ENTOMOLOGY

VECTOR BIOLOGY

Anopheles culicifacies Complex

Bionomics and Distribution Pattern of Members

Anopheles culicifacies collected from highly malarious villages in the District Gadchiroli (Maharashtra) were cytologically examined for sibling species composition. Results revealed that species B and C were sympatric in these villages with predominance of species C (>80%), an established vector of malaria. Similarly, in study villages of Districts Kanker and Bastar (Chhattisgarh) species B and C were sympatric with predominance of latter. In District Hazaribagh (Jharkhand) species A, B and C were sympatric in study villages but species B was predominant comprising 69.4% of the total identified. In Karnataka, cytological examination of *An. culicifacies* samples from study villages of District Bijapur revealed prevalence of species A and B, the former being predominant comprising 88.9%, whereas in District Dharwad only species B was found prevalent in the study villages. In District Udaipur (Rajasthan) species A and B were found sympatric and species A was polymorphic for i^1 inversion. In all the above mentioned districts *An. culicifacies* sibling species were primarily zoophagic.

Molecular Diagnostic Assays for the Identification of Members

The two regions, intertranscribed sequence 2 (ITS2) of rDNA and cytochrome oxidase II (COII) of mitochondrial DNA were analyzed to find species-specific variations to differentiate the so far reported five sibling species of *An. culicifacies* complex. The ITS2 amplicon (~ 500 bp) digested with *Rsa* I could differentiate the five sibling species into two groups—A/D from B/C/E group (Fig. 1). DNA sequencing of these two amplicons (ITS2 and COII) and their se-

HIGHLIGHTS

- In *An. culicifacies* complex, sequence alignment of COII region was utilized to design primers that could differentiate all the five species in two step PCR assays. Microsatellite markers isolated from *An. culicifacies* species A were used for genotyping species A populations from different geographical areas.
- Allele-specific PCR assay developed to differentiate members of *An. fluviatilis* complex was validated using cytologically identified specimens. The results of PCR assay were found to be in agreement with those of cytotaxonomy.
- An album of GIS predicted distribution of anopheline species in India has been produced in form of a CD. It also contains blowup maps of GIS predicted district-wise favourable areas and the validation of GIS predicted distribution through reported surveys.
- Field trials revealed that indoor residual spraying of bendiocarb was highly effective in controlling the densities of DDT and HCH resistant *An. culicifacies*.
- Field evaluation of Agnique MMF (a monomolecular surface film) and Triflumuron (an insect growth regulator) revealed their effectiveness against immature stages of vector mosquitoes.

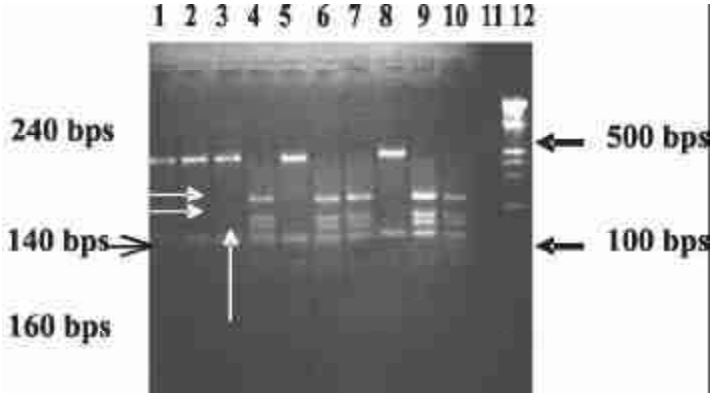

Fig. 1: Rsa I RFLP in ITS2 region of rDNA among the *An. culicifacies sensu lato* collected from Jabalpur. Lanes 1,2,3,5 & 8: Species C; Lanes 4,6,7,9 & 10: Species D; Lane 11: Negative control; and Lane 12: 100 bp ladder

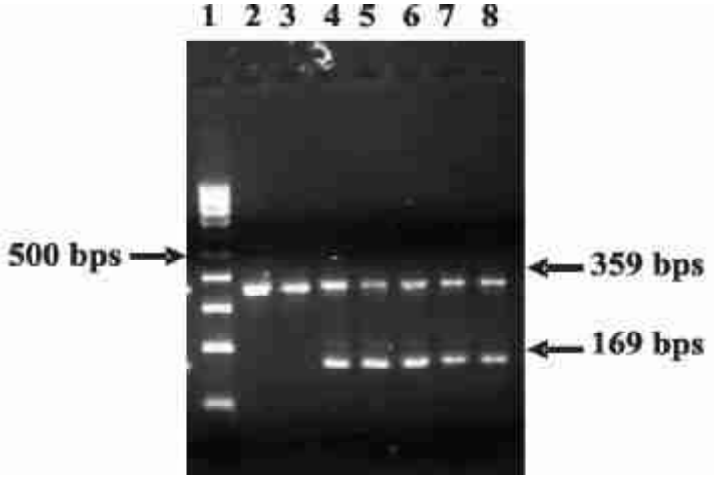

Fig. 2: Result of allele-specific PCR assay to differentiate species A and D among the *An. culicifacies* complex with the primers designed from COII region of mtDNA. Lane 1: 100bp ladder; Lanes 2&3: Species A; Lanes 4–8: Species D from Jabalpur

