 utilizes deltamethrin and a synergist, piperonyl butoxide (PBO) incorporated into monofilament polyethylene yarn of 100 denier (Warp-knitted fabric, with weight of  $40 \pm 15\% \text{ g/m}^2$ ) at the target dosages of 4.0 g a.i./kg and 25 g a.i./kg of netting material, respectively. The

side panels of PermaNet 3.0 are made of multi-filament polyester fibres, treated with deltamethrin in a resin coating ( $75 \pm 5\% \text{ denier}$ ). The side netting has two parts: a strengthened lower part, so called border (70 cm) by using  $75 \pm 5\% \text{ denier}$  (weight  $40 \pm 10\% \text{ g/m}^2$ ) and a side panel made of  $75 \pm 5\% \text{ denier}$  (weight  $30 \pm 10\% \text{ g/m}^2$ ). The target dosage of deltamethrin on the side panels is 115 mg a.i./m<sup>2</sup> of the border and 85 mg a.i./m<sup>2</sup> of the remaining of the side panels. PermaNet 2.0 net is made of knitted poly-filament polyester fibres and is treated with deltamethrin to a target concentration of 55 mg/m<sup>2</sup>. About 3000 nets have been received and samples of 30 nets of each type have been sent to the WHO authorized laboratory for testing the target dose.

Three villages Rali, Old Rali and Kanawal in Anand district have been included in the study. The census of households (1500) in these villages has been completed. The species *An. culicifacies* and *An. stephensi* (F1) were found susceptible to deltamethrin (0.05%). Bio-efficacy, chemical assay and household surveys on each type of net have been planned according to the protocol. Community counselling, coding and preparation of master lists for distribution of nets are in progress.

### **Support to the State Vector Borne Disease Control Programme**

#### **Assessment of malaria situation and monitoring of anti-malaria activities in Ahmedabad Municipal Corporation (AMC)**

On the request of Commissioner of AMC, a team of the National Institute of Malaria Research Field Unit, Nadiad and Regional Office of Health & Family Welfare, Ahmedabad assessed the ongoing vector borne disease control activities in AMC. The team visited two Urban Health Centres one each in central and new west zone from 30–31 May 2013. Comparison of routine surveillance data for January to May 2012 and 2013 showed marginal increase in May 2013 as compared to previous year 2012. Up to May 2013, >95% malaria was due to *P. vivax* (1982/2059) infection. Nearly, 70% malaria cases were reported from the south, new west and eastern zones. Dengue cases have increased from 46 in May 2012 to 100 during corresponding period in 2013. The team visited slums/societies under 2 Urban Health Centres of 2 wards in 2 zones of AMC. Based on the current



Larval survey in river front, Ahmedabad.

disease burden, preparedness of staff posted at UHC and muster, field visits, surveys, the IRS issues were discussed with ADHO, Medical Officer and Jr. Assistant Entomologist. Larval surveys were carried out to find breeding potential of vector mosquitoes. In order to minimize malaria and dengue cases, a strict supervision by all the level of officers, arrangement of workers for active surveillance in slums, anti-larval activities and participation of community in IRS were urgently required. The observations and recommendations were submitted to the Commissionerate of AMC for necessary action.

#### **Independent assessment of the mosquito control measures in certain districts of Gujarat**

The Joint Director, VBDPC (Gujarat), Gandhinagar has requested the National Institute of Malaria Research Field Unit, Nadiad and Regional Office of Health & Family Welfare, Ahmedabad to asses the ongoing Anti-malaria activities in Godhra, Dahod and Valsad, Rajkot, Surendranagar, Banaskantha and Kutchch districts and Vadodara Municipal Corporation (VMC). In October 2013, a team of NIMR, Nadiad has visited Vadodara Municipal Corporation, Godhra and Dahod districts. In VMC, three Urban Health Centres (UHCs), societies/slums/villages and construction sites and in Godhra and Dahod districts three PHCs, one CHC and two villages in each PHC were visited. The preparedness for mosquito control in monsoon, work schedule, advance programme of supervisors and Medical Officers and anti-mosquito measures were discussed in the presence of Biologists/District Malaria Officers. In general, the State has reported

three folds increase in dengue in 2013 (1707 cases) as compared to 2012 (445 cases) up to the month of September. Malaria cases in the state have declined to 44,123 in September 2013 as compared to 58,372 cases in September 2012 but it has increased in certain PHCs in Dahod and Godhra districts. The observations, present status of staff, mosquito breeding sites and remedial measures have been reported to the Joint Director, VBDPC, Gandhinagar for necessary action.

#### **Malaria Clinic**

Malaria diagnostic support was provided to the Civil Hospital and the data so generated have been used for sentinel monitoring of malaria situation in the district. In 2013, 7031 febrile patients have been screened for malaria, of which 131 had malaria infection ( $Pv = 117$ ;  $Pf = 13$  and Mix = 1). The slide positivity rate was 1.86%. All the confirmed malaria patients were provided radical treatment by Medical Officers of Civil Hospital, Nadiad. In general, there is decline in malaria cases but proportion of *P. falciparum* has increased (Fig. 7). The age-wise distribution of cases indicates highest

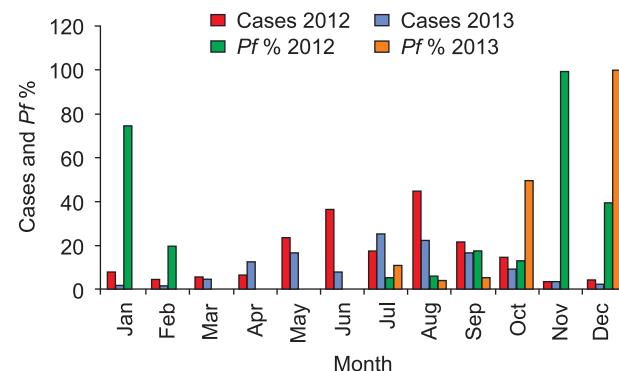


Fig. 7: Malaria cases reported at NIMR Clinic, Nadiad.

**Table 18. Age and sex-wise distribution of malaria cases reported at NIMR Clinic**

Age group (yr)	Male			Female			Total		
	BSE	Pv	Pf	BSE	Pv	Pf	BSE	Pv	Pf
< 1	51	0	0	23	1	0	74	1	0
1 to 4	322	1	0	234	1	1	556	2	1
5 to 8	300	4	0	237	2	0	537	6	0
9 to 14	333	8	0	320	7	0	653	15	0
> 15	2734	66	7	2477	27	6	5211	93	13
Total	3740	79	7	3291	38	7	7031	117	14*

\*One mixed case ( $Pv + Pf$ ); BSE – Blood slide examined; Pv – vivax; Pf – *P. falciparum*.

cases in adult age group in this area (Table 18).

### **Important meetings attended**

#### **Dr HC Srivastava**

1. Participated in SACTO programme organized by the State Health Department at Gandhinagar and delivered lecture on Importance of community participation in mass drug administration on 5 January 2013.
- 2 Attended the meeting for Inter-sectoral coordination in Vector Borne Disease Control, organized by the Commissionerate of Health at Gandhinagar on 24 April 2013.
3. Participated in the meeting on “Inter-sectoral collaboration” for Prevention and control of mosquito borne diseases at Ahmedabad Mahanagar Seva Sadan, Ahmedabad on 28 May 2013.
4. Attended review meeting to discuss issues related with High Court PIL in 2011 on 3 June 2013. The meeting was called by the Commissionerate of Health, at Gandhinagar.
5. Delivered a lecture on “Conquering malaria-with special reference to industrial estates/township” in short-term training programme on “Updates in public health for factory medical officers” of Gandhinagar, Thaltez and Ahmedabad at the Indian Institute of Public Health on 24 June 2013.
6. Attended pre-mission field visit review meeting for “World Bank Mission in connection with Malaria Control Project” at the Commissionerate of Health, Gandhinagar on 9 July 2013.
7. Attended 24th NVBDCP sub-committee meeting on 30 July 2013 to review the VBDC activities in various districts of Gujarat and provided inputs on IRS and alternative methods for vector control. The meeting was called by the Additional Director (Health), Gandhinagar.
8. Attended review meeting on Vector Borne Diseases Control in Gujarat and presented the

results of Assessment of mosquito control activities in Ahmedabad Municipal Corporation, Ahmedabad, at NVBDCP Regional Office for Health & Family Welfare Ahmedabad on 12 September 2013.

9. Dr HC Srivastava and Mr VS Malaviya felicitated “World Malaria Day” at the District Malaria Office, District Panchayat, Kheda on 25 April 2013, and delivered a speech on “Invest in future in defeating malaria”.

### **Trainings/Workshops/Seminars/Conferences/Symposia organized/participated**

1. On the request of Commissionerate of Health, Gandhinagar, Re-orientation training courses in malaria microscopy for laboratory technicians of Primary Health Centres were organized at this Field Unit, (one batch 18–25 person each) in March, July, August, September and October 2013.
2. Training courses on Malaria surveillance and patient care were organized for 3 batches of first year students (5 in each batch) of Nursing College, Civil Hospital Nadiad from January–February 2013. The training included active surveillance, blood smear preparation, treatment and patient care. A live demonstration on mosquito life cycle was also organized.
3. A 3-day training for 45 students (Master of Social Works) as interviewers/ supervisor and 7 NVBDCP consultants was organized at Gujarat Vidhyapeeth, Ahmedabad from 10–12 September 2013 under “Phase-II Baseline household surveys project” supported by the NVBDCP (World Bank) to carry out household surveys in 80 villages of Gujarat.
4. Dr HC Srivastava participated as faculty in a workshop organized by the Joint Director, Office, Dadra & Nagar Haveli on ‘Prevention and control of malaria and other vector borne diseases’ from 14–15 May 2013 for supervisor and multi-purpose health workers at Dadra and Nagar Haveli.



## 3.7

### Panaji (Goa)

#### Proteogenomic analysis of *Anopheles stephensi* Liston, a principal vector of malaria in India

*Anopheles stephensi* is a major vector of malaria in urban areas of India. Knowledge on proteogenomics of this vector is limited. Hence, we have initiated comprehensive studies on proteome of this species and carried out annotation of genes responsible for these proteins with particular focus on those involved in *Plasmodium* infection. Comprehensive proteomic analysis of *An. stephensi* was carried out to characterize proteins expressed in using high resolution LTQ-OrbitrapVelos mass spectrometer.

We searched our mass spectrometry derived peptide data against a computationally predicted *An. stephensi* protein database and so far, we have identified 8000 proteins. Out of which, 33 proteins identified were involved in various known biological process. These 33 proteins were classified into 15 categories based on Gene Ontology annotations of biological processes as per VectorBaseDB (Fig. 1).

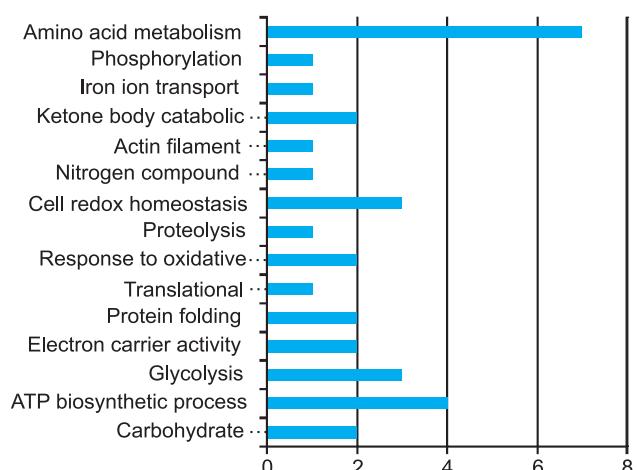


Fig. 1: Classification of the identified mid gut proteins of female *An. stephensi* in biological process based on the gene ontology annotations of biological process as per VectorBaseDB.

Eight proteins were identified from the mid gut of female *An. stephensi*, which are involved in vector parasite interactions. These agonistic molecules are associated with the *Plasmodium* development in mosquito *An. gambiae* and their role has been confirmed using knockdown studies in *An. gambiae*. They are attractive candidates for elucidating their role in parasite development, invasion and survival in *An. stephensi*. Further data analysis is in progress.

#### Characterization of dengue/DHF, chikungunya and yellow fever vector *Aedes aegypti* Linn. proteome

*Aedes aegypti* is a major vector of dengue, west nile virus and yellow fever in many parts of the world. The genome of *Ae. aegypti* has been described. However, proteome is not yet characterized. Hence, we have taken up this study to elucidate proteins with emphasis on those functionally involved in disease infection in this vector and transmission process.

Proteins were extracted from *Ae. aegypti* by homogenizing the tissue in 0.5% SDS using ultrasonication. The extracted proteins were quantified by Lowry method using Bio-Rad DC Protein assay. In-gel trypsin digestion of proteins obtained was carried out and trypsin digested peptides were further analyzed on high resolution LTQ-OrbitrapVelos mass spectrometer. The MS/MS data were searched against a protein database comprised of known and predicted proteins reported from *Ae. aegypti* using software.

So far we have identified 944 and 1912 proteins from the salivary gland and mid gut of *Ae. aegypti* using in-gel digestion approach, respectively, 628 proteins were present in both the salivary gland and mid gut. The unique proteins identified in salivary gland and mid gut were 316 and 1284,

respectively. These proteins will be further characterized based on their biological functions. Further data analysis is in progress.

### Epidemiology and malaria evolution in south Asia

Both parasite and vector are evolving rapidly in south Asia and becoming a global concern. Therefore, it is important to study prevalence of vectors and their competence for *Plasmodium* infection from time-to-time. Three geographical areas in Goa were selected for mosquito collection using 233 CDC traps. A total of 3357 mosquitoes were identified based on standard morphological keys. Identification of anophelines was further confirmed by PCR amplification of ITS region using specific primers. Mosquitoes were assayed for blood meal and also *P. falciparum* and *P. vivax* infections using Nested PCR after separating their heads and abdomen.

A total of 321 anopheline specimens comprising of five species were caught in three malaria high risk towns along the coast of Goa. Among these were *An. stephensi*, *An. subpictus*, *An. barbirostris*, *An. annularis* and *An. jamesii*. *Anopheles stephensi* and *An. subpictus* were found to be positive for human blood indicating their anthropophilic nature. *Anopheles subpictus* which was hitherto considered as non-vector of malaria in this region showed infection (mixed infection for *P. falciparum* and *P. vivax*) in comparison to a well-known vector *An. stephensi* infection rate as revealed by Nested PCR.

In Goa, construction sites and overhead tanks were primarily targeted for *An. stephensi* control but the present study is forcing us to revisit the earlier perspective of *An. stephensi* to be the sole transmitter of malaria in urban Goa. Therefore, vector control focus needs to be enlarged covering this and other species which may be involved in transmitting malaria in this area.

### Larvicidal activity of leaf extracts of plant GIC against *Anopheles stephensi* Liston, *Culex quinquefasciatus* Say and *Aedes aegypti* Linn.

We evaluated larvicidal and pupicidal activity of methanol and chloroform extracts of dried leaves of plant, code named GIC against urban malaria vector *An. stephensi* Liston, filariasis vector *Cx. quinquefasciatus* Say, dengue and chikungunya vector *Ae. aegypti* Linn. Methanol and chloroform leaf extracts showed both larvicidal and pupicidal

effect after 24 and 48 h of exposure. Further, LC<sub>50</sub> and LC<sub>90</sub> values were calculated for each time interval. Methanol and chloroform leaf extracts showed different larvicidal activities against these three species.