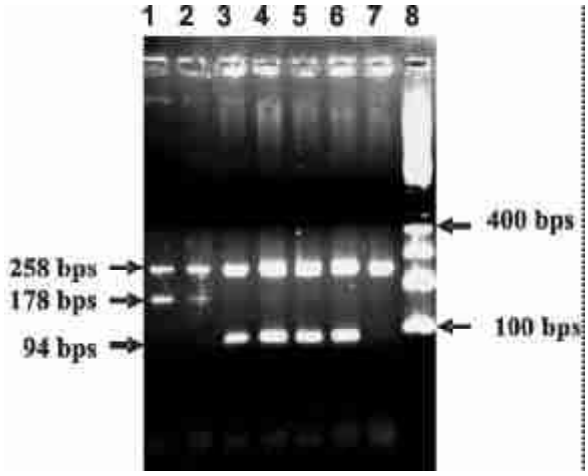

Fig. 3: Result of allele-specific multiplex PCR assay to differentiate species B and E among the *An. culicifacies* complex with primers designed from COII region of mtDNA. Lanes 1–2: Species E from Rameswaram; Lane 3–6: Species C from Bastar; Lane 7: Negative control; and Lane 8: 100 bp marker

quence alignment showed species-specific differences. The ITS2 was not used for primer designing to differentiate the species as the primers for this differentiation have already been designed and evaluated from D3 and D2 regions of 28S rDNA (Annual Report 2001). However, the sequence alignment of COII was utilized to design primers that could differentiate all the five species in two PCR assays on the pre-grouped A/D and B/C/E species by D3/D2-PCR assay. The approach followed for differentiating all the five members of *An. culicifacies* complex was: First—D3/D2 PCR assay to differentiate A/D from B/C/E; Second—A-D-PCR to differentiate species A from species D (Fig. 2); and Third, B-C-E-PCR to differentiate the three species, B from C from E (Fig. 3). These PCR assays are being validated for field use.

Studies on Microsatellite Markers for *An. culicifacies* Species A (WHO/TDR Funded Project in Collaboration with Yale University, USA)

This year 14 new microsatellite markers were isolated from *An. culicifacies* species A and thus the total number of microsatellite markers isolated by Malaria Research Centre, for this species, has increased to 31. Three populations of *An. culicifacies* species A from Districts Kheda (Gujarat), Sonepat (Haryana) and Bijapur (Karnataka) were genotyped using 10 most promising microsatellite markers and two populations from Districts Allahabad (U.P.) and Udaipur (Rajasthan) were genotyped using five microsatellite markers.

Anopheles fluviatilis Complex

Distribution, Bionomics and Biology of Sibling Species

Mapping of the geographical distribution of *An. fluviatilis* sibling species continued. Samples examined from Districts Mandya and Gulbarga (Karnataka) revealed the prevalence of only species T in these districts which was found polymorphic for q^1 inversion. *An. fluviatilis*

collected from Iran Shahr, Baluchistan (Iran) were also examined and species T was found prevalent in this area.

Cytological examination of *An. fluviatilis* population from villages under Laksar PHC of District Hardwar (Uttaranchal) revealed the existence of a new inversion homozygote in this area. The break points for new inversion were identified and the inversion genotype on chromosome arm 2 was observed as $+q^1r^1s^1$. Collections made during summer (April–May) and post-monsoon season (September–December) from villages Dargahpur and Ismilepur showed prevalence of this new cytological variant in sympatric association with species T and U. A total absence of inversion heterozygotes between them suggests the possible existence of a new species in *An. fluviatilis* complex. It is noteworthy to mention that PHC Laksar contributes bulk of malaria cases in the district with *An. fluviatilis* in highest proportion (36%) among 13 anopheline species recorded from this area. Therefore, studies have been initiated to resolve the taxonomic status of the new cytological variant and explore its role in malaria transmission.

Efforts were made to establish cyclic colonies of species S and U. Short-term cultures of species U could be established and bidirectional reciprocal crosses were made between species T and U. The hatchability ranged from 70–83%. In both the crosses the F_1 hybrid females were found with normal reproductive organs but the hybrid males were sterile with testis devoid of sperms. These observations showed that apart from pre-mating barriers there exist post-zygotic barriers between species T and U.

Molecular Characterization of Members of *An. fluviatilis* Complex—Development of Species-specific Markers and Microsatellite Markers (ICMR-Genomics Project)

Validation of diagnostic PCR assay for the identification of members: In the year 2001, a PCR assay was developed for the differentiation of members of *An. fluviatilis* complex, which is based on the sequences of D3 domain of 28S rDNA. The PCR assay was tested against over 700 mosquitoes collected from different parts of India (Districts–Hardwar, Udham Singh Nagar, Sundargarh, Malkangiri, Koraput, Gulbarga and Mandya) having different sympatric associations. Among these 147 specimens were also examined chromosomally for the validation of PCR assay. The PCR assay was found to differentiate unambiguously all the members of the complex and the results of PCR assay were in agreement with that of cytotaxonomy in the areas where fixed diagnostic inversions are present. In District Mandya of Karnataka, where q^1 inversion polymorphism exists—heterozygotes ($q^1/+^{q1}$) were found in Hardy-Weinberg equilibrium, all specimens were identified as species T. In the absence of chromosomal marker, this population was considered as species T due to resemblance with species T in biological characteristics—zoophagy and resting in cattlesheds.

Anopheles minimus Complex

Molecular Characterization of *An. minimus* from Assam

Based on 28S-D3 rDNA sequences of *An. minimus* species A and C, primers were designed for the differentiation of species A and C. The primers tested against *An. minimus* were collected from Districts Dibrugarh and Sonapur of Assam. Out of 20 samples tested, all were

identified as species A. To confirm the result, two samples of each population mentioned above were sequenced. The sequenced data confirmed that these mosquitoes were indeed species A.

Mapping of Indian Anophelines (Funded under ICMR Task Force Project on GIS and RS)

GIS Based Distribution of *An. culicifacies* in India

An. culicifacies is widely distributed throughout the country except in Andaman and Nicobar Islands. It is abundant in the plains but less prevalent in eastern part of India. Though the species is of the plains but also reported from higher altitudes—Nainital (1600 m), Kashmir (3000 m) etc. Therefore, from the altitude layer 0–3000 m was selected as the base layer and the most favourable range was taken as 0–1350 m with temperature > 20°C. Rainfall is an important factor in regard to *An. culicifacies*, the species being monsoon-associated and the maximum breeding occurs after the rains. High rainfall areas ≥ 2400 mm and very low rainfall areas ≤ 200 mm were deleted. Maps were integrated and the resulting map is shown in Fig. 4.

Blowup of distribution of *An. culicifacies* in northeastern India is shown in Fig. 5. The dots in the districts indicate the reported distribution of the species. In all districts where the species has been reported, GIS also predicts favourable areas for its distribution.

GIS Mapping of *An. minimus*

The resultant map after integration of thematic maps—soil, forest cover, rainfall, temperature and altitude using GIS mapped the areas favourable for *An. minimus.* Surveys were conducted to validate the results in favourable and unfavourable areas by our team as well as by an independent team and sites were selected both from reported and nonreported areas. Surveys were conducted in four states—Uttaranchal in north, West Bengal, Assam and Meghalaya in northeast. In the northeast a stretch of 900 kms was covered. Amazingly, *An. minimus* were collected from all locations in GIS predicted favourable zone (Table 1). In two districts—Champawat, Banbasa areas of Uttaranchal and Dhubri of Assam, in the former, the species was reported to have disappeared after 1950s, and latter, it was not reported in earlier entomological surveys, during present surveys in both the places *An. minimus* was encountred (Malaria Research Centre—Annual Reports). Thus besides validation of GIS prediction, reappearance of *An. minimus* at Banbasa and first report from Dhubri was established.

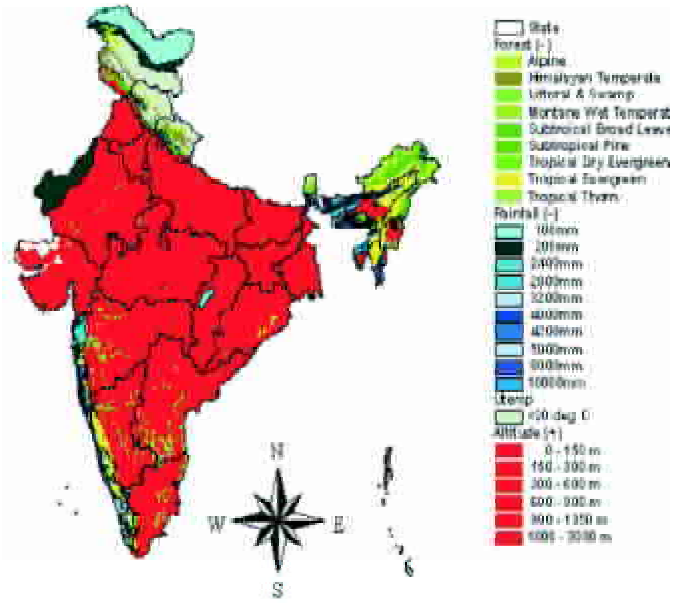

Fig.4: GIS predicted distribution of *An. culicifacies* subject to condition, the area being an rural/urban area

GIS predicted precisely the locations in these districts to conduct entomological surveys and the species could be found there. Blind survey was conducted by independent team in both favourable and unfavourable areas. In favourable areas the species was found and in unfavourable areas on the border of Karbi-Anglong, it was found to be absent.

Using GIS, percentage of favourable area for distribution of *An. minimus* in different states was estimated. It showed that most of the area in northeastern states was favourable for *An. minimus*. In Mizoram favourable area is about 90.61%, and in Manipur, Nagaland, Tripura and Assam 70, 35, 33 and 25% respectively. In other states it was less than 10% except Kerala. There are some favourable areas in Kerala and Maharashtra, till date there is no report of *An. minimus* from these areas. Most favourable corridors for distribution of *An. minimus* were also mapped by stratifying favourable

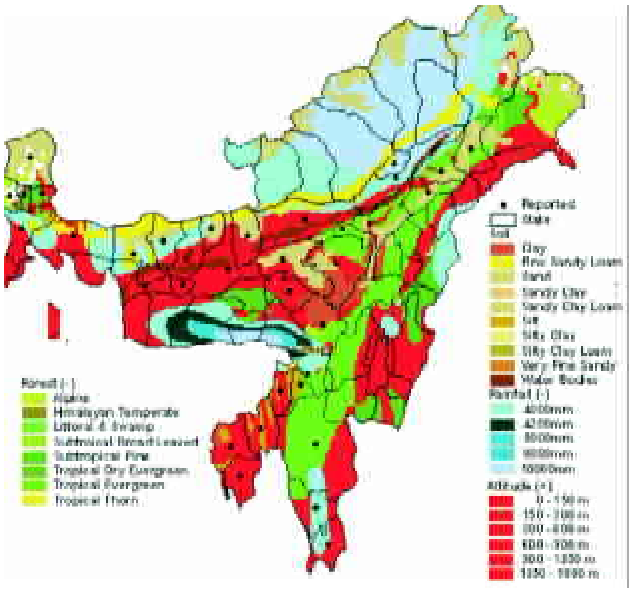

Fig.5: Blowup of distribution of *An. culicifacies* in north-eastern India, the dots represent the districts, where *An. culicifacies* was reported. Red colour shows areas favourable for distribution in that particular district

Table 1. Validation of *An. minimus* in GIS predicted areas by precision surveys

Collection site/ District/State	Period of survey	MHD* of *An. minimus*	Larval density**	Remarks
Banbasa, Uttaranchal	May 2001	0.25	0.00	Reappearance of species
	Jul 2001	0.53	0.02	
	Aug 2001	0.73	0.03	
Jalpaiguri, West Bengal	Oct 2001	1.70	0.08	
Dhubri, Assam	Oct 2001	0.91	0.06	Reported first time
Kamrup, Assam	Oct 2001	21.80	1.40	
Barpeta, Assam	Oct 2001	Not done	0.18	
Burnihat, Meghalaya	Oct 2001	1.16	Not done	
Shillong, Meghalaya	Oct 2001	0.33	Not done	
Darrang, Assam	Jun/Jul 2001	4.00		
Goalpara, Assam	Sep 2001	21.00		
Karbi-Anglong, Assam	Oct 2002	0.00		*An. minimus* was not found in GIS predicted unfavourable area

*MHD: No. of mosquitoes collected per man per hour; **Larval density—No. of larvae per dip.