Methanolic extract of leaf showed highest larval mortality against *An. stephensi* (100%) with LC<sub>50</sub> of 11 ppm and LC<sub>90</sub> of 28 ppm, followed by *Ae. aegypti* (82%) with LC<sub>50</sub> = 45 ppm, LC<sub>90</sub> = 112 ppm and *Cx. quinquefasciatus* (47%) with LC<sub>50</sub> = 65 ppm and LC<sub>90</sub> = 177 ppm, respectively at 24 h. Similarly, methanolic extract showed highest pupicidal activity against *An. stephensi* with LC<sub>50</sub> = 0.675 ppm and LC<sub>90</sub> = 1.41 ppm, followed by *Cx. quinquefasciatus* with LC<sub>50</sub> = 20.19 ppm, LC<sub>90</sub> = 115.02 ppm and *Ae. aegypti* with LC<sub>50</sub> = 24.69 ppm, LC<sub>90</sub> = 185.76 ppm at 24 h. Methanolic extract was found to be more effective than the chloroform against all the tested mosquito species. However, further work is required for identification and isolation of bioactive components present in leaf in order to develop eco-friendly larvicide of plant origin. Further work is in progress.

### Quality assessment of LLINs procured by NVBDCP-DHS by periodical bioassays

Long-lasting insecticide nets were introduced by the National Vector Borne Disease Control Programme in slum populations and among migrant construction workers. It was envisaged that duration of effectiveness of the target vectors of major vector borne diseases should be concurrently assessed. Cone bioassays were carried out periodically (every quarter) using laboratory populations of *An. stephensi*, *Cx. quinquefasciatus* and *Ae. aegypti* as per the WHO standards to assess the quality of LLINs procured by NVBDCP-DHS, Goa.

After 24 months of usage and seven washes, 73.5% mortality was recorded in the laboratory-bred *An. stephensi*, whereas 57.5 and 87.5% mortality was observed in the case of *Cx. quinquefasciatus* and *Ae. aegypti*, respectively (Fig. 2). Without washing, the percent mortalities were 95, 97.5 and 97.5% in *An. stephensi*, *Cx. quinquefasciatus* and *Ae. aegypti*, respectively. It may be noted that absolute mortality was observed with unwashed LLINs and those washed at quarterly interval with ordinary detergent until 21 months. It was observed that LLINs of a particular brand used in Goa were effective for up to two years.

## PANAJI (GOA)

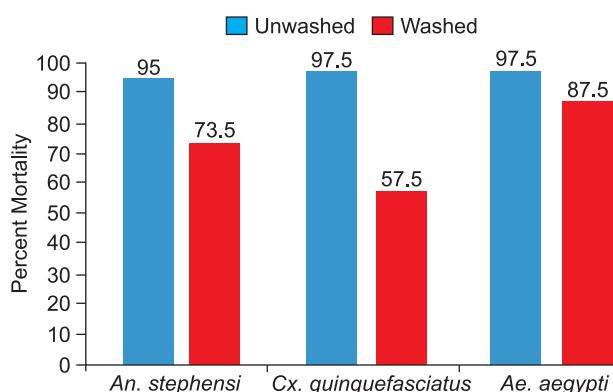


Fig. 2: Effect of 7 washes (in 24 months) on mortality of adult *An. stephensi*, *Cx. quinquefasciatus* and *Ae. aegypti* exposed to LLIN using cone bioassays.

### Insecticide susceptibility of *Anopheles stephensi*, *Aedes aegypti*, *Aedes albopictus* and *Culex quinquefasciatus* in Goa

Information on insecticide susceptibility of vectors is of great practical importance for NVBDCP, Goa and needs to be generated and updated from time-to-time. Therefore, we studied status of insecticide resistance in *An. stephensi* against which IRS is selectively carried out in slums albeit sparingly and LLINs impregnated with deltamethrin are being used in high risk population in urban slums in Goa. Laboratory-bred fully-fed vector females were exposed ( $n = 25/\text{replicate}$ ) in four replicates/insecticide, while two replicates of control females ( $n = 25$ ) were also concurrently maintained. Females were exposed for 1 h to standard WHO papers of 4% DDT, 5% malathion and 0.05% deltamethrin.

The mortality observed in *An. stephensi* adults to these insecticides was 53.2, 73 and 92%, respectively. On the other hand, the mortality

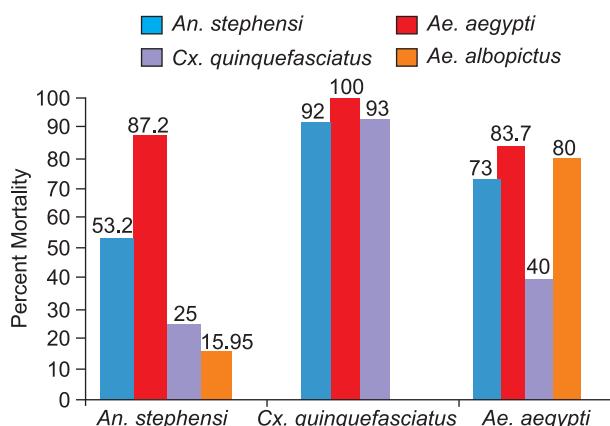


Fig. 3: Results of bioassays showing varying susceptibility of important vectors to insecticides in Goa.

observed in *Ae. aegypti* adults was 87.2% against DDT, 100% against deltamethrin and 83.7% against malathion. The mortality observed in *Ae. albopictus* adults was 15.95% against DDT and 80% against malathion. The mortality observed in *Cx. quinquefasciatus* adults was 25% against DDT, 93% to deltamethrin and 40% against malathion (Fig. 3).

There is varying resistance to DDT, malathion and deltamethrin in four vector species tested. The least resistance was found against deltamethrin in first three species, however, tolerance is growing in *An. stephensi* and *Cx. quinquefasciatus*. However, complete susceptibility to deltamethrin was found in *Ae. aegypti*.

### Vector surveillance in malaria high risk areas of Goa

Periodic vector surveillance was carried out in different malaria high risk areas by NIMR field teams especially in malaria high risk areas. The surveillance reports are provided to the Health Officers of the respective PHCs on day-to-day basis for antilarval treatment with a copy to the SPO for follow up. Antilarval treatment is also being rendered promptly on the spot by PHC spraymen who accompany the NIMR vector surveillance team followed by antilarval measures initiated by the Health Officers of PHC/UHC in the entire survey area based on the reports submitted by NIMR. High risk PHCs/UHCs covered in Goa for vector surveillance and monitoring were Candolim, Mapusa, Margao, Panaji, Ponda and Vasco.

The results of the vector surveillance carried out in five malaria high risk PHCs/UHCs areas of Goa, viz. Candolim, Mapusa, Margao, Panaji and Vasco from April 2013 to March 2014 are summarized in Table 1. Altogether, 215 (0.81%) breeding habitats out of 26,665 surveyed were positive for mosquito immatures which included 120 (0.5%) *Anopheles* spp, 76 (0.29%) *Culex* sp and 19 (0.07%) *Aedes* species. The species emerged from these samples were *An. stephensi*, *An. subpictus*, *An. vagus*, *An. jamesi*, *Cx. quinquefasciatus*, *Cx. vishnui*, *Cx. tritaeniorhynchus*, *Ae. aegypti*, *Ae. albopictus* and *Ae. vittatus*. The surveillance results are summarized in Table 1. Breeding site-wise results are given in Table 2. Further data analysis is in progress on seasonal population trends of vectors of malaria and other vector borne diseases to check the correlation between vector prevalence and disease incidence.

**Table 1.** Results of surveillance in high risk PHCs during the period between April 2013 and March 2014

PHC/ UHC/ CHC	No. of visits	Total breeding sites checked	Number and percent positive for							
			Total		Anophelines		Culicines		Aedines	
			Nos.	%	Nos.	%	Nos.	%	Nos.	%
Candolim	34	5262	39	0.74	22	0.4	15	0.29	2	0.04
Mapusa	33	5188	39	0.75	21	0.4	17	0.33	1	0.02
Margao	36	5980	33	0.55	20	0.3	13	0.22	0	0
Panaji	29	4344	14	0.32	9	0.2	4	0.09	1	0.02
Vasco	36	5891	92	1.56	48	0.8	27	0.46	15	0.25
Total	168	26665	217	0.81	120	0.5	76	0.29	19	0.07

**Table 2.** Percent positivity breeding site-wise in high risk PHCs during the period between April 2013 and March 2014

S.No.	Breeding sites	Total breeding sites checked	No. (+)ve	Percent (+)ve
1.	Barrel	1350	4	0.30
2.	Water collection	17141	62	0.36
3.	Bucket	11	0	0
4.	Cement tank	155	3	1.94
5.	Curing water	6818	113	1.66
6.	Drain	107	4	3.74
7.	Flower pot	26	2	7.69
8.	GTPL	306	2	0.65
9.	Lift Column	292	17	5.82
10.	Misc	71	1	1.41
11.	OHT PL	93	3	3.23
12.	Pits	11	0	0
13.	Pot	1	1	100.00
14.	Swimming pool	9	1	11.11
15.	Septic tank	29	2	6.90
16.	Sump	40	2	5.00
17.	Tin	175	0	0
18.	Well	1	0	0
19.	Tyre	29	0	0
	Total	26665	217	0.81

### Monitoring of vector populations and evaluation of impact of interventions with particular emphasis on dengue and chikungunya vectors in and around Goa airport and seaport

As per International Health Regulations to which India is a signatory, no vector should be found

breeding in ports of entry and ground crossings and in 400 m perimeter of these establishments. Goa has a seaport and an international airport in Vasco-da-Gama. Periodical vector surveillance was undertaken covering variety of breeding habitats to monitor vectors of malaria, filaria, JE, dengue and chikungunya. Areas surveyed were Dabolim airport (airport complex & new terminal construction complex), Airport periphery (labour huts, Mangor hill, New Vaddem and Dhaktale wado), Seaport (MPT port area) and seaport periphery (MPT colony/ Sada slum and private residential area).

The results of surveillance are summarized in Table 3. Altogether, 120 (0.89%) breeding habitats out of 13,470 surveyed were positive for mosquito immatures which included 12 (0.09%) *Anopheles* spp., 14 (0.10%) *Culex* sp and 94 (0.70%) *Aedes* species. The species emerged from these samples were *Ae. vittatus*, *Ae. aegypti*, *Ae. albopictus*, *Cx. quinquefasciatus* and *Cx. vishnui*. A total number of 790 houses/premises were checked during the survey period, of which 98 (House Index = 12.41) were found positive for *Aedes* breeding sites. The container and breteau indices were 0.89 and 11.90, respectively. The results of area-wise surveillance are summarized in Table 4. Results of surveillance breeding site-wise are given in Table 5.

Findings of the surveys in the port areas suggest that most of the breeding was restricted to barrels,

**Table 3.** Results of surveillance in port areas from April 2013 to March 2014

Area (No. of visits made)	Total breeding sites checked	Number and percent positive for							
		Total		Anophelines		Culicines		Aedines	
		Nos.	%	Nos.	%	Nos.	%	Nos.	%
Airport (10)	2372	22	0.93	9	0.38	5	0.21	8	0.34
Airport periphery (9)	3704	28	0.76	1	0.03	0	0.00	27	0.73
Seaport (8)	1874	23	1.23	0	0.00	7	0.37	16	0.85
Seaport periphery (14)	5520	47	0.85	2	0.04	2	0.04	43	0.78
Total (41)	13470	120	0.89	12	0.09	14	0.10	94	0.70

**Table 4. Results of surveillance in port areas from April 2013 to March 2014: Standard indicators of Aedes vector surveillance**

Area (No. of visits made)	No of premises checked	No. of premises (+)ve	Container index	House index	Breteau index
Airport (10)	51	18	0.93	35.29	15.69
Airport periphery (9)	296	23	0.76	7.77	9.12
Seaport(8)	70	16	1.23	22.86	22.86
Seaport periphery (14)	373	41	0.85	10.99	11.53
Total (41)	790	98	0.89	12.41	11.90

**Table 5. Breeding site-wise contribution to mosquito populations in port areas from April 2013 to March 2014**

S.no.	Breeding site	Total breeding sites checked	No. (+)ve	Percent (+) ve
1.	Barrel	1492	47	3.15
2.	Bottle	1122	0	0
3.	Bucket	1124	3	0.27
4.	Cement tank	40	0	0
5.	Coconut shell	2375	0	0
6.	Drain	119	8	6.72
7.	Flower pot	2529	2	0.08
8.	Fountain	10	0	0
9.	Grinding stone	17	3	17.65
10.	GTPL	193	4	2.07
11.	OHT PL	98	0	0
12.	Pot	176	5	2.84
13.	Scrap MC	198	4	2.02
14.	Sump	4	0	0
15.	Tin	1908	16	0.84
16.	Tyre	446	10	2.24
17.	Miscellaneous	1619	18	1.11
.	Total	13470	120	0.89.

**Table 6. Results of entomological surveys in Mumbai during investigations**

S.No.	Area	No. checked	Positive for Aedes (%)	Breeding sites surveyed
1.	E-ward, Madanpura	22	0	Water collections, ground tanks (plastic drums), tins, tarpaulins
2.	Kamathipura, S.P. Road	15	1 (6.7%)	Water collections, tarpaulins, ground tanks (plastic drums) and odd containers
3.	N-ward, Barvenagar, B-Block, Ghatkopar (W)	43	0	Ground tanks (plastic drums), tarpaulins and odd containers
4.	G/N Ward, Jagruti Industries, Mogul Lane, Mahim	8	0	Ground tanks (plastic drums) and odd containers
5.	G/N Ward Polychem Industries Corporation, Mogul Lane, Mahim	3	0	Ground tanks (plastic drums) and odd containers
6.	Municipal Vocational School, Mambala Tank Road, Matunga (W)	5	0	Ground tanks (plastic drums) and odd containers
	Total	96	1 (1.04%)	



Central team conversing with Dr Arun Bamne, Medical Officer of Health, Municipal Corporation of Greater Mumbai.



The team members visiting the field and inspecting a barrel for *Aedes* breeding in Mumbai.

### Salient observations regarding *Aedes* breeding potential

- Plastic drums were the main water storage containers in the visited areas which are supporting breeding.
- On top of huts and small shops, tarpaulin sheeting formed channels and depression due to the weight of stones, tyres, etc., ideal for *Aedes* breeding during monsoon and post-monsoon seasons.
- Coconut shells, tarpaulins, small/big earthen pots, plastic cups, tyres, bottles, thermocol, etc. were the main breeding sites outdoors.
- Flower pots, money plant containers, Fengshui containers were main breeding sites indoors.
- Abandoned constructions, Railway yards and Port areas have huge potential of breeding during the monsoon period.

### Entomological assessment of dengue situation in Mumbai

- In the month of September 2013, a total number of 892 potential *Aedes* breeding sites were enumerated/reported from all the 24 wards and E-ward being the highest with 197 breeding sites followed by 113 in K/E and 103 in M/E wards.
- Emergence from the above breeding sites showed *Ae. aegypti* and *Ae. vittatus* species. No *Ae. albopictus* were reported so far as per the records of MCGM.
- Till 19 October, a total of 1099 positive breeding sites of *Aedes* were recorded from ward E which was the highest number of breeding sites in 2013 as per the data provided by MCGM.

### Epidemiological assessment of dengue situation in Mumbai

- As per the data of MCGM, in 2013 till 20 October, total number of confirmed dengue cases reported were 698. There were 6 deaths due to dengue, 3 deaths each in the months of August and September. In the year 2012, the number of confirmed cases was 1008 and 5 deaths. Confirmed dengue cases and deaths from the year 2010 to 2013 are given in Table 7.
- Of total number of 698 cases, ward E recorded highest 79 (11.32%) cases followed by ward F/N 71(10.17%) and ward G/N 69 (9.89%), respectively.
- Month-wise trend of confirmed dengue cases and deaths from the year 2010 to 2013 are given in Fig. 4.
- Cases and deaths due to dengue show increasing trend from the year 2010 to 2013.
- The ELISA confirmatory test and dengue rapid test were carried out in 5 hospitals and 16 health posts.
- In 2013, 16,003 suspected samples were checked for IgM-Mac ELISA, of which 698 (4.36%) were found positive. Similarly, 24,352

Table 7. Confirmed dengue cases and deaths in Mumbai during the years 2010 to 2013

Year	Dengue cases	Deaths
2010	115	3
2011	416	3
2012	1008	5
2013 (Till 24 October)	698	6

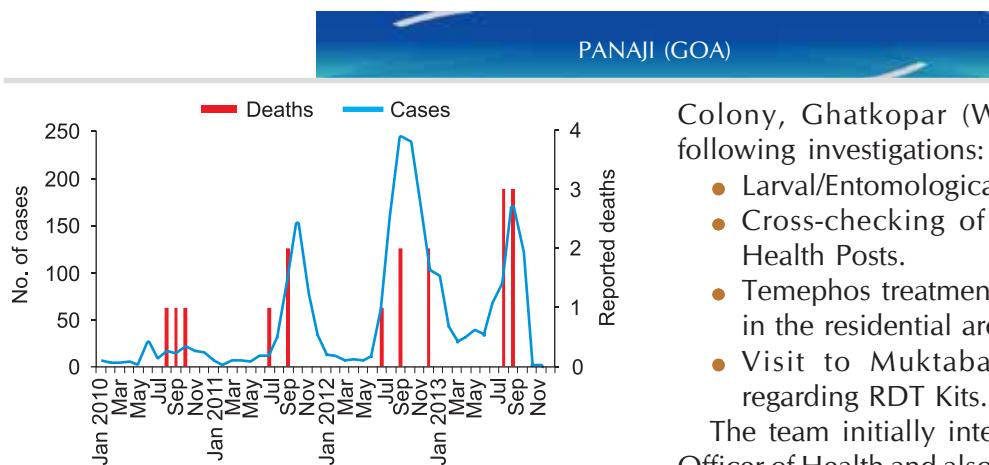


Fig. 4: Trend of dengue in Mumbai from January 2010 to November 2013.

**Table 8. Results of dengue diagnosis with ELISA (IgM) and RDK carried out in Mumbai during the dengue outbreak by MCGM**

Month	ELISA			RDK		
	No. of samples	Positive cases	Percent (+)ve	No. of samples	positive cases	Percent (+)ve
Jan 2013	1454	94	6.46	796	53	6.66
Feb	1211	41	3.39	511	31	6.07
Mar	1162	24	2.07	652	24	3.68
Apr	1400	29	2.07	745	27	3.62
May	1164	37	3.18	658	23	3.50
Jun	1371	32	2.33	791	130	16.43
Jul	2245	66	2.94	1581	101	6.39
Aug	3134	87	2.78	4272	389	9.11
Sep	1738	168	9.67	6649	1338	20.12
Oct	1124	120	10.68	7697	1257	16.33
Total	16003	698	4.36	24352	3373	13.85

suspected samples were checked by Rapid Diagnostic Test Kits, of which 3373 (13.85%) were found positive. Dengue positive cases reported in Mumbai as diagnosed by ELISA test and RDT Kits from January to October 2013 are shown in Table 8.

### Visit to Microbiology Department of KEM Hospital, Mumbai

Dr Kalpana Baruah and Dr Hemanth Kumar visited the Microbiology Department of KEM Hospital, Mumbai and discussed about the discrepancies found in the results obtained by RDT diagnostic kits for dengue in comparison with IgM ELISA.

### Visit to N-ward of MCGM

The team visited B-block Barve Nagar, MCGM

Colony, Ghatkopar (W) and carried out the following investigations:

- Larval/Entomological surveys.
- Cross-checking of dengue records at the Health Posts.
- Temephos treatment of the water containers in the residential areas.
- Visit to Muktabai Hospital laboratory regarding RDT Kits.

The team initially interacted with the Medical Officer of Health and also with Pest Control Officer of N-ward. The data of dengue cases were reviewed. In the month of September 2013, 87 suspected dengue cases were reported. Till date in October 2013, 78 suspected dengue cases were reported in N-ward. It was informed by MOH of N-ward that 21 rallies for IEC coverage were conducted in the local slum and non-slum areas. The MOH further informed, that there are 220 C.H.V. the visiting team covering 10 Health Posts in that ward.

Barve Nagar Health Post was then visited along with the Medical Officer. The team visited B-block Barve Nagar, MCGM Colony along with AMO of the Health Post and C.H.V. The team inspected 35 houses in which 43 containers were checked. No larvae was detected in these containers. All were found covered with either cloth or plastic sheet. The accompanying C.H.V. had treated all the containers in that area as a preventive measure. The work of C.H.V. was found satisfactory. The team interviewed the locals about their knowledge and practices regarding dengue and their responses were found satisfactory. The households also informed that abate treatment is carried out on weekly basis.

The AMO of the Health Post informed that during the month of September 2013, 11 suspected dengue cases were reported from that area. In October, 17 suspected dengue cases were reported till date. As per the Health Post records approximately 12,000 containers were recorded in that area. Laboratory in the Muktabai Hospital was visited and availability of RDT kits for dengue was checked.

Variety of breeding places such as coconut shells, barrels, small pots, flower pots and other odd articles were found in the field and most of them were dry. The residents conveyed that they were told about the continuing dengue problem and how to take preventive measures.

## Recommendations/Suggestions

### Vector Control

1. Antilarval measures such as source reduction, chemical larvicide and environmental management should be undertaken promptly for reducing the transmission risk.
2. Depressions in the tarpaulin on the roof tops should be removed. If necessary, executive orders may be issued banning of tyres on the roofs especially in shanties and shops in the slums.
3. Abandoned construction sites should be enumerated and treated in each ward in the monsoon and post-monsoon period.
4. Residents of slums and 'chawls' should be instructed to cover the water storage drums with cloth or plastic sheets to prevent breeding.
5. Cross-checking of activities should be undertaken by an external organization periodically.

### Surveillance

1. Active surveillance to be sustained in all the wards, in high risk populations especially in slums. There should be epidemic preparedness and rapid response for case management.
2. For early case reporting and management, number of sentinel surveillance sites with laboratory support should be increased and similarly referral services should be strengthened.
3. Private health sector (diagnostic labs) should be involved in sentinel surveillance.
4. For epidemiological assessment, dengue cases detected in the private sector should be included in MCGM statistics.

### Community & peoples participation

1. IEC and BCC campaigns are being undertaken by MCGM. However, there is scope for further strengthening these through electronic and print media such as TV, Cable network, radio, pamphlets, posters, banners, hoardings, etc. which should be displayed at key places such as local trains, buses, bus stops, post offices, public offices, railway workshops, seaports, schools, colleges, kiosks, chawls and slums.
2. Supporting intervention such as health

education, IEC and BCC should be intensified during the inter-epidemic period, monsoons and at the time of transmission.

3. Intersectoral collaboration/coordination should be increased to take preventive measures by the concerned organizations especially railways, railway workshops, ports, bus yards, bus workshops, depots, defence establishments and airport.

### Participation in important Meetings, Conferences, Workshops, etc.

#### Dr Ashwani Kumar

1. Attended the Science of malaria eradication and managing the end of malaria advanced courses in Barcelona and Sitges organized by ISGlobal of Barcelona University, Spain from 5 to 13 May 2013.
2. Attended Goa State task force meeting of NVBDCP, Directorate of Health Services, Goa held at Conference Hall, Secretariat, Porvorim under the Chairmanship of Principal Secretary (Health) on 15 May 2013.
3. Invited as Temporary Advisor to Regional Director, SEARO for Inter-country training on Ship Sanitation Inspection and issuance of Ship Sanitation Certificate organized by WHO-SEARO Office, New Delhi at Kochi from 24–26 June 2013.
4. Invited as Temporary Advisor for Regional meeting on International Health Regulations (2005) : Core Capacities at the Point of Entry, organized by WHO-SEARO Office, New Delhi at Kochi from 27–28 June 2013.
5. Attended 3rd meeting of Evidence Review Group (MPAC) in WHO HQs at Geneva, Switzerland from 8–9 July 2013.
6. Invited to be a Member of Vector Control Advisory Group by the Department of Neglected Tropical Diseases, WHO, Geneva, Switzerland and attended its first meeting from 10–12 July 2013.
7. Attended third Annual Workshop of International Centres of Excellence in Malaria Research at Guilin, China from 21–23 August 2013.
8. Presented Plenary talk on IVM in malaria elimination: Implementation, evidence and impact assessment in VI Multilateral Initiatives on Malaria (MIM) conference held at Durban,

South Africa from 6–10 October 2013.

9. Presented (Invited: Plenary session) a paper entitled "Impact of insect growth regulating (IGR) compound, Novaluron 10% EC (mosquiron), spraying in Goa, India: Phase-III evaluation of the effectiveness on mosquito vector control in urban settings" in the International Conference on Entomology organized by the Department of Zoology and Environmental Sciences, Punjabi University, Patiala, Punjab, India from 21–23 February 2014.
10. Dr Ashwani Kumar & Dr Hemanth Kumar participated in FICCI meeting on" DST-Lockheed Martin, FICCI India Innovation Growth Programme 2014 at "Hotel Cidade De Goa", Near Vainguinim, Dauna Paula, Panjim, Goa on 23 December 2013.
11. Sh. AK Mohanty attended XII International Conference on Vector and Vector Borne Diseases (ICOV-12) organized by Department of Zoology, Mohan Lal Sukhadia University, Udaipur, Rajasthan from 16-18 September, 2013. Poster presentation, Abstract entitled "Proteomic landscape of Indian vector of malaria, *Anopheles stephensi*".

#### **Dr Hemanth Kumar (SRS)**

1. Attended a Brainstorming conference on Dengue Scenario in India: Disease burden, surveillance and control in Madurai organized by the Centre for Research in Medical Entomology (ICMR) and presented a paper entitled "Impact of insect growth regulating (IGR) compound, Diflubenzuron 25% WP (Bi-Larv) spraying on Aedines in Goa, India" from 25–26 July 2013.
2. Participated in 83rd Annual session and the symposium on "Space for Human Welfare organised by The National Academy of Sciences of India at NIO and Goa University, Goa from 5–7 December 2013.
3. Scientists and staff participated and represented NIMR, FU, Panaji in II Science Expo 2013 organized by Sansa Foundation Exhibition, New Delhi at Hotel Fidalgo, Panaji from 28–30 September 2013.

#### **Awards, Prizes received**

Dr Ashwani Kumar, Scientist F was awarded Bio-Tech International Award 2013 for Excellence in



Dr Ashwani Kumar being presented with Bio-Tech International Award for Excellence for the Promotion of Bio-control of malaria by Dr V.M. Katoch, Secretary, DHR, Govt. of India and DG, ICMR in the International Conference on Vector Borne Diseases at Udaipur.

Promotion of Bio-control of Vectors in an International Conference on Vectors organized by the National Academy of Vector Borne Diseases at Udaipur on 18 September 2013 at the hands of Dr VM Katoch, Secretary, DHR and DG, ICMR.

#### **Conferences/Meetings/Workshops/Trainings organized**

1. Training of 16 laboratory technicians of health services and Goa Port Hospital in 4 batches was conducted in 2013.
2. TOT for officials from Goa, Karnataka and Maharashtra: 30 Officers trained.
3. Field training of Medical and Health Officers of NVBDCP Goa was conducted.
4. Training of 4 field staff members of Voluntary Health Organization, Goa engaged by NVBDCP Goa was conducted.
5. Dr Hemanth Kumar conducted a programme from 16–17 July 2013 for the staff of Voluntary Health Association of Goa, Panaji, Goa. A lecture on Introduction on Malaria and other Vector Borne Diseases: Problem and control was given.
6. As a part of health education NIMR, FU, Goa has supplied CD on malaria, exhibits, leaflets and pamphlets on malaria and vector borne diseases to Montessori Primary School Goa Velha for organizing their school Health day on 20 July 2013.
7. Dr Ashwani Kumar & Dr Hemanth Kumar delivered lectures and supervised/guided field visits from 5–6 August 2013 to the batch of 10 students of M.Sc. Public Health Entomology from Vector Control Research

- Centre (ICMR), Puducherry who visited NIMR, Goa as a part of their academic tour.
8. B.Sc. (Microbiology), Royal College of Arts, Science & Commerce, Thane (27 students) visited NIMR, Field Unit as a part of their academic tour. They were lectured on Malaria and other Vector Borne Diseases: Problems and control and were shown the laboratory and other malaria-related things on 18 November 2013.
  9. A two-day field visit on 11 and 18 November 2013 was conducted for the students of B.Sc. Biotechnology, Dhempe College of Arts and Sciences, Mirmar, Panaji, Goa. They were shown the mosquito breeding sites, sampling and collection of larvae etc. They were also acquainted with the control methods undertaken by the programme.
  10. B.Sc. (Zoology), Government College, Calicut, Kerala (25 students) visited NIMR, Field Unit on 2 December 2013 as a part of their academic tour. They were lectured on Malaria and other Vector Borne Diseases: Problem and control and were shown the laboratory and other malaria-related things.
  11. Dr Ashwani Kumar served as an Expert and

delivered lectures in a training programme on Mosquito control in and around Marmagoa seaport & Goa Airport, organised by the Port Health Organisation, Marmagoa Port Trust, Vasco-da-Gama, Panaji, Goa, from 28–30 January 2014. Dr Hemanth Kumar, participated and guided field survey in the programme.

12. Final year B.Sc. (Zoology) 29 students of the Postgraduate Department of Zoology, Nirmala College, Ernakulum (Kerala) visited NIMR, Field Unit on 4 February 2014 as a part of their academic tour. They were lectured on Malaria and other Vector Borne Diseases: Problem & control and were shown the laboratory and other malaria-related things.

#### **Ph.D. students working**

1. Mr Ajeet Kumar Mohanty (Goa University) *Ph.D. topic:* Midgut proteome analysis of female *Anopheles stephensi* Liston, a vector for human malaria
2. Ms Rakhi Dhawan (Goa University) *Ph.D. topic:* Proteogenomic analysis of midgut and salivary gland in *Aedes aegypti* Linn.



## 3.8

### Raipur (Chhattisgarh)

#### **Phase-III evaluation to compare insecticidal efficacy and household acceptability of DawaPlus 2.0 long-lasting insecticidal mosquito nets with deltamethrin-treated nets (PermaNet 2.0) in India**

Evaluation of DawaPlus 2.0, a deltamethrin-treated polyester net is being made to compare its insecticidal efficacy, household acceptability and longevity over a period of three years in comparison to PermaNet 2.0, a WHO approved deltamethrin-treated LLIN in selected villages of Kanker and Balod districts. A total of 10 villages have been selected in Kanker and Balod districts for the evaluation purpose. No LLINs were previously provided in these villages by the State Health Department. However, community-owned nets were observed in some of the households. Out of 10 villages, six were from Charama block, Kanker district and four from Gurur block, Balod district. The population census data of these 10 villages were recorded by our research team during August-September 2013. The total population of these villages is 4055 (2016 males and 2039 females) residing in 919 houses. Based on the population census data a total number of 1812 LLINs (891 DawaPlus 2.0 and 921 PermaNet 2.0) were distributed so as to cover the entire households of the selected villages.