Table 2. GIS predicted most favourable corridors for *An. minimus*

Category	Altitude (m)	Rainfall (mm)	Temp. (°C)	Forest cover
Category 1 (Most favourable)	0–600	2000–2800	22.5–25	Evergreen
Category 2 (Medium favourable)	600–900	2800–3200	20–22.5	Moist deciduous
Category 3 (Less favourable)	900–1800	3200–4000	< 20	Moist deciduous

The species population is likely to be most stable in category 1 and least in category 3.

zones in high, medium and low categories. The stratification was based on forest cover, temperature, rainfall and altitude, range identified for three zones is given in Table 2.

An. minimus in Banbasa (Uttaranchal)

To validate the GIS prediction, *An. minimus* were collected from Banbasa area of Uttaranchal state though it has been reported to have disappeared from this area in early 50's. The surveys were conducted in post-monsoon (October and November 2001), pre-monsoon (March, April and May 2002) and monsoon (June-July 2002) seasons, per man hour density of the species was recorded as 0.9, 0.26 and 0.67 and the larval density per dip as 0.5, 0.2 and 0.4 respectively. The larvae collected from the streams were reared individually in the laboratory till adult emergence. Individual specimens of larval and pupal skins were mounted and examined for the identification characters. The most important character of *An. minimus* larvae is the presence of branched seta-0 on abdominal segments III–VI, and this character was recorded in 22 mounted slides prepared in different seasons, which confirms the presence of *An. minimus* in Banbasa, a GIS predicted area.

GIS Based Distribution of *An. stephensi* —An Urban Vector

An. stephensi is found throughout the country except in Andaman and Nicobar Islands and higher altitudes. It is responsible for transmission of malaria in urban/peri urban areas of the country but the recent epidemics of malaria in Jodhpur and Bikaner districts of Rajasthan were attributed to this species. *An. stephensi* breeds in domestic containers such as tanks, cisterns, coolers, wells, etc. It has also been found breeding in irriga-tion channels and in rice fields. Distribution of *An. stephensi* is shown in Fig. 6, subject to the condition, the area is urban/peri urban. Altitude range from 0–900 m, temparature >20°C and rainfall < 2400 mm have been taken as favourable.

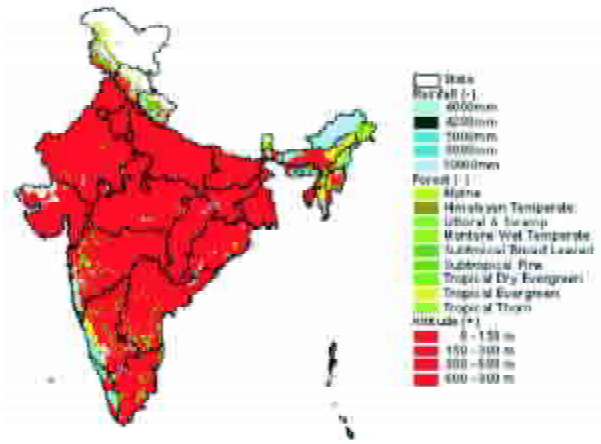

Fig. 6: GIS predicted distribution of *An. stephensi*, subject to the condition, the area being an urban/peri urban area

Besides mapping distribution of major malaria vectors, all 58 anophelines have been mapped using GIS and a CD has been produced consisting of an album with the objective to make ready to use product. This album consists of 58 maps each

showing the GIS predicted distribution of anopheline species in India, along with the blowup maps of GIS predicted district-wise favourable areas and the validation of GIS predicted distribution through reported surveys (Fig. 7).

The technique can delineate the areas favourable for any species of flora and fauna, which is very useful for precision surveys. The technique is fast and can be easily duplicated at desired scale in other parts of the country/world. Since this unique technique identifies the location for precision surveys in an area where the species is likely to be found, thus location wise control activity may be implemented which may be effective in terms of results and cost.

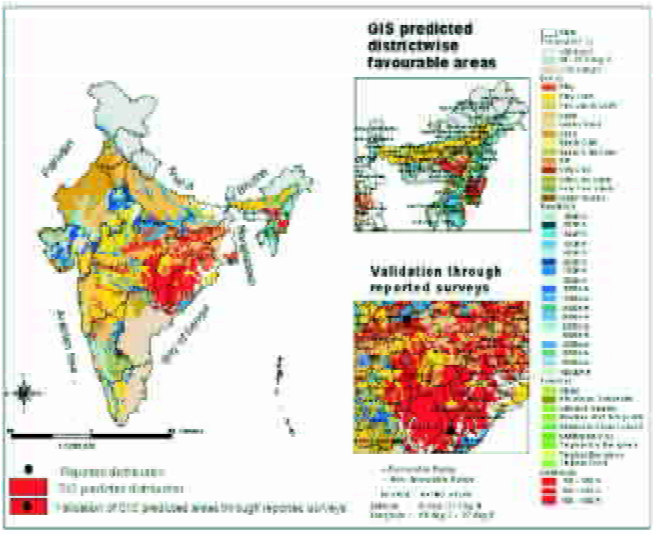

Fig. 7: GIS predicted distribution of *An. sergentii* in India, a blowup showing northeastern states and validation through reported distribution is also depicted in insets

Spiracular Index and Bioecology of *An. minimus* in Kamrup District, Assam

A total of 1811 *An. minimus* were collected from 12 villages of Kamrup district, Assam, located in evergreen-forested zone in pre-monsoon (April–May 2002) and post-monsoon (September–October 2001) season. Out of 1811 *An. minimus*, 665 (MHD 15.7) were collected in pre-monsoon and 1146 (21.8 MHD) in post-monsoon season. During pre-monsoon and post-monsoon 410 and 310 specimens were dissected respectively for spiracular index. In pre-monsoon, the average spiracular length of *An. minimus* was 0.085 mm (± 0.01), average thorax length was 0.94 mm (± 0.05) and spiracular index was 9.04 (± 0.74) whereas in post-monsoon average spiracular length was 0.08 mm (± 0.01), average thorax length was 0.94 mm (± 0.07) and spiracular index was 9.41 (± 0.70) as calculated by using the technique and formula of Vinogradaskaya 1969. *An. minimus* population does not show any seasonal fluctuation in their spiracular index ($p < 0.05$). In pre-monsoon the humidity recorded was 75–81% whereas in post-monsoon the humidity was 72–95%.

It may be noted that *An. minimus* is a small sized mosquito of wet zone as it measures 2–3 mm as compared to *An. stephensi* type form 5–6 mm and *mysorensis* 4–5 mm of arid zone—the xerophilic species. The spiracular index of *An. minimus* was higher 9.4 as compared to that of *An. stephensi* type form 9.09 and *mysorensis* 7.69. This establishes that *An. minimus* is a hygrophilic species.

Bioecology

Breeding habitat: *An. minimus* was found breeding in slow moving streams with grassy margins. A total of 575 larvae were collected and identified. Out of these 235 were identified

as *An. minimus* and others were associated species—*An. splendidus, An. jeyporiensis* and *An. maculatus*. It is noteworthy to mention that the breeding was observed in those streams which were near to the houses (< 1000 m). Fifteen *An. minimus* emerged from the larvae collected from rice fields and small irrigation channels adjacent to the houses.

Resting collection: Searches made inside the houses and outdoor shelters yielded 665 specimens of the species. Out of these 640 (96.24%) accounted for indoor collection during pre-monsoon season (April–May). Similarly, during post-monsoon collection (September–October) out of 580 total collected 565 (97.41%) were from indoors. The preferred indoor resting sites included under surface of the furniture, mosquito net on the bed, hanging clothes, cobwebs and walls. The most preferable resting site was under surface of the furniture in both the seasons—pre-monsoon and post-monsoon as 290 (60%) and 245 (55%) specimens were collected respectively.

Indoor Resting Behaviour of *An. stephensi* in an Arid Zone (District Jodhpur, Rajasthan)

Success of malaria control by IRS largely depends on resting behaviour of the vector mosquitoes on sprayable surfaces. Endophilic (indoor resting) and endophagic (indoor feeding) species are known to be highly vulnerable to this strategy. The success rate with exophilic (outdoor resting) and endophagic species depends on the time spent by the species on sprayable surfaces during pre- and post-biting rest. Therefore, studies on resting behaviour of *An. stephensi* were carried out to assess its amenability to control through indoor residual spray in an Arid zone of District Jodhpur, Rajasthan during March–April 2002. In the present study, resting behaviour of the species during all its movement rhythms covering 24 hours period related to: (i) swarming/mating; (ii) pre- and post-biting rest; (iii) after feed resting between hopping movements; (iv) night and daytime resting; and finally (v) diel activity movements in response to temperature changes were carried out in both unsprayed and spayed villages. Analysis revealed that about 95 and 97% of *An. stephensi* preferred to rest on unsprayable surfaces—household objects in unspyayed and spayed villages. The most preferred resting sites in both groups of villages under household objects were hanging clothes, utensils and cupboards. There was no significant difference in resting behaviour of the species in both groups of villages ($p>0.05$). Pre- and post-biting rest period ranged from 5–15 and 15–25 min respectively. After biting out door, species starts entering the rooms at around 2330 hrs. Maximum entries—56 and 62% of the species into the rooms were observed during third quarter (0100–0400 hrs) in unsparayed and sprayed villages respectively, and coincided with fall in ambient temperature indoor below 30^{O}C. Statistically there was no significant difference in the entry of mosquitoes ($p > 0.05$) in both the groups of villages. Therefore, control of *An. stephensi* in study area requires an integrated control strategy based upon intersectoral, environmental, larviciding with chemical/biolarvicide and use of larvivorous fish wherever feasible. Such a control strategy offers cost-effective and sustainable option than indoor residual spray.

VECTOR–PARASITE INTERACTION

Cyclical Transmission of Rodent Malaria

Laboratory cyclical transmission of rodent malaria parasites—*P. yoelii yoelii, P. chabaudi, P. berghei* and *P. vinckei* was revived in BALB/c mice using *An. stephensi* as vector. Cyclical transmission of *P. vinckei* was also maintained through other vectors—*An. culicifacies* species A and *An. fluviatilis* species T.