To ensure that the target dose of the insecticide applied is within the WHO specified limits of  $80 \text{ mg/m}^2 \pm 25\%$ , 30 nets each of DawaPlus 2.0 and LLINs PermaNet 2.0 were destructively sampled as per the procedure given in the project protocol and 150 sub-samples each of the above candidate LN were sent to the WHO Collaborating Centre, Gembloux, Belgium. The baseline results of chemical analysis were found in accordance with the target dose of insecticide (deltamethrin) as per WHO specifications. As per the standard WHO procedure baseline cone bioassays are in progress

on samples of both types of LLINs. Based on the results of the cone bioassays, the nets were distributed in June 2014.

#### **Impact of insecticide resistance in malaria vectors on the effectiveness of combination of indoor residual spraying (IRS) and long-lasting insecticidal nets (LLINs) in India: A multidisciplinary approach**

The project has been launched in Kondagaon district in southern Chhattisgarh. A Field Unit has been established at Kondagaon, where several categories of staff have been recruited and field work has been initiated. Population census, cross-sectional survey and adult susceptibility tests were carried out to generate the baseline data. Insecticide susceptibility tests were carried out in about 25 sub-centres of Keshkal block of Kondagaon district which has been selected as the study area. Tests were carried out against wild caught *An. culicifacies* using diagnostic dosages of DDT, malathion, deltamethrin and bendiocarb. Cross-sectional blood sample survey was carried out in 47 villages (pop. 69,554) of the block and about 12,037 blood slides were collected, of which about 221 (*Pf* 189; *Pv* 22; *Pm* 1; *Pv+Pf* 7; *Pv+Pf+Pm* 1; *Pf+Pm* 1) were found positive for malaria giving slide positivity rate of about 2%. On the basis of the level of deltamethrin resistance in *An. culicifacies* and malaria endemicity in different villages, about 85 villages selected for the study would be randomized into two groups to receive intervention in the form of IRS alone or IRS in combination with LLIN to observe the effect of insecticide resistance on malaria vectors and malaria morbidity. The progress of the project was discussed in a review meeting of project investigators of five sites held at Nairobi, Kenya in January 2014. Various project activities as envisaged in the protocol are under progress.

## **Large-scale (Phase-III) evaluation of efficacy, fabric integrity and community acceptability of Olyset Plus long-lasting insecticidal nets compared with Olyset Net in India**

The LLINs (Olyset Plus and Olyset) have been received from the company and 30 LLINs of each brand were destructively sampled (5 samples/net) as per the standard WHO Protocol. These samples (300) were sent to WHO Collaborating Centre, Gembloux, Belgium for baseline chemical analysis and have been reported to meet the WHO specifications. It is planned to distribute the nets during June in the villages of Kanker district.

Olyset Plus LN is made of 150 denier high density mono-filament polyethylene yarn containing 2% permethrin (w/w) corresponding to 20 g a.i./kg (about 800 mg a.i./m<sup>2</sup>) and piperonyl butoxide (PBO) 1% (w/w), as synergist, corresponding to PBO 10 g/kg (about 400 mg of PBO/m<sup>2</sup>) which is incorporated in all the fibres on all the sides and also on the roof. Permethrin and the synergist migrate through the net by diffusion at a constant ratio of 2:1, thus ensuring enhanced efficacy against mosquitoes. The rate at which permethrin and PBO migrate to the surface of the net has been adjusted to provide rapid regeneration, making the net active again within 1–2 days after washing. Olyset Net is a 100% high density polyethylene, 150 denier net, blended with permethrin 2% (w/w) as active ingredient, corresponding to about 1000 mg of active ingredient/m<sup>2</sup>.

Cone-bioassays using laboratory-reared susceptible *An. stephensi* are in progress. So far the results of bioassays on both the nets have shown that 3 min exposure provides 100% knockdown and 100% mortality. Further bioassays are in progress.

## **A longitudinal study on the bio-efficacy, fabric integrity and survivorship of field distributed long-lasting insecticidal nets for malaria control in Chhattisgarh state**

The State Health Department has distributed large numbers of long-lasting insecticidal nets in various districts of the state during different phases of the implementation of World Bank supported malaria control project. From the programme point of view, it is pertinent to know whether these nets are actually being used by the communities (usage practices), their washing behaviour and longevity of the distributed LLINs. Household surveys were carried out to collect information on the net usage

behaviour of the community, fabric integrity of the LLINs distributed and their survivorship after more than three years of use. Follow-up studies were carried out for two types of LLINs, viz. PermaNet 2.0 distributed by the State Health Department in the villages of Antagarh block, Kanker district and DuraNet distributed by NIMR Field Unit, Raipur in 5 villages of Basti PHC in Bilaspur district. The information as per WHO guidelines were collected on the structured questionnaires during house-to-house visits in the selected villages. Analysis of data so far collected and further work are in progress

## **Technical support to the programme**

### **Cross-checking of malaria blood slides**

Cross-checking facility for malaria slides was provided by the Field Unit. During the reported period, 12,434 malaria slides received from 16 districts of Chhattisgarh through the State Programme Officer (Malaria), Raipur were cross-checked at the Field Unit. Discrepancy of 1.8% was observed in positive slides and discrepancy of 0.5% was observed in negative slides. Results were communicated to the concerned agency.

### **Malaria Clinic**

A total of 44 persons with fever attended the malaria clinic at the Field Unit. One blood smear was found to be positive for *Pf* infection and one for *Pv* infection.

### **Orientation training of MBBS/BHMS students**

1. From the Govt. Medical College, Raipur 68 students of III year MBBS were imparted one-day orientation training on various aspects of vector-borne diseases and their control.
2. One-day orientation training on malaria was imparted to 15 students of final year BHMS of Maharana Pratap Homeopathic College, Raipur.

### **Refresher training to laboratory technicians**

The Field Unit has received a grant of Rs.10 lakh from the State Health Department to organize refresher training courses in malaria microscopy for the laboratory technicians working in the PHCs/ CHCs.

### **Training to newly recruited project staff**

Training in laboratory and field entomological

techniques to the newly recruited field staff of IIR (WHO) project, Kondagaon was imparted during May-June 2013.

### **Meetings/Workshops attended/organized**

#### **Dr RM Bhatt**

1. Attended 16th WHOPES Working Group meeting held at WHO (HQ), Geneva from 22–30 July 2013 as Temporary Adviser to review the reports of testing and evaluation of pesticides.
2. Attended a meeting held at Nairobi, Kenya from 27–30 January 2014 to review the progress of the project on the Implications of Insecticide Resistance (IIR) being implemented in Kondagaon district of Chhattisgarh and to prepare an activity plan and budget for 2014–15.

#### **Dr GDP Dutta**

1. Attended monthly review meeting of District Malaria Officers which was chaired by the Director, Health Services, on 1 June 2013.

#### **Drs SN Sharma and GDP Dutta**

1. Participated in the Intersectoral co-ordination

committee meeting held at DHS, Chhattisgarh, New Raipur on 29 May 2013.

2. Attended Malaria Clinic Committee meeting held at RLTRI Hospital on 6 November 2013.
3. Assisted RS Yadav, WHO Geneva during his visit to Field Unit, Raipur from 29–30 January 2014.

#### **Dr SN Sharma & MP Singh**

1. Attended meeting chaired by the Commissioner, Municipal Corporation, Raipur for the control of vector-borne diseases in urban areas of Raipur on 22 August 2013. A local RAC chaired by Dr PL Joshi, Former Director of NVBDCP and Retd. Prof. MKK Pillai (Delhi University) was held at Raipur on 12 October 2013 to review the research activities carried out so far by the Field Unit and provide guidance on future course of action. The meeting was also attended by the local RAC Members — Dr Kamlesh Jain, Nodal Officer, DHS, Prof. AK Gupta, HoD, SoS in Life Sciences, Pt. Ravishankar Shukla University, Raipur and Dr V Jaipraksah, SPO-VBD.



# 3.9

## Ranchi (Jharkhand)

Jharkhand state is highly endemic for malaria and contributes about 7% of the total malaria cases in the country. It has relatively stable malaria transmission with a yearly average slide positivity rate (determined from both passive and active surveillance) of 15% over the last three years. *Plasmodium falciparum* accounts for 44% of the cases while *P. vivax* accounts for 56%. The forest, hilly terrain, favourable climate, inaccessible area, tribal culture and social unrest has aggravated the malaria situation. On the basis of epidemiological information, out of 24 administrative districts, 13

districts, namely West Singhbhum, Simdega, Sahibganj, Lohardaga, Gumla, Dumka, Ranchi, Khunti, East Singhbhum, Saraikela, Pakur, Godda and Latehar have been identified as most vulnerable to malaria. After the introduction of new epidemiological tools, RDT for diagnosis, ACT for the treatment of *falciparum* malaria and long-lasting insecticidal nets (LLINs), the API of Jharkhand state reduced from 3.63 to 2.84% in the year 2013 (Fig. 1). In the year 2014, in addition to the new epidemiological tools, the NVBDCP has introduced bivalent (RDT) kits for diagnosis. Other vector-

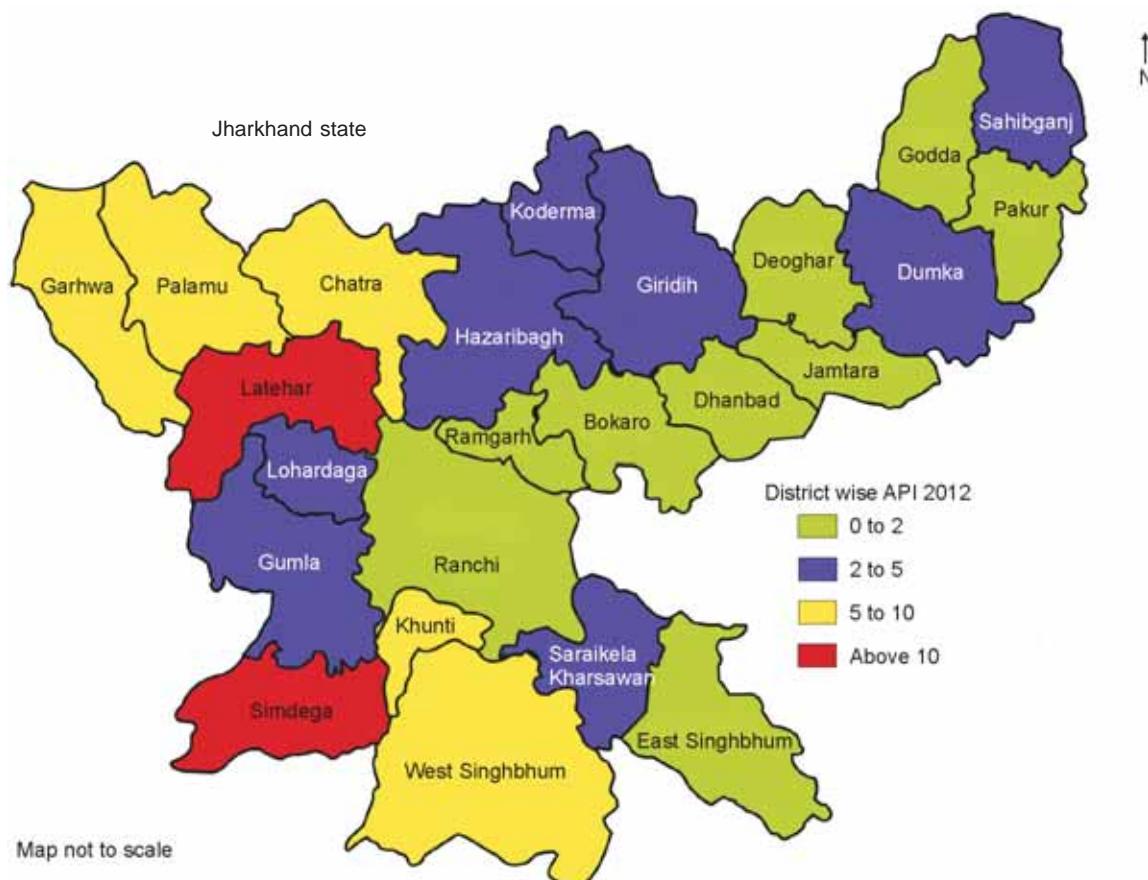


Fig. 1: District-wise annual parasite incidence (API) of malaria in Jharkhand state (Source: SVBDCP).

borne diseases contributing to prevalent in the state are filariasis, dengue, chikungunya, Japanese encephalitis and kala-azar. There is very scanty information on the transmission dynamics of various vector-borne diseases and health seeking behaviour of the tribal population is very poor. To find solution to control vector-borne diseases in Jharkhand state, the National Institute of Malaria Research Field Unit is working in close collaboration on the need-based research programme of the state government.

### Mosquito fauna survey

The survey for the species composition of anopheline mosquitoes was undertaken in West Singhbhum district (Saranda forest and Nuamundi area) during January to October 2013. A total of 21 species of anopheline mosquitoes were collected from Saranda forest area (Table 1) and 18 species of anopheline mosquitoes were collected from Nuamundi area (Table 2 — border of Jharkhand and Odisha states) (Fig. 2). The four major malaria vectors *An. culicifacies*, *An.*

**Table 1. Percent composition of Anophelines in West Singhbhum district (Saranda forest area)**

S.No.	Anopheline species	Total mosquitoes	% composition	S.No.	Anopheline species	Total mosquitoes	% composition
1.	<i>An. annularis</i>	648	17.4	11.	<i>An. jeyporiensis</i>	13	0.34
2.	<i>An. culicifacies</i>	986	26.48	12.	<i>An. karwari</i>	3	0.08
3.	<i>An. fluviatilis</i>	1016	27.28	13.	<i>An. moghulensis</i>	2	0.05
4.	<i>An. minimus</i>	298	8	14.	<i>An. nivipes</i>	3	0.08
5.	<i>An. aitkenii</i>	2	0.05	15.	<i>An. philippinensis</i>	6	0.16
6.	<i>An. barbirostris</i>	78	2.09	16.	<i>An. roperi</i>	3	0.08
7.	<i>An. barbumbrosus</i>	14	0.37	17.	<i>An. serpentii</i>	6	0.16
8.	<i>An. crawfordi</i>	12	0.32	18.	<i>An. stephensi</i>	3	0.08
9.	<i>An. insulaeflorum</i>	3	0.08	19.	<i>An. splendidus</i>	334	8.97
10.	<i>An. jamesii</i>	12	0.32	20.	<i>An. subpictus</i>	268	7.19
				21.	<i>An. tessellatus</i>	13	0.34



**Fig. 2:** *Anopheles minimus* collection sites from the forest of West Singhbhum (Nuamundi area).



Fig. 3: *Anopheles minimus* adult and larval collection sites.

**Table 2. Percent composition of anophelines in West Singhbhum (Nuamundi area)**

S.No.	Anopheline species	Total mosquitoes	% composition	S.N	Anopheline species	Total mosquitoes	% composition
1.	<i>An. annularis</i>	745	22.24	10.	<i>An. jeyporiensis</i>	20	0.59
2.	<i>An. culicifacies</i>	968	28.90	11.	<i>An. karwari</i>	3	0.08
3.	<i>An. fluviatilis</i>	1095	32.69	12.	<i>An. moghulensis</i>	2	0.05
4.	<i>An. minimus</i>	286	8.53	13.	<i>An. nivipes</i>	3	0.08
5.	<i>An. barbirostris</i>	67	2	14.	<i>An. pallidus</i>	88	2.62
6.	<i>An. barbumbrosus</i>	16	0.47	15.	<i>An. philippinensis</i>	6	0.17
7.	<i>An. crawfordi</i>	12	0.35	16.	<i>An. roperi</i>	3	0.08
8.	<i>An. insulaeflorum</i>	3	0.08	17.	<i>An. serpentii</i>	6	0.17
9.	<i>An. jamesii</i>	13	0.38	18.	<i>An. tessellatus</i>	13	0.38

*annularis*, *An. fluviatilis* and *An. minimus* were collected from Saranda jungle and Nuamundi area. *Anopheles minimus* is the dominating mosquito species in Nuamundi area. From Sarju forest area of Latehar district, 16 species of anopheline mosquitoes were collected. *Anopheles minimus* ( $n=298$ ) were collected from West Singhbhum district of Saranda-Singhbhum range. In addition to this, *An. minimus* were collected from the nearby villages (Fig. 3) of Noamundi and Badajamda PHC. Sequencing of 28S rDNA confirmed that specimens identified morphologically as *An. minimus sensu lato* were actually *An. minimus sensu stricto*.