Immune Response of *An. stephensi* against *Micrococcus leutus*

Insects are known to mount a strong immune response against any invading parasite. Success of sporogony largely depends upon the ability of insect to combat the parasite and/or the ability of parasite to evade the immune response. Response of *Anopheles stephensi*, an urban malaria vector to bacterial infection and to sterile injury is studied in immature stages. Proteins being the first conceivable product of gene action are studied using SDS-PAGE as tool. Denaturating SDS-PAGE analysis revealed induction of a 42 kDa polypeptide in female pupae of urban malaria vector— *An. stephensi* upon infection with gram-positive bacteria *Micrococcus leutus*, that was absent upon septic injury. Septic injury seems to activate phenoloxidase cascade, as melanin formation is responsible for wound healing in insects. Our study identifies up-regulation of 35 and 65 kDa proteins in larvae and pupae both in whole body and tissue specific expression. It is worth mentioning here that serine proteases have molecular weight of 35 kDa whereas, phenoloxidases have molecular weight in the range of 60–70 kDa. Besides these five, polypeptides (20, 45, 52, 70 and 150 kDa) are also enhanced in their expression upon injury. Similary, studies on the immunity of refractory and susceptible strains of *An. culicifacies* in response to rodent malaria parasite and bacteria are in progress.

INSECTICIDE RESISTANCE

Present Status of Insecticide Resistance in *An. stephensi* from Delhi

Susceptibility tests against larvae and adult mosquitoes of *An. stephensi* strain collected from southDelhi revealed partial resistance in adult mos-quitoes to DDT and a very high degree of resistance to malathion and dieldrin, however, the mosquitoes were fully susceptible to other insecticides (Table 3). Among the larvicides tested, this strain was found to be highly resistant to fenthion but it was fully susceptible to teme-

Table 3. Susceptibility of *An. stephensi* strain from south Delhi to insecticides

Insecticide	% mortality after 24 h	Time in min	
		LT_{50}	LT_{90}
DDT (4%)	62.5	50.6	114.2
Malathion (5%)	5	6.54**	10.63**
Fenitrothion (1%)	79 (100)*	23.93	63.32
Propoxur (0.1%)	100	20.8	59.5
Deltamethrin (0.05%)	100	2.36	9.76
Lambdacyhalothrin (0.05%)	100	3.33	16.22
Cyfluthrin (0.15%)	100	4.11	14.17
Etofenprox (0.1%)	100	10.73	24.4

*Exposure time 2 h; ** Time in hours.

Table 4. Larval susceptibility of *An. stephensi* from south Delhi to some larvicides

Larvicides	LC_{50} (ppm)	LC_{90}
Temephos	0.0098	0.0199
Fenthion	>0.125	>0.125

phos (Table 4). Further, studies will be carried to monitor present status of susceptibility to various insecticides in the field populations of *An. stephensi* from Rajasthan; and resistance mechanism and inheritance pattern of resistant gene in this strain.

A New Focus of Malathion-resistant *An. culicifacies in* District Chhindwara, M.P.

Surveys were carried out in the villages of Mohkher block in District Chhindwara (Madhya Pradesh) in November and December 2001; and March 2002 and in the villages in Pandhurna block on the Madhya Pradesh-Maharashtra border in December 2001 and March 2002. In insecticide susceptibility tests the population was resistant to DDT (36–84%). To malathion Mohkher population was 14–40% resistant while in Pandhurna the resistance was in the range of 36–63%. Mohkher block was under regular indoor sprays of DDT while in Pandhurna block indoor spray in public health programme was discontinued about a decade ago. In both the areas malathion was never sprayed in public health programme. The observed resistance to malathion in *An. culicifacies* could be attributed to selection by the pesticides being used in agriculture on cash crops—cotton, vegetables, oil seeds, etc. Serial synergist-insecticide bioassays on the field populations indicated the involvement of malathion carboxylesterase as the major mechanism of malathion resistance though the involvement of mixed function oxidases as a secondary mechanism could not be ruled out.

VECTOR CONTROL

Follow-up Studies on the Impact of Indoor Residual Spraying of Bendiocarb 80% W.P. (Carbamate) against DDT and HCH Resistant Malaria Vector—*An. culicifacies* in Malaria Endemic Villages of Distt. Ghaziabad (U.P.) [Contract Research Project with M/s. Hoechst]

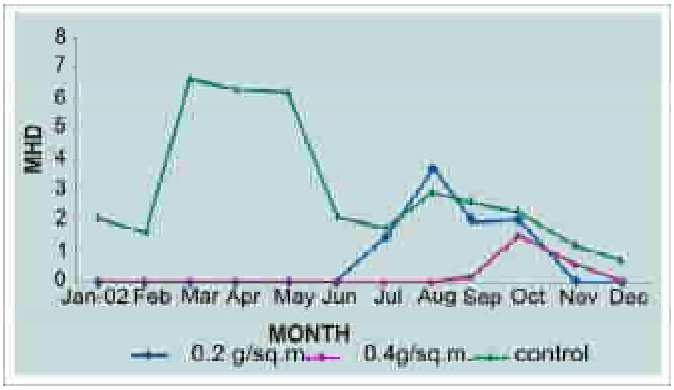

Fig. 8: Man hour density of *An. culicifacies* in selected villages of Distt. Ghaziabad, U.P.

Follow-up studies were carried out during the year to study the efficacy of bendiocarb in interrupting the malaria transmission. The SPR in village sprayed with bendiocarb @ 0.2 g/m^2 ranged from 0 to 6.4, in 0.4 g/m^2 bendiocarb sprayed village the SPR ranged from 0 to 4.5 where as in the control village the SPR ranged from 0 to 44.4. The SfR was nil in both the sprayed villages whereas in the control village few cases were reported. The study indicates that bendiocarb spraying has resulted in interruption of malaria transmission in the experimental villages when compared to the control village. Ento-

mological studies revealed that MHD of *An. culicifacies* was found to be in the range of 1.5–3.7 during the transmission season (July to October) in 0.2 g/m^2 bendiocarb sprayed village and in 0.4 g/m^2 bendiocarb sprayed village the MHD of *An. culicifacies* was in the range of 0–1.5 whereas in the control village it was in the range of 1.7–3.0 in the same period (Fig. 8). This clearly sug-gests that bendiocarb residual spraying is highly effective in controlling the mosquito densities.

Laboratory and Field Evaluation of Teknar HP-D (*B. thuringiensis* var *israelensis*) [Contract Research Project with M/s. Margo Bio-controls Pvt. Ltd.]

Efforts have been initiated to evaluate the bioefficacy of *Bacillus thuringiensis* against target species of mosquitoes. Preliminary laboratory results revealed that *Bti* formulation has broad-spectrum larvicidal activity against mosquitoes. Cent per cent mortality in *Cx. quinquefasciatus, An. stephensi* and *Ae. aegypti* was observed in laboratory trials with Teknar @ 0.1 ppm. Field trials are in progress.

Field Evaluation of Mosquito Larvicide and Pupicide Agnique MMF in different Urban Habitats against Malaria Vector, *An. stephensi* [Contract Research Project under WHOPES]

Evaluation of Agnique MMF—Poly (Oxy-1, 2-ethanediyl), a-isoctane cyl-w-hydroxy1, a mono-molecular surface film, developed by M/s. Cognis Corporation was carried out in the representative breeding habitats of *An. stephensi* in urban areas of National Capital Region, Delhi. Study was carried out in two parts—simulated field conditions and natural field conditions.

In simulated field testing cement tanks of 1 x 1 x 1 m size were used and the efficacy of larvicide, effective doses and duration of effect against *An. stephensi* were determined. The impact of larvicide applied in different doses was studied by monitoring number of adults emerged in specially designed mosquito traps. The efficacy of the test larvicide was assessed by measuring the larval density in water storage cemented tanks and wells in natural field conditions. Results showed that in simulated field testing the Agnique MMF caused 100% inhibition of emergence of *An. stephensi* adults from water storage tanks upto one week at the application rate of 0.4, 0.6 and 1 ml/m^2 and > 95% up to second week at all the three doses. Dose of 1 ml/m^2 was effective up to third week when 99.5% inhibition of adult emergence was achieved.

In natural field trials the results of Agnique MMF in water storage tanks with breeding of *An. stephensi* and *An. subpictus*, the dose of 2 ml/m^2 was effective. The per cent reduction in late instars was more than 75 on Day 4 and 100% control was achieved in one week and remained up to second week. Results with MLO when treated @ 20 ml/m^2 also gave the same results. Hence, the results clearly indicated that dose of 2 ml/m^2 of Agnique MMF can be selected for the effective control up to two weeks period.

In wells the dose of 1 ml/m^2 gave about 75% control of late instars of *An. stephensi* on Day 2 and remained 100% up to two weeks. Agnique MMF was safe against larvivorous fish (*Gambusia affinis*) and notonectid bug (*Anisops sardae*). Since the Agnique MMF is odor-

less, invisible, monomolecular film and spreads rapidly across standing water, it could be one of the choices in the larval control programmes in urban areas.

Field Evaluation of Insect Growth Regulator—Triflumuron against Larvae of Mosquito Vectors [Contract Research Project with M/s. BAYER (India) Ltd.]

Efficacy of triflumuron, an IGR compound was carried out in the representative breeding habitats of *An. culicifacies* and *Cx. quinquefasciatus* in rural areas of north Delhi and in the district Sonepat of Haryana state. In small-scale fields trials triflumuron was sprayed at doses of 0.25, 0.5 and 1 ppm. Habitats selected for *An. culicifacies* were pools, ditches and paddy fields. For *Cx. quinquefasciatus* habitats selected were polluted drains, pools and cement tanks. Results showed the reduction in immatures in all the doses tested. At the dose of 1 ppm the pupal production was nil after one week which remained so even up to four weeks. The occurrence of delayed mortality in larvae indicated the effective developmental inhibition potential of this compound.

Evaluation of the Impact of DDT and Malathion Indoor Residual Spraying being used in Malaria and Kala-azar Control Programmes on the Disease Prevalence—A Multicentric Study

A nine-month multicentric study was conducted (July 2001 to March 2002) involving two other ICMR institutes—VCRC, Pondicherry and RMRIMS, Patna, Bihar. A common protocol was prepared for both entomological and parasitological evaluations. MRC evaluated the efficacy of indoor residual spray of DDT in six districts —Bareilly (Uttar Pradesh), Mandya (Karnataka), Chhindwara (Madhya Pradesh) and Kamrup (Assam) and of malathion in two districts—Kheda (Gujarat) and Hardwar (Uttaranchal). Indoor residual spray was conducted under the supervision of MRC staff to achieve the desired coverage. Evaluation was carried out during pre- and post-spray periods on different aspects—vector abundance, species composition, susceptibility status of mosquitoes to insecticides, disease prevalence, etc.

Results of the evaluation indicated effectiveness of DDT sprays against *An. minimus*, a principle vector in northeast states. *An. culicifacies* was found resistant to DDT in susceptibility tests in all the areas of study and DDT spraying was not found very effective in controlling this species. However, in some study areas this species exhibited excito-repellency against DDT for few weeks after spray registering a decrease in vector density while the results of evaluation against malathion indicated effectiveness of malathion sprays in controlling the vector—*An. culicifacies* in District Hardwar and in District Kheda the species exhibited differential susceptibility in different areas.