Mosquito blood meal (MBM) analysis of *An. minimus* collected from Nuamundi area revealed positive for human blood (anthropophilic index 60%). *Anopheles minimus* collected from both the forested areas were subject to sporozite ELISA for vector incrimination and molecular analysis. Studies are in progress.

#### Field evaluation of NetProtect LLIN (impregnated with deltamethrin) against malaria vectors and its impact on malaria in Jharkhand

Field evaluation of NetProtect – a long-lasting insecticidal net (LLIN) incorporated with

deltamethrin @ 55 mg/m<sup>2</sup> manufactured by Best Net Insect Control was initiated during December 2010 to evaluate its impact on malaria vectors, *An. culicifacies*, *An. annularis* and *An. fluviatilis* and its impact on malaria transmission in one of the highly endemic areas in Jharkhand state. Cross-sectional and longitudinal surveys were carried out to collect the entomological and parasitological data from deltamethrin-incorporated nets, untreated nets and no net villages. Cone and ring net bioassays were performed in the laboratory. Similarly wash resistance, studies of deltamethrin incorporated nets are completed.

During the third year study, indoor resting density of *Anopheles* vectors *An. culicifacies*, *An. fluviatilis* and *An. annularis* was monitored in each study village. The MHD of *An. culicifacies*, *An. fluviatilis* and *An. annularis* showed declined trend in NetProtect village as compared to untreated and no net villages (Figs. 4–6). There was a reduction in malaria incidence in NetProtect area as compared to the untreated and no net villages (Fig. 4). So far, no adverse event of the net was observed.

The community acceptance of NetProtect was very high as there was 90 to 94% compliance rate of the net usage in the study villages and no adverse

## RANCHI (JHARKHAND)

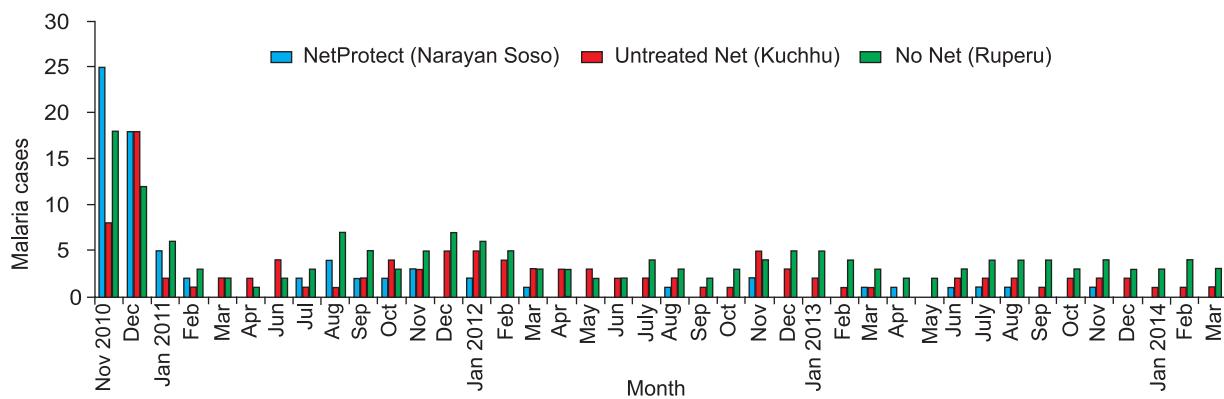


Fig. 4: Malaria incidence in population using NetProtect, untreated net and no net.

event of the net was observed. The third year field evaluation has been completed.

### Filariasis survey

Human filariasis survey was carried out in tribal population of Santhal Pargana (Dumka, Godda, Pakur, Sahibganj and Jamtara districts) (Fig. 5). The above districts are dominated by Santhal, Munda, Oraon, Ho, Lohra and Mohali tribes. The microfilaria (mf) survey was carried out by cross-sectional method in one PHC of each district. The mf rate in the district of Dumka 5.18, Godda 10.13, Pakur 11.5, Sahibganj, 7.65 and Jamtara 12.1%. The age and sex-wise positivity in Poraiyahat PHC of Godda district and Nala PHC of Jamtara district are summarized in Table 3.

### Godda district (Poraiyahat PHC)

Filariasis survey was carried out in Poraiyahat PHC (Godda district) of Jharkhand state (Fig. 6).

The district is dominated by Santhal, Kharia, Bhumij, Kharwar, Oraon and Munda tribes. A total of 2922 persons were surveyed for filariasis, out of which microfilariae were detected in 296 individuals with an overall prevalence of 10.13% (Table 3). The prevalence of mf was significantly higher in males (11.49 vs. 8.68% in females,  $\chi^2 = 8.44, p > 0.05$ ) (Table 3). The youngest mf detected was in a 5 year old girl and a 5 year old boy and oldest mf carrier was an 80 years old male.

A total 2544 microfilariae were observed in the blood smears of 296 positive persons. The mean mf density per positive person was 8.59 per 20 mm<sup>3</sup> of blood. The mean mf density in males and females was 8.49 and 8.73%, respectively and mf ranged between 1 and 108 in males and 1 and 80 in females. The maximum number of parasites found in a single slide was 108 and mf identified as *W. bancrofti*. The highest mf rate was detected in Ratanpur village (17.71%) and the lowest mf rate



Fig. 5: Filariasis survey in Santhal Pargana— Dumka, Godda, Pakur, Sahibganj and Jamtara districts of Jharkhand state.

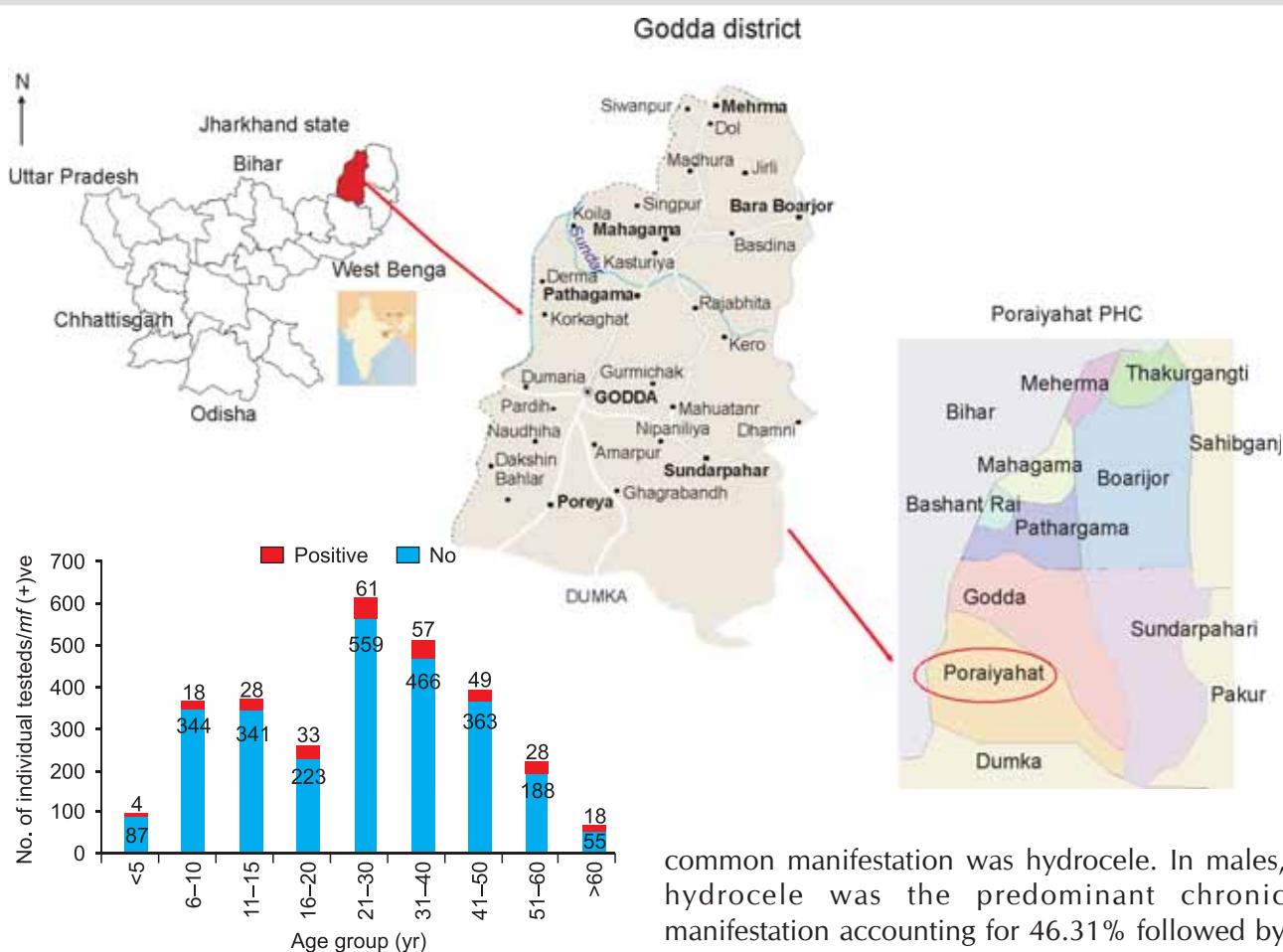


Fig. 6: Study area showing age-wise distribution of microfilaria data in Poraiyahat PHC of Godda district, Jharkhand state (after 6 rounds of MDA); BSC: 2922; mf (+)ve: 296; mf rate (%): 10.13).

was in Hariyari (5%) (Table 3). A total of 447 patients had manifestations of filariasis, out of which 52 (11.6%) being positive for *mf*. The most

common manifestation was hydrocele. In males, hydrocele was the predominant chronic manifestation accounting for 46.31% followed by lymphoedema (37.14%). The overall filarial disease rate and filarial endemicity rate were 16.15 and 26.28%, respectively. The manifestation rate was 22.2% among males and 9.6% among females. The filarial endemicity rate was also significantly ( $p < 0.05$ ) higher among males (33.69%) than females (18.28%).

Table 3. Village-wise microfilaria rate, disease rate and filarial endemicity rate in Poraiyahat PHC of Godda district

Village	Population	Blood slide collection			Microfilaria positive			Microfilaria rate (%)			Manifestation			Disease rate (%)	Filaria endemicity rate (%)
		Male	Female	Total	Male	Female	Total	Male	Female	Total	Male	Female	Total		
Hariyari	915	63	37	100	1	4	5	1.58	10.81	5	13	8	21	21	26
Sundiha	759	62	56	118	5	4	9	8.06	7.14	7.62	21	12	33	27.9	35.52
Siktiya	828	56	76	132	11	1	12	19.64	1.31	9.09	7	5	12	9.09	18.18
Petbi	616	79	62	141	3	4	7	3.79	6.45	4.96	18	3	21	14.8	19.76
Baksara	775	53	55	108	5	5	10	9.43	9.09	9.25	10	4	14	12.9	22.15
Chotte Baksara	582	123	114	237	14	6	20	11.38	5.26	8.43	35	12	47	19.8	28.23
Deodand	1439	126	133	259	11	9	20	8.73	6.76	7.72	47	22	69	26.6	34.32
Kusum tola	821	82	78	160	11	12	23	13.41	15.38	14.37	23	8	31	19.3	33.67
Jagannathpur	646	161	150	311	24	22	46	14.90	14.66	14.79	37	11	48	15.4	30.19
Ratanpur	555	122	132	254	31	14	45	25.40	10.60	17.71	33	17	50	19.6	37.31
Bhatinda	685	135	123	258	13	6	19	9.62	4.87	7.36	14	10	24	9.30	16.66
Bagmara	1556	122	119	241	5	9	14	4.09	7.56	5.80	6	6	12	4.97	10.77
Sarpa	605	178	154	332	23	17	40	12.92	11.03	12.04	32	9	41	12.34	24.38
Angiabandh	530	92	75	167	11	8	19	11.95	10.66	11.37	17	5	22	13.17	24.54
Nawadih	414	51	53	104	5	2	7	9.80	3.77	6.73	22	5	27	25.96	32.69
Total	11726	1505	1417	2922	173	123	296	11.49	8.68	10.13	335	137	472	16.15	26.28

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**Table 4. Age and sex-wise microfilaria positivity rate in Poraiyahat PHC of Godda district**

S.No.	Age group (yr)	Blood slide collection			Microfilaria positive			Microfilaria rate		
		Male	Female	Total	Male	Female	Total	Male	Female	Total
1.	<5	42	49	91	2	2	4	4.76	4.08	4.39
2.	6–10	183	179	362	10	8	18	5.46	4.46	4.97
3.	11–15	188	181	369	14	14	28	7.44	7.73	7.58
4.	16–20	122	134	256	18	15	33	14.75	11.19	12.89
5.	21–30	319	301	620	33	28	61	10.34	9.30	9.83
6.	31–40	261	262	523	26	31	57	9.96	11.83	10.89
7.	41–50	211	201	412	41	8	49	19.43	3.90	11.89
8.	51–60	115	101	216	17	11	28	14.78	10.89	12.96
9.	>60	64	9	73	12	6	18	18.75	66.66	24.65

The presence of *mf* among persons with clinical disease is lower than in persons without clinical disease, but was still high. It was observed that *mf* rate increased steeply with age up. However, in females the *mf* rate decreased from 41–50 years of age (Table 4). BinaxNow (antigen detection ICT card) showed 33.33% (10/30) positive among 2–5 years children indicating high transmission in this area. All the 296 *mf* positive cases were treated with standard dose of DEC (diethylcarbamazine) 6 mg/kg/day for 12 days. Six rounds of mass drug administration (MDA) was carried out in Poraiyahat PHC of Godda district.

During the entomological survey, a total of 190 female *Cx. quinquefasciatus* were collected from human dwellings. The man hour density (MHD) of *Cx. quinquefasciatus* was 19. In all, 184 mosquitoes were dissected for the presence of different stages of filarial larvae. Out of which 12 were found to be positive for L1 and L2 stages showing 6.52% of infection rate, and 6 were positive for L3 larvae, showing 3.26% of infectivity rate.

*Mansonia annulifera* (42), *Ma. uniformis* (21) were collected and dissected but none was found

to be positive for L1, L2 and L3 stages.

### Jamtara district (Nala PHC)

Filariasis survey was carried out in Nala PHC (Jamtara district) of Jharkhand state (Fig. 7). The district is dominated by Santhal, Munda, Oraon, Bhumij, Kharia, Lohra, Chero, Bedia and Mahali tribes.

A total of 2582 persons were surveyed for filariasis, out of which microfilariae were detected in 314 individuals with an overall prevalence of 12.16% (Table 5). The prevalence of *mf* was significantly higher in males (11.91 vs. 12.53% in females,  $\chi^2 = 9.98$ ,  $p > 0.05$ ) (Table 6). The youngest *mf* carrier was a 5 year old girl and oldest *mf* carrier was an 85 year old male.

A total of 2326 microfilariae were observed in the blood smears of 314 positive persons. The mean *mf* density per positive person was 7.4 per 20 mm<sup>3</sup> of blood. The mean *mf* density in males and females was 9.75 and 4.04% and the *mf* ranged between 1 and 50 in males and 1 and 78 in females, respectively. The maximum number of parasites found in a single slide was 78. The *mf* identified

**Table 5. Village-wise microfilaria rate, disease rate and filarial endemicity rate in Nala PHC of Jamtara district**

Village	Population	Blood slide collection			Microfilaria positive			Microfilaria rate (%)			Manifestation			Disease rate (%)	Filaria endemicity rate (%)
		Male	Female	Total	Male	Female	Total	Male	Female	Total	Male	Female	Total		
Khaira	1617	214	170	384	21	15	36	9.81	8.82	9.37	50	28	78	20.3	29.67
Dalawad	1534	212	135	347	30	20	50	14.15	14.81	14.40	88	18	106	30.54	44.94
Mohanpur	1155	275	184	459	46	36	82	16.72	19.56	17.86	58	15	73	15.90	33.76
Chakanderkuri	761	28	8	36	1	0	1	3.57	0	2.77	12	4	16	44.45	47.22
Challipada	108	21	27	48	3	3	6	14.28	11.11	12.5	14	10	24	50	62.5
Babupur	517	178	136	314	31	23	54	17.41	16.91	17.19	33	17	50	15.9	33.09
Jivanpur	456	34	45	79	1	4	5	2.94	8.88	6.32	10	13	23	29.1	35.42
Siarkitiya	877	149	90	239	4	4	08	2.68	4.44	3.34	27	9	36	15.06	18.4
Godapiyal	621	180	103	283	27	17	44	15	16.50	15.54	65	10	75	26.5	42.04
Dumariya	510	99	60	159	11	4	15	11.11	6.66	9.43	24	15	39	24.5	33.93
Kalipahari	852	163	71	234	10	3	13	6.13	4.22	5.55	74	14	88	37.6	43.15
Total	9008	1553	1029	2582	185	129	314	11.91	12.53	12.16	455	152	607	23.5	35.66

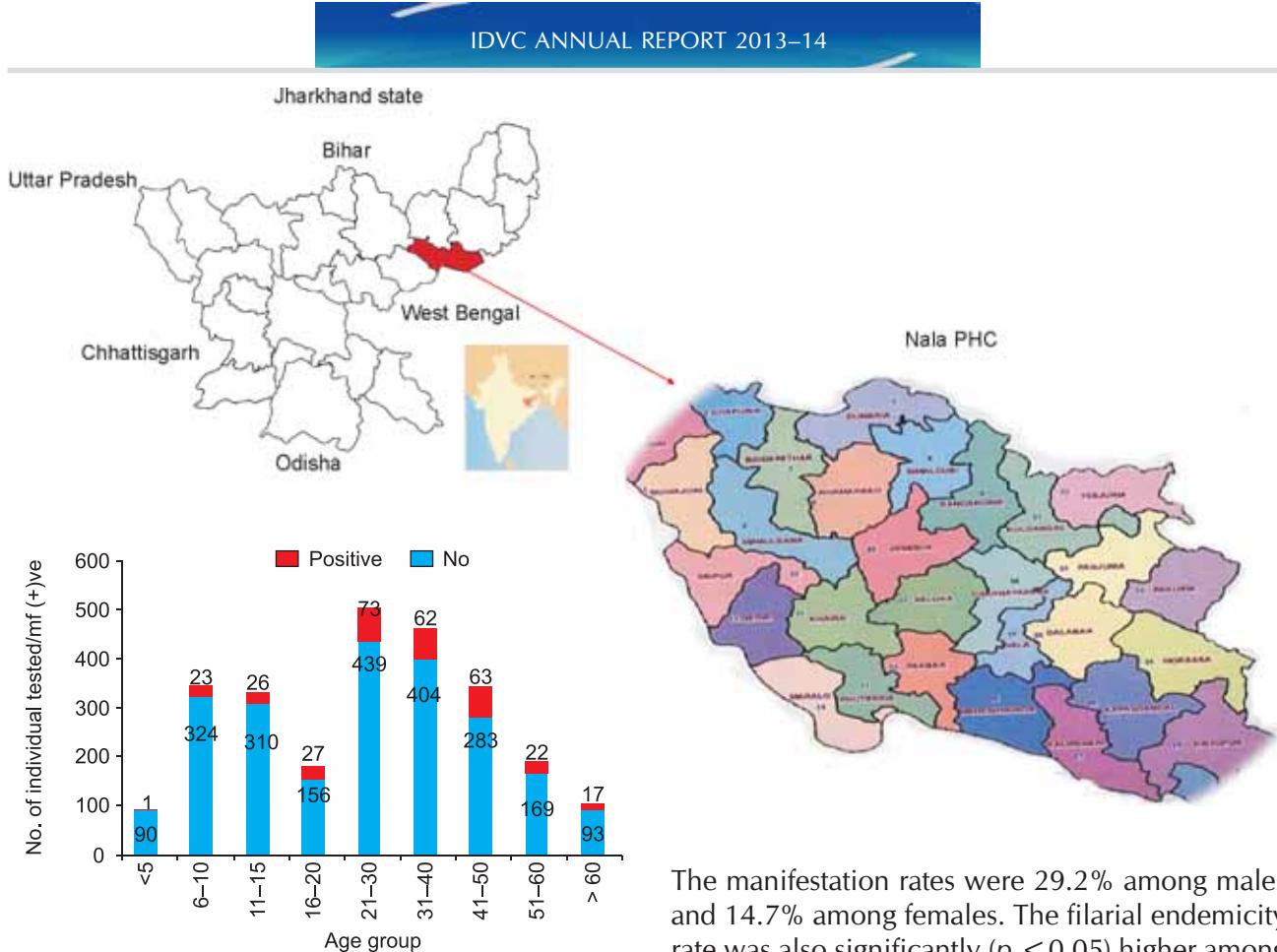


Fig. 7: Study area showing age-wise distribution of microfilaria data in Nala PHC of Jamtara district, Jharkhand state. (No MDA); BSC: 2582; mf (+)ve: 314; mf rate (%): 12.16.

was *W. bancrofti*. The highest mf rate was detected in Mohanpur village (17.86%) and the lowest mf rate was in Chakanderkuri (2.77%). A total of 576 patients had manifestations of filariasis and, out of which 71 (11.6%) being positive for mf. The most common manifestation was hydrocele. In males, hydrocele was the predominant chronic manifestation accounting for 56.6% followed by lymphoedema 25.1%. The overall filarial disease and filarial endemicity rates were 23.5 and 35.66%.

The manifestation rates were 29.2% among males and 14.7% among females. The filarial endemicity rate was also significantly ( $p < 0.05$ ) higher among males (41.1%) than females (27.2%). The presence of mf among persons with clinical disease is lower than in persons without clinical disease, but was still high. It was observed that mf rate increased steeply with age up. However, the mf rate decreased from 51–60 years in both the sexes. BinaxNow (antigen detection card) showed 40% (12/30) positive among 2–5 years children indicating high transmission in this area. All the 314 mf positive cases were treated with standard dose of DEC (diethylcarbamazine) 6 mg/kg/day for 12 days. No mass drug administration (MDA) was carried out in Nala PHC.

Table 6. Age and sex-wise microfilaria positivity rate in Nala PHC of Jamtara district

S.No.	Age group (yr)	Blood slide collection			Microfilaria positive			Microfilaria rate		
		Male	Female	Total	Male	Female	Total	Male	Female	Total
1.	<5	52	39	91	0	1	1	0	2.56	1.09
2.	6–10	189	158	347	13	10	23	6.87	6.32	6.62
3.	11–15	190	146	336	10	16	26	5.26	10.95	7.73
4.	16–20	123	60	183	14	13	27	11.38	21.66	14.75
5.	21–30	331	181	512	52	21	73	15.70	11.53	14.25
6.	31–40	262	204	466	34	28	62	12.97	13.72	13.30
7.	41–50	211	135	346	36	27	63	17.06	20	18.20
8.	51–60	125	66	191	17	5	22	13.6	7.57	11.51
9.	>60	70	40	110	9	8	17	12.85	19.51	15.31
	Total	1553	1029	2582	185	129	314	11.91	12.53	12.16

During the entomological survey, a total of 411 female *Cx. quinquefasciatus* were collected from human dwellings. The man hour density (MHD) of *Cx. quinquefasciatus* was 41.1 and 403 mosquitoes were dissected for the presence of different stages of filarial larvae. Out of which 42 were found to be positive for L1 and L2 stages showing 10.4% of infection rate, and 18 were positive for L3 larvae, showing 4.46% of infectivity rate.

*Culex quinquefasciatus* was incriminated as the vector for transmission of filariasis in all the five districts. The *mf* was identified as *W. bancrofti*. The report of *mf* positive cases was sent to the DMO and treatment with DEC was provided by the PHC and NIMR staff. BinaxNow (antigen detection card) showed high transmission in all the PHCs. The present study highlights the problem of filariasis in the Jharkhand state. The survey will be helpful for LF elimination programme in this state through MDA. Community health education and video clipping programmes were carried out to raise awareness among patients and alleviate the suffering.



Filariasis survey during night camp.

#### A randomized controlled trial of Artesunate + Sulphadoxine-Pyrimethamine (ASP) vs ASP + Primaquine for decreasing malaria transmission in Jharkhand state, India

Artemisinin combination therapies (ACTs) are now the first line of treatment against malaria in most of the countries including India. A key benefit of ACTs is their activity against immature gametocytes resulting in reduction of malaria transmission. Addition of a single dose of PQ alongside the ACT would be desirable to eliminate mature gametocyte population. However, despite recommendations, use of PQ must involve careful safety assessment because of hemolysis. It is

known to cause in individuals with glucose-6-phosphate dehydrogenase (G6PD) deficiency and also for unknown reasons in individuals with wild-type G6PD allele when administered in combination with ACTs. Very few studies have been carried out in malaria endemic regions of India to assess the effectiveness and safety of PQ alongside ACTs for reducing gametocyte transmission.

The trial is being carried out at Jonha Additional PHC, Rinchi Hospital, Jonha in Ranchi district. The study area was dominated by Munda, Oraon, Bedia, Lohra and Birhor tribes. This is a randomized open labelled clinical trial. Cases of malaria were detected by passive surveillance and received artesunate plus sulfadoxine-pyrimethamine in a standard 3-day course (4 mg/kg each day + 25/1.25 mg/kg stat). They were randomized to receive a single dose of either primaquine or no drug (0.75 mg base/kg; usual adult dose 45 mg). Within the patients selected to receive primaquine, they were randomized to receive primaquine on D0 or D2. Thus, the allocation ratio for no PQ, D0 PQ, and D2 PQ is 2:1:1. Patients were tested for haemoglobin, parasite counts, G6PD, and patients were seen weekly for a total of 28 days according to WHO protocol. A total of 197 patients were screened. The enrolment is complete with 110 patients and the data are being analysed. A 28-day follow up was completed in 106 patients where four patients showed lost to follow up and two patients showed *P. falciparum* positivity on Day 28 (Fig. 8). The exact status of two positive cases will be confirmed after PCR analysis. The parasitic density of the enrolled patients ranged from 199-149438/ $\mu$ l.

Patients were also tested for haemoglobin (%), parasite count, G6PD deficiency and follow up as per WHO protocol. Blood spots were collected on Whatman 903 saver card on scheduled days of follow up for RNA extraction and PCR. A total of nine patients showed G6PD deficiency. The parasite clearance was rapid and complete clearance of parasitaemia was observed within 48 h in all the patients. Proportion of patients with gametocytes was 17% (19/110) at the time of enrolment. Further microscopic quantification of gametocytes showed proportion of patients with gametocytaemia on Day 1, 2 and 3 as 11, 9 and 6%, respectively.

Microscopic observation revealed that there was no significant difference in gametocyte clearance

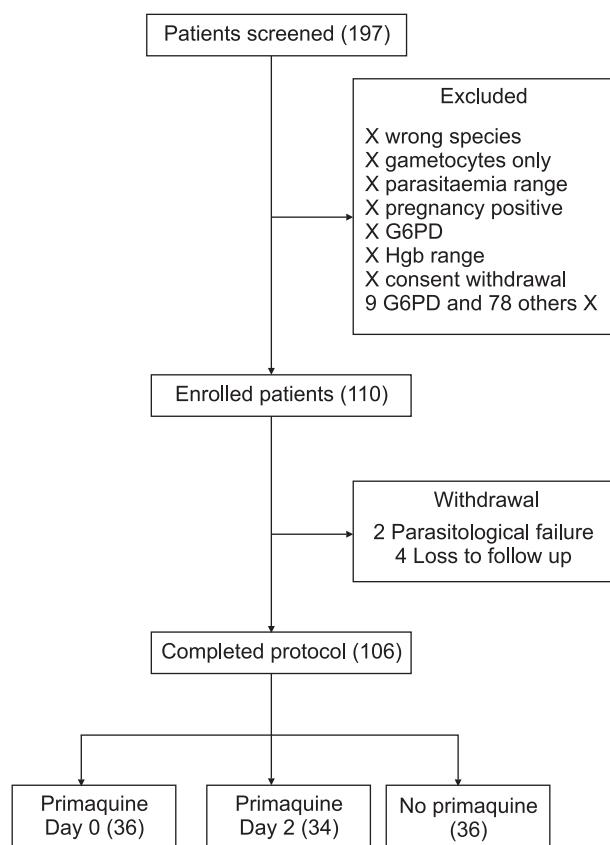


Fig. 8: Summary flow diagram of study recruitment.

in three arms. There was no increased risk of haemolysis due to primaquine therapy in non-G6PD deficient patients. Molecular analysis of the samples is in progress.

#### Enrolment by arm

Arm	N	Mean Hb (g/dL) D0	Mean Hb (g/dL) D7	Gameto-cytaemia D0 (%)
ASP	36	10.77	10.16	18.4
ASP+PQ D0	36	10.35	9.84	21.0
ASP+PQ D2	34	10.38	10.02	18.9

#### Malaria Clinic

Malaria Clinic at the Field Unit provides free diagnosis facility to the patients attending the clinic. A total of 1174 patients attended the clinic up to March 2014, out of which 70 cases were positive for malaria. Of these, 20 cases were positive for *P. vivax* and 50 cases were positive for *P. falciparum*. Overall SPR was 5.9% , SFR was 4.2% and 6 *P. falciparum* positive patients showed gametocytes in the peripheral blood (Table 7).



Surveillance of malaria cases.

Table 7. Malaria Clinic data of IDVC Field Unit, Itki, Ranchi (2013–14)

Month	BSC	Total (+)ve	Pv	Pf	SPR	SFR
Mar 2013	86	5	1	4	5.8	4.6
Apr	74	3	2	1	4.0	1.3
May	50	2	2	0	4.0	0
Jun	81	7	3	4	8.6	4.9
Jul	123	8	3	5	6.5	4.0
Aug	139	9	2	7	6.4	5.0
Sep	149	6	1	5	4.0	3.3
Oct	136	6	1	5	4.4	3.6
Nov	104	7	1	6	6.7	5.7
Dec	97	5	1	4	5.1	4.1
Jan 2014	35	3	1	2	8.6	5.7
Feb	48	5	1	4	10.4	8.3
Mar	52	4	1	3	7.6	5.7
Total	1174	70	20	50	5.9	4.2

#### Filaria Clinic

A Filaria Clinic is functioning at IDVC Field Unit, Itki, Ranchi. A total of 46 patients of filariasis attended the clinic during the year. Most of the cases were old cases of filariasis. These cases were with acute manifestation of filariasis starting from hydrocele to elephantiasis. One case of epididymo-orchitis was observed. Only two patients had multiple manifestations (4.34%) (Table 8). A total of 85 mf positive cases were treated with DEC.

Table 8. Clinical profile of 46 patients of filariasis

Symptom/Sign	No. of Patients*	Percentage
Elephantiasis	6	13.04
Lymphoedema(Hand)	1	2.17
Lymphoedema(Leg)	17	36.95
Hydrocele	20	43.47
Lymphadenitis	0	0
Epididymo-orchitis	1	2.17
Lymphangitis	1	2.17

\*2 patients had multiple manifestations (4.34%)



Affection of the toes along with the leg and foot (Elephantiasis).



Elephantiasis of leg with skin nodules.



Elephantiasis of superior extremity.

### Future Programme

1. To study the prevalence of G6PD deficiency and haemoglobinopathies in tribal population of Jharkhand state.
2. To study the haemoglobinopathies, its genetic epidemiology with special reference to alpha thalassemia and mass screening programme in malaria endemic tribal population of Jharkhand state.
3. Genotype of *W. bancrofti* of Jharkhand state.
4. Identification of foci of kala-azar and the reasons for its persistence in Santhal Pargana (Sahibganj, Pakur, Dumka and Godda).

### Technical support provided to the programme (SVBDCP)

1. MDA evaluation of Deoghar and Godda districts of Jharkhand state.
2. Epidemiological and entomological surveys for the control of malaria in Angara PHC, Ranchi district, Jharkhand.
3. Preparation of annual malaria action plan.
4. Filariasis survey in Deoghar, Santhal Pargana (Godda, Pakur, Dumka and Sahibganj districts) and Jamtara.

5. Monitoring of insecticide resistance in malaria vectors.
6. Capacity building in the field of training, malaria microscopy, surveillance to MPW, MTS, ANM, Sahiya and health workers.

### Health education and IEC activities

Four health camp were organized in Jonha and Narayan Soso villages. A meeting was jointly organized in Ranchi Trust Hospital, Jonha. The use of long-lasting insecticide-treated nets (LLINs) for protection against mosquitoes and malaria was discussed among the tribal people. Role of RDT and ACT combination therapies was discussed. Lectures and demonstration regarding malaria and use of LLIN were delivered to the tribal people.

In addition to this, 10 health education camps were organized in PHC Shikaripara of Dumka district, Hirapur PHC of Pakur district, Nala PHC of Jamtara district, Pathan PHC of Sahibganj district and Poraiyahat PHC of Godda district, endemic for filariasis. Role of MDA in controlling filariasis and LLIN were discussed among the tribal people

### Dr MK Das

1. An invited speaker in the National planning workshop for district level Health Officers of Bihar, Patna Medical College (National Vector Borne Disease Control Programme from 12–16 March 2013) in collaboration with the National Institute of Health & Family Welfare, Munirka, New Delhi on "Vector of public health importance and control strategy" and "Elimination of filariasis from India" conducted.
2. As a plenary speaker in National symposium on Innovation in Science and Technology for inclusive development, Indian National Science Congress Association Bengaluru Chapter and Mysore University, Mysore on "Malaria in the primitive tribe of Andaman & Nicobar Islands" on 4 January 2014.

### ICMR Travel Grant Award

- ICMR award for International Fellowship of Senior Biomedical Scientist 2013.

### Trainings undertaken

- Training on Molecular methods of malaria

parasite detection including molecular surveillance methods for monitoring drug resistant malaria parasites and monitoring of insecticide resistant malaria vectors using the CDC bottle assay during the visit to CDC Atlanta, USA from 6 to 20 February 2013.

- Training on Safety survival skills—Part I :

General responsibilities (course ID: CDC OHS SSSI) at CDC Atlanta, USA on 8 February 2013.

- Training in Safety survival skills—Part 2: Laboratory safety (course ID: CDC OHS SSSI) at CDC, Atlanta, USA on 8 February 2013.



## 3.10

### Rourkela (Odisha)

#### Comprehensive case management programme in Odisha

The MMV funded Comprehensive Case Management Programme was launched in collaboration with the Government of Odisha, in four districts with the primary objective to assess the impact of comprehensive case management system of uncomplicated malaria on its transmission in different transmission settings. Puintala, Athamallick, Hindol and Nuagaon have been taken as intervention blocks, whereas Saintala, Bhuban, Chhendipada and Khajuripada have been taken as

control blocks in Bolangir, Dhenkanal, Angul and Kandhamal districts, respectively (Fig. 1). Recruitment and training of project staff were completed by May 2013 and the CCM project units started functioning in the intervention blocks from July 2013. The existing staff of the PHCs were also trained.

In the low endemic Bolangir district, the ABER, SPR and API, respectively were 17.1, 1.1 and 0.2 in the intervention block, whereas the corresponding indicators in the control block were

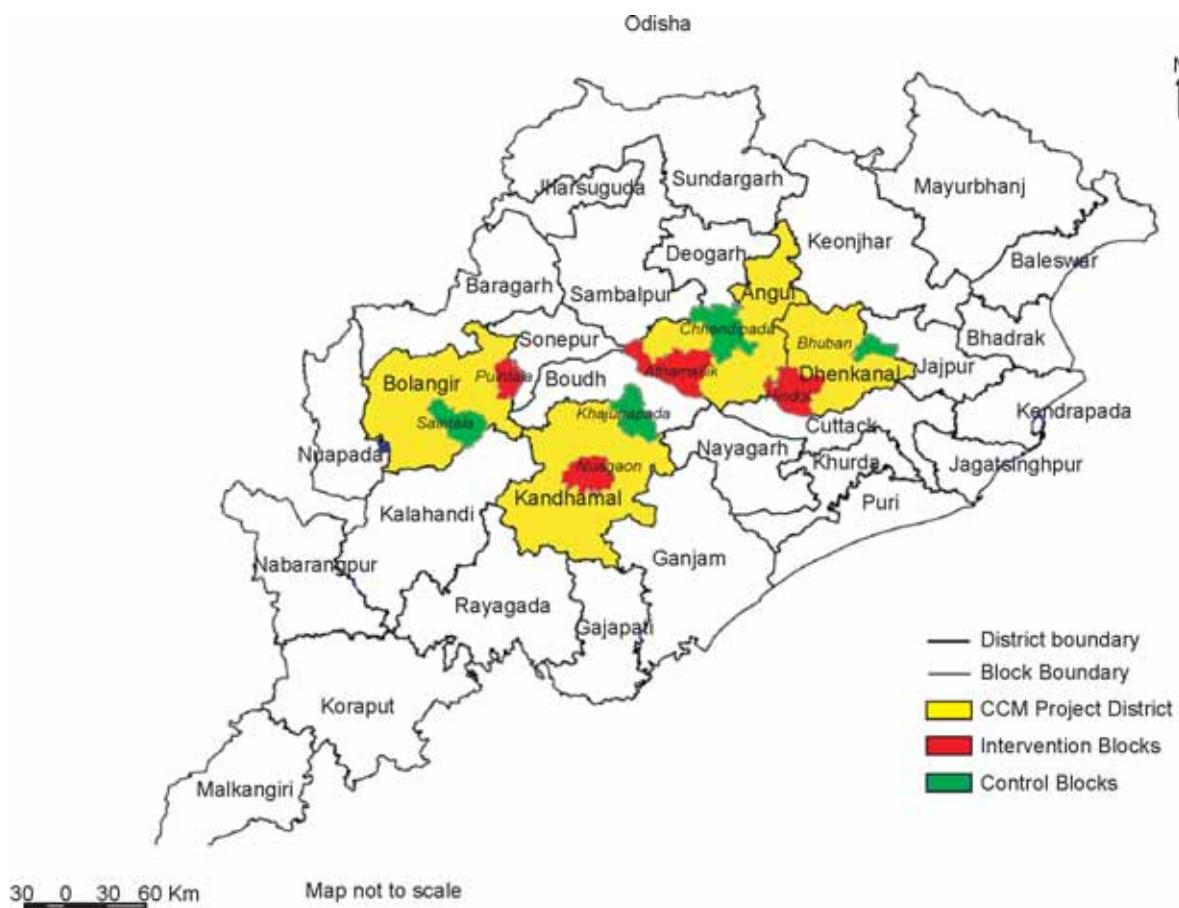


Fig. 1: Map showing the intervention and control blocks in four districts of Odisha under CCM project.

16.1, 2.5 and 0.4, respectively during the year 2013–14 (Fig. 2). In the intervention block of meso-endemic Dhenkanal district, the ABER, SPR and API were 12.5, 2.3 and 2.9, whereas in the control block the corresponding indicators were 11.7, 0.7 and 0.1, respectively during the year of reporting (Fig. 3). In the high endemic Kandhamal district, the ABER, SPR and API were 22.1, 4.5 and 9.9, respectively in the intervention block, whereas the corresponding indicators in the control block were 29.0, 8.6 and 2.5, respectively (Fig. 4). In the hyper-endemic Angul district, the ABER, SPR and API were 19.2, 14.9 and 28.7, respectively in the intervention block, whereas the corresponding indicators in the control block were 19.4, 2.0 and 3.8, respectively during the year of reporting (Fig. 5).

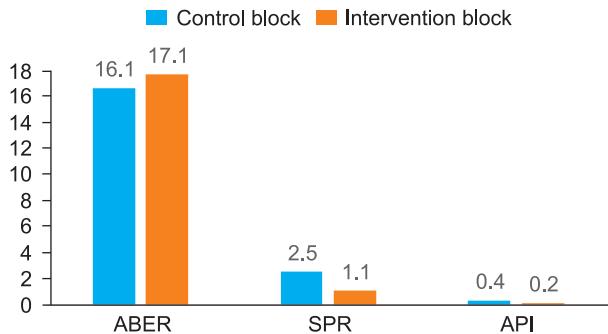


Fig. 2: Malaria indicators in the control and intervention blocks of low endemic Bolangir district.

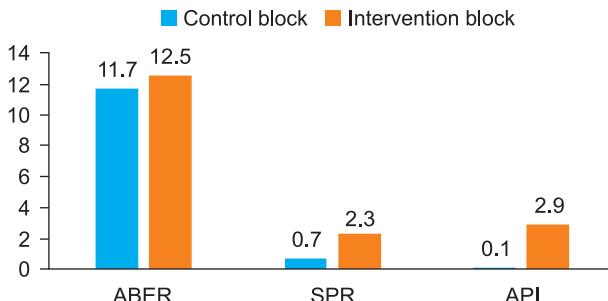


Fig. 3: Malaria indicators in the control and intervention blocks of meso-endemic Dhenkanal district.

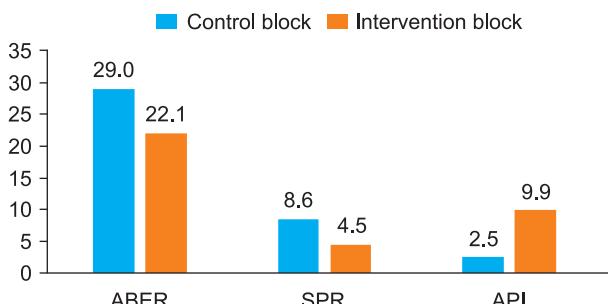


Fig. 4: Malaria indicators in the control and intervention blocks of high endemic Kandhamal district.

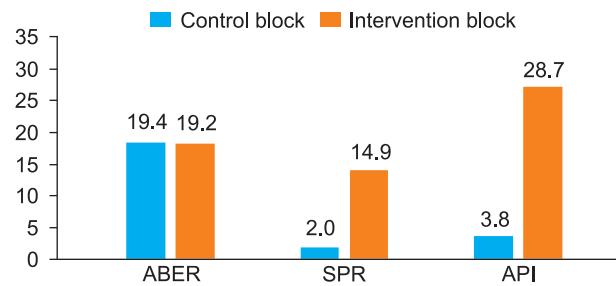


Fig. 5: Malaria indicators in the control and intervention blocks of hyper-endemic Angul district.

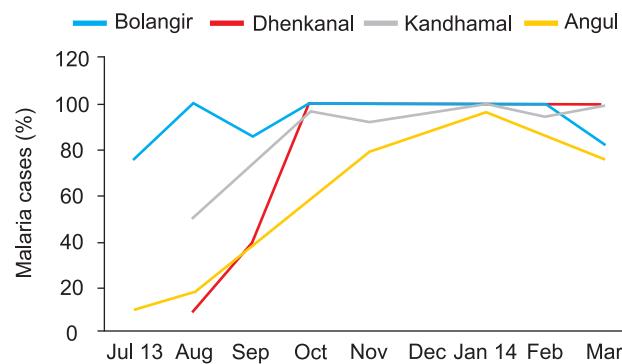


Fig. 6: Follow-up percentage of malaria cases in the intervention blocks of different districts.

In the intervention blocks of all the four districts, *P. falciparum* and *P. vivax* cases were followed up on Day 5 and Day 14 for drug compliance and adverse events, respectively since July 2013. The overall compliance rate of follow up in Bolangir, Dhenkanal, Kandhamal and Angul districts was 90.2, 69, 66 and 60.8%, respectively. However, the month-wise compliance rate of follow up in these districts ranged from 10.1 to 100% (Fig. 6).

One death due to malaria was reported from each of the intervention and control blocks of



Monitoring of work under CCM project.



Monitoring of work under CCM project.

Bolangir district, whereas no death due to malaria was reported from any of the other three districts. Various works under CCM project were closely monitored.

### Eco-epidemiology and transmission of complex malaria in India (under NIH funded CSCMi project)

#### Cross-sectional study

A cross-sectional study was conducted in 11 villages located in forest, plain and riverine areas around Rourkela under Bisra, Kuarmunda and Birkera CHCs of Sundergarh district, Odisha. During the year 2013-14, a total of 1223 subjects were enrolled, out of which 106 were found

positive for malaria under microscopic examination comprising of 88 *P. falciparum*, 15 *P. vivax* and 3 mixed infections (Table 1). The SPR was 8.7 and 85.8% of the total malaria cases were *P. falciparum*. The SPR of children belonging to the age groups <5, 5-9 and 10-17 were 11.5, 12.0 and 14.1, respectively which were higher than the SPR of 8.7 for the adults. In terms of enrolment, females (55.8%) outnumbered males (44.2%). However, the SPR of female subjects was less (7.0) in comparison to that of male subjects (10.7).

Rapid diagnostic test was performed in 1211 cases in which 77 cases were found positive for malaria with TPR of 6.4. The specificity and sensitivity of RDT were 59.4 and 98.7%, respectively in comparison to the microscopic results.

#### Clinic study

During the year of reporting, a total of 570 subjects were enrolled in the clinic study, out of which 22 were found positive for malaria under microscopic examination comprising of 14 *P. falciparum* (63.6%) and 8 *P. vivax* (36.4%). The SPR in the age groups <5, 5-9, 10-17 and adults were 2.2, 0, 7.3 and 3.8, respectively. The SPR of male subjects was 5.1, whereas that of female subjects was 2.1. In terms of enrolment, males (58.9%) outnumbered females (41.1%) in contrast to the cross-sectional study (Table 2).

Table 1. Sex and age group-wise malaria cases detected during the cross-sectional study

Sex/Age group (yr)	Microscopy						RDT							
	BSE	Pf	Pv	Pm	Mixed	Total	SPR	No.	RDT	Pf	Pv	Pm	Mixed	Total
Male	540	46	10	0	2	58	10.7	534	21	5	7	5	38	7.1
Female	683	42	5	0	1	48	7.0	677	27	3	4	5	39	5.8
<5	130	13	1	0	1	15	11.5	128	9	0	1	4	14	10.9
5-9	150	15	2	0	1	18	12.0	147	8	1	3	3	15	10.2
10-17	156	19	3	0	0	22	14.1	155	8	3	4	1	16	10.3
>17	787	41	9	0	1	51	6.5	781	23	4	3	2	32	4.1
Total	1223	88	15	0	3	106	8.7	1211	48	8	11	10	77	6.4

Table 2. Sex and age group-wise malaria cases detected in the clinic study

Sex/Age group (yr)	Microscopy						RDT							
	BSE	Pf	Pv	Pm	Mixed	Total	SPR	No.	RDT	Pf	Pv	Pm	Mixed	Total
Male	336	11	6	0	0	17	5.1	334	9	4	2	0	15	4.5
Female	234	3	2	0	0	5	2.1	234	2	2	1	1	5	2.1
<5	46	1	0	0	0	1	2.2	46	0	0	0	0	0	0
5-9	47	0	0	0	0	0	0	46	0	0	0	0	0	0
10-17	82	3	3	0	0	6	7.3	82	3	2	1	0	6	7.3
>17	395	10	5	0	0	15	3.8	394	8	4	2	1	15	3.8
Total	570	14	8	0	0	22	3.9	568	11	6	3	1	21	3.7

Rapid diagnostic test was performed in 568 cases in which 20 cases were found positive for malaria with the TPR of 3.5. The specificity and sensitivity of RDT were 86.4 and 99.6%, respectively in comparison to the microscopic results.

### Entomological studies

#### Environmental monitoring

A total of 36 automatic data loggers were fixed in 6 villages, viz. Birkera, Jhirpani, Santoshpur, Rangamati, Mahaliapali and Sarla located in forest, plain and riverine areas under Bisra and Birkera CHCs in order to monitor temperature and humidity. These loggers were fixed in a range of indoor (28) and outdoor (8) resting places of mosquitoes. Temperature and humidity were monitored, at hourly intervals, through these loggers from July 2013 onwards.

The minimum and maximum temperatures recorded in the study villages from July 2013 to March 2014 fluctuated in between 12.7°C and 34.7°C. The month-wise minimum and maximum temperatures recorded in all the 6 villages were almost same (Fig. 9). The minimum and maximum humidity fluctuated in between 42.2 and 92.3%

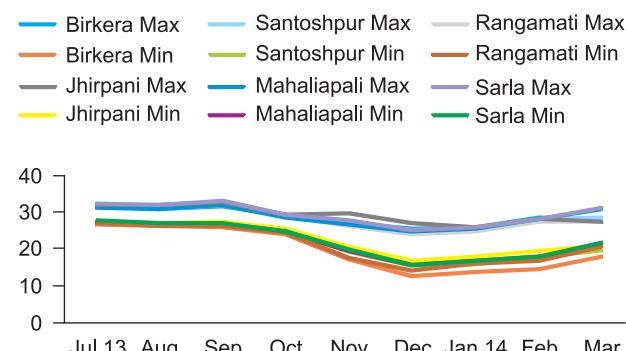


Fig. 9: Minimum and maximum temperatures recorded in the study villages.

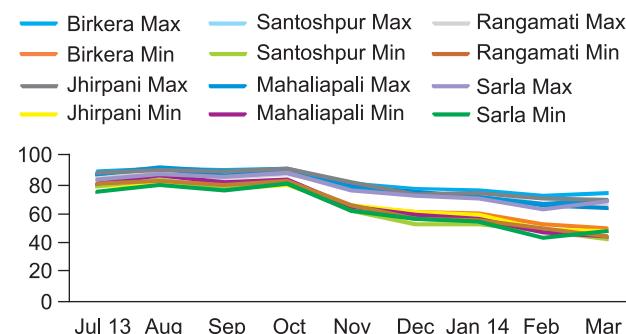


Fig. 10: Minimum and maximum humidity recorded in the study villages.

during the corresponding period. The minimum and maximum humidity were also almost identical in all the 6 study villages (Fig. 10).

#### Vector composition

Hand catches of anopheline mosquitoes were done in the study villages from October 2013 to March 2014. The percentage composition of *An. fluviatilis* was 0.2, 3.1, 6.3, 12.8, 2.9 and 9.0 during the months of October to March, respectively. Similarly, the percentage compositions of *An. culicifacies* during the corresponding period was 9.6, 4.9, 12.9, 22.8, 50.3 and 38.1, respectively (Fig. 11)

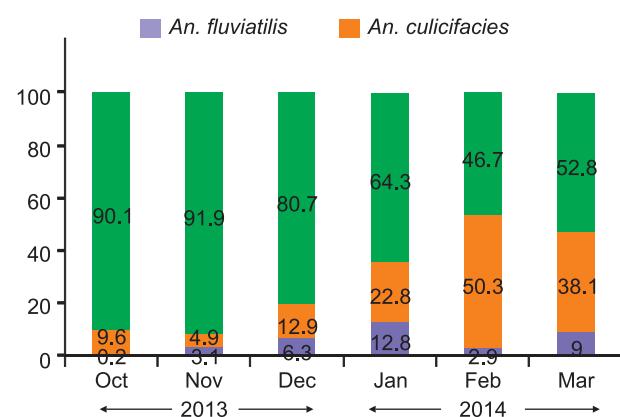


Fig. 11: Percentage composition of vectors in the study villages.

#### Blood meal analysis

Blood meals of 125 *An. culicifacies* and 88 *An. fluviatilis* were analyzed for feeding preference and detection of sporozoites. In *An. culicifacies*, bovine, bovine + human, bovine + goat + human, human and goat + human bloods were found in 14.1, 76.8, 6.4, 1.6 and 0.8% cases, respectively. In *An. fluviatilis*, bovine, bovine + human, bovine + goat + human and human bloods were found in 13.6, 77.2, 4.5 and 4.5% cases, respectively (Fig. 12). Sporozoites were not detected in any of the mosquito species.

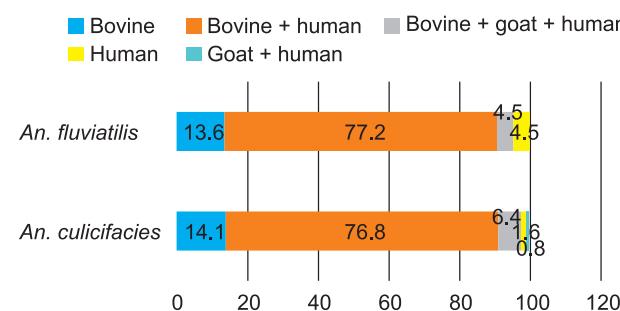


Fig. 12: Percentage of mosquitoes found to have fed with the bloods of human and other animals.

## A Phase-II/III randomized clinical trial of the efficacy and safety of artesunate + sulphadoxine-pyrimethamine and artesunate + mefloquine to treat uncomplicated falciparum malaria in pregnancy

The multi-centric study was being carried out simultaneously at Rourkela, Ranchi and Jamshedpur. In Rourkela, 83 subjects were recruited for the study from a cohort of 3040. The range of parasite density on Day-0 was 32 per ml to 95,280 per ml of blood and all the subjects became negative on D-3. A 63-day follow up was completed in all the 83 subjects and there was no treatment failure.

In all, 81 subjects have completed deliveries. Normal birth was observed in 60 (74%) cases, whereas low birth weight (< 2500 g) and pre-term deliveries ( $\leq 36$  weeks) were observed in 18 (22.2%) and 3 cases (3.7%), respectively. Adverse events to mothers were noticed in only 4 (6.2%) cases, whereas adverse events to children were noticed in 6 cases (9.2%).

A 42-day post-partum follow up was completed in 80 cases. There was no death at all among both the mothers and children. Re-infection with malaria parasite was observed in only one of the subjects (Table 3).

**Table 3. Results of the therapeutic efficacy and safety of the drugs for the treatment of uncomplicated falciparum malaria in pregnancy (up to 31-3-2014)**

<b>Therapeutic efficacy (n=83)</b>	
Number of volunteers/participants in the cohort	3040
Number of patients enrolled	83
Number of patients who have completed	83
63-day follow up	
Loss to follow up	0
Number of treatment failure	0
Number of patients who have completed deliveries	81
Number of patients who have completed 42-day post-partum follow up	80
Number of patients in which reinfection with malaria parasite was observed	1 (1.2)
<b>Birth outcome (n=81)</b>	
Normal birth	60 (74)
Low birth weight (< 2500 g)	18 (22.2)
Pre-term deliveries ( $\leq 36$ weeks)	3 (3.7)
Miscarriage	0
Still birth	0
Neonatal death (28 days)	0
<b>Adverse events (n=65)</b>	
Adverse events to mother	4 (6.2)
Adverse events to children	6 (9.2)

Figures in parentheses indicate percentage.

## Studies on the *Aedes* mosquito in the epidemic prone area of Sundergarh district, Odisha

Sundergarh district is considered as epidemic prone area where 63 and 69 dengue cases have been reported during 2011 and 2012, respectively. Larval survey was carried out in September-October 2013 and in March 2014 in the localities where the dengue cases were reported. During September-October, 2013 the house, container, breteau and pupal indices were 38.7, 24.4, 74.2 and 142.7, respectively. In the subsequent survey carried out in the month of March 2014, the corresponding indices were 28.3, 22, 29.1 and 20.9, respectively (Table 4).

**Table 4. Results of survey of *Aedes* breeding in Sundergarh district, Odisha**

Month/Year	House index	Container index	Breteau index	Pupal index
Sep-Oct 2013	38.7(124)	24.4(377)	74.2	142.7
March 2014	28.3(134)	22.0(176)	29.1	20.9

The matter was discussed with the local authorities, the District Medical Officer and the VBD Consultant for preventive measures to be taken for the control of *Aedes* breeding. Extensive house-to-house campaign was carried out to increase the awareness of people regarding breeding habitats of *Aedes* mosquito, the threat posed by it and to motivate the community for the prevention and control of dengue.

## Studies on insecticide susceptibility status of malaria vectors in six western districts of Odisha

The susceptibility status of *An. culicifacies* to DDT 4%, malathion 5% and deltamethrin 0.05% was studied in Sundergarh district, Odisha during September–October 2013 and January 2014. *Anopheles culicifacies* was found to be resistant to DDT 4% with a mortality rate of 12.9%. The vector was found to be very tolerant to malathion 5% and deltamethrin 0.05% with mortality rate of 84.3 and 88.6%, respectively (Table 5).

**Table 5. Results of insecticide susceptibility study carried out in Sundergarh district, Odisha**

Insecticides	No. of mosquitoes tested	No. of mosquitoes knocked-down in 24 h	Percentage mortality
DDT 4%	350	45	12.9 R
Malathion 5%	210	177	84.3 VR
Deltamethrin 0.05%	510	452	88.6 VR

### Malaria Clinic

During the year April 2013 to March 2014, a total of 4840 patients with fever reported to the clinic run by the NIMR Field Unit, out of which 162 patients were found positive for malaria comprising of 70 *P. vivax*, 88 *P. falciparum*, 1 *P. malariae* and 3 with mixed infection. The SPR, SFR and Pf% were 3.3, 1.9 and 56.2, respectively (Table 6).

**Table 6. Parasitological data of Malaria Clinic at NIMR Field Unit, Rourkela from April 2013 to March 2014**

Month	BSE	(+ve)	Pv	Pf	Pm	Mixed	SPR	SFR	Pf%
Apr 2013	290	4	2	2	0	0	1.4	0.7	50.0
May	284	4	4	0	0	0	1.4	0	0
Jun	491	8	4	2	0	2	1.6	0.8	50.0
Jul	800	26	11	15	0	0	3.3	1.9	57.7
Aug	452	18	6	11	0	1	4.0	2.7	66.7
Sep	481	13	4	9	0	0	2.7	1.9	69.2
Oct	394	15	7	8	0	0	3.8	2.0	53.3
Nov	403	32	8	23	1	0	7.9	5.7	71.9
Dec	298	25	13	12	0	0	8.4	4.0	48.0
Jan 2014	327	7	5	2	0	0	2.1	0.6	28.6
Feb	289	8	5	3	0	0	2.8	1.0	37.5
Mar	331	2	1	1	0	0	0.6	0.3	50.0
Total	4840	162	70	88	1	3	3.3	1.9	56.2

### Trainings/Workshops/Meetings/Health Education Camps organized and attended

#### Training programmes organized

1. An induction level job-specific training programme was organized for the Senior Research Officers and Block Level Managers recruited under CCM project with emphasis on operational malaria control programme from 28 to 31 May 2013. The MMV funded

project was launched in four districts of Odisha in collaboration with the Government of Odisha.

2. An orientation training programme was organized for the students of B.Sc. (Nursing), Ispat General Hospital, Rourkela on Mosquito borne diseases and their control on 3 February 2014.
3. Organized a training programme on 14 March 2014 for the volunteers of a social organization called "Hand Maids of Merry" working in the backward areas of Sundergarh district to develop the knowledge, attitudes and practices of tribal communities. A total of 35 village level Voluntary Community Health Workers (Female) of the social organization participated in the training programme where training on the Prevention and control of malaria and other mosquito borne diseases with special emphasis on personal protection measures including LLIN and acceptance to IRS was given.

#### Meeting attended

1. Mr K Padhan, OIC attended a meeting of the Joint Monitoring Mission at Bhubaneswar on 2 March 2014. He briefed the activities undertaken by the NIMR Field Unit, Rourkela having relevance to the operational programme of the NVBDCP before the Joint Monitoring team visited Odisha state.

#### Health awareness camps organized

1. A team from NIMR Field Unit comprising of Health Educators, LTs and other staff actively



Field training of CCM project staff at Kapatamunda sub-centre under Bisra CHC, Sundergarh.



Health camp being organized by NIMR Field Unit, Rourkela at Vedvyas Shishu Mela.

- participated in the "Malaria Samadhan Shivir" organized in different Blocks/CHCs by the State Health Department during November, 2013 to increase awareness among the tribal people about Prevention and control of malaria through exhibitions.
2. A team from NIMR Field Unit participated in the "Vedvyas Shishu Mela" at Vedvyas Gurukul Ashram, Rourkela on 9 February 2014 in which a health camp was organized by providing free diagnosis and treatment for malaria. An exhibition was also organized on the Prevention and control of malaria. The programme was organized in collaboration with Youth Hostels Association of India, Ispat General Hospital, SAIL, Kalinga Vaishya Yuvak Sangha, and District Drugs Distributors Association.
3. Several health awareness camps and exhibitions were organized in different tribal villages and educational institutions in Rourkela and in rural areas of Sundergarh district, Odisha.

□

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3. Chandra G, Ghosh SK. Use of fish in biological and environmental control of disease vectors. In: *Biological and Environmental Control of Disease Vectors*. Editors Cameron MM, Lorenz LM, Wallingford: CABI 2013; pp. 25–41.
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7. Haq Sarfarazul, Srivastava Harish Chandra. Efficacy of *Aphanus dispar* (Rüppell): An indigenous larvivorous fish for vector control in domestic tanks under the Sardar Sarovar Narmada project command area in District Kheda, Gujarat. *J Vector Borne Dis* 2013; 50: 137–40.
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12. Mishra Neelima, Gupta Ruchi, Singh Sagya, Rana Roma, Shahi Bhartendu, Das Manoj Kumar, Anvikar Anup, Valecha Neena. Insights following change in drug policy: A descriptive study for antimalarial prescription practices in children of public sector health facilities in Jharkhand state of India. *J Vector Borne Dis* 2013; 50: 271–7.
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