Laboratory (Phase-I) Evaluation of Phenthoate against Urban Mosquito Vectors—*Anopheles stephensi* and *Culex quinquefasciatus* [Contract Research Project with M/s. EID Parry India Ltd., Chennai]

A new organophosphorus insecticide phenthoate was tested against urban malaria vector *An. stephensi* and pest mosquito *Culex quinquefasciatus* in the laboratory to determine its efficacy against larvae and adult mosquitoes and to compare the results of its bioefficacy with that of

malathion, another organophosphorus insecticide which is being used at present as an adulticide. Mosquito colonies—*An. stephensi* and *Cx. quinquefasciatus* maintained in the insectary of MRC were used. The efficacy of phenthoate was determined against larvae and adult mosquitoes as per WHO recommended procedure. The results of bioefficacy against phenthoate are given in Tables 5–8. Phenthoate was more effective against *Cx. quinquefasciatus* larvae (LC_{50} = 0.0217 ppm) than *An. stephensi* (LC_{50}= 0.368 ppm). However, against adult mosquitoes of *An. stephensi*, phenthoate did not produce any mortality, when exposed to insecticide impregnated papers at a concentration of 4%, whereas against *Cx. quinquefasciatus*, complete mortality was observed even at 2% concentration (LT_{50}= 11.9 min).When the sus-

Table 5. Efficacy of phenthoate against III instar larvae of *An. stephensi* and *Cx. quinquefasciatus*

Mosquito species	Concentration in ppm		?²(df)
	LC_{50} (95% confidence limit)	Regression equation	
An. stephensi	0.368 (0.248–0.453)	Y = 3.129X ± 6.3579	0.755 (2)
Cx. quinquefasciatus	0.0217 (0.0132–0.0302)	Y = 2.0671X ± 5.7486	4.337 (3)

Table 6. Efficacy of phenthoate against adult mosquitoes of *An. stephensi* and *Cx. quinquefasciatus*

Mosquito species	% concentration		?² (df)
	LC_{50} (95% confidence limit)	Regression equation	
An. stephensi	> 4%	—	—
Cx. quinquefasciatus	0.33% (0.108–0.508)	Y = 2.2495X ± 6.08	1.376 (2)

Table 7. Efficacy of phenthoate 2% (lethal time) against *Cx. quinquefasciatus*

Mosquito species	LT_{50} (Time in min)	Regression equation	?² (df)
Cx. quinquefasciatus	11.98 (7.045–16.78)	Y = 2.1686X ± 2.66	2.239 (2)

Table 8. Comparative susceptibility of adult mosquitoes of four strains of *An. stephensi* against malathion and phenthoate

An. stephensi strain	Per cent mortality	
	Malathion 5%	Phenthoate 4%
Wild strain, Delhi	18	6.6
Mutant strain (Black larvae)	50	66.6
Mutant strain (Golden yellow larvae)	70	80
Mutant strain (White eye larvae)	86.6	100

ceptibility of *An. stephensi* to phenthoate was compared with that of to malathion, it showed cross-resistance to both the insecticides.

Prospecting of Botanical Pesticides (Screening of Bioactivity of Plant Extracts against Mosquitoes Particularly *Anopheles* spp) [DBT Funded Collaborative Project]

This study was undertaken as a part of a collaborative project funded by DBT included various laboratories as shown in Table 9. Bioactivity of various plant extracts/fractions/formulations received from five extracting laboratories was determined against mosquitoes particularly the malaria vector *An. stephensi* using standard protocol which included larvicidal, adulticidal and IGR activities, oviposition deterrency and mosquito repellency. During the reporting period 107 samples were received of which 50 samples have been screened for different activities as mentioned above. Of these 16 samples showed positive results for larvicidal activity and 8 samples showed adultcidal activity (Table 10).

Larvicidal Properties of Crude Aqueous Extract of Leaf of a Coded Plant – SP2

Larvicidal efficacy of the crude aqueous extract of the leaf of a coded plant – SP2 was tested against five species of mosquitoes of three genera—*An. culicifacies* species A and C, *Cx. quinquefasciatus*, *An. stephensi* and *Ae. aegypt*i. The calculated lethal concentration for 50% mortality (LC_{50}) of the exposed larvae was in the range of 0.25 to 0.59 ml/L. The efficacy was in the order *Cx. quinquefasciatus* (0.25ml/L) > *An. culicifacies* species A (0.26) >*An. stephensi* (0.33) > *Ae. aegypti* (0.36) > *An. culicifacies* species C (0.59).

Table 9. Activities in various laboratories

Activity	Responsible laboratories
Collection, preservation and taxonomic identification of plants	RRL, Jammu RRL, Trivandrum NBRI, Bangalore
Extraction and fractionation of herbal products	IIT, Delhi FRI, Dehradun EID, Parry (India) Ltd., Bangalore RRL, Trivandrum RRL, Jammu
Bioassay testing of the efficacy against agricultural pests	IHBT, Palampur EID, Parry (India) Ltd., Bangalore
Bioassay testing of the efficacy against mosquitoes *(An. stephensi)*	MRC, Delhi and Hardwar
Formulation of the selected samples	IIT, Delhi IPFT, Gurgaon

Table 10. Bioactivity of plant extracts against *An. stephensi*

S. No.	Sample code number	Bioactivity of plant extract		
		Larvicidal	Adulticidal	Repellency
1.	NBDB(4)-002B-07-P-13a	(+)	(–)	(–)
2.	NBDB(4)022B-08-P-10a	(+)	(–)	(–)
3.	NBDB(4)023B-07–P-10a	(+)	(–)	(–)
4.	NBDB(4)042B-07-P-10a	(+)	(–)	(–)
5.	NBDB(2)008A-06-P-01a	(+)	(+)	(+)
6.	NBDB(2)010A-06-P-10a	(+)	(–)	(–)
7.	NBDB(2)010A-06-P-10b	(+)	(–)	(–)
8.	NBDB(4)-001B-08-P-13a	(+)	(–)	(–)
9.	NBDB(3)-022i-08-P-11e1	(+)	(–)	(–)
10.	NBDB(2)-005A-07-P-10a	(+)	(+)	(–)
11.	NBDB(2)-005A-07-P-10b	(+)	(+)	(–)
12.	NBDB(2)-005A-07-P-10c	(+)	(+)	(–)
13.	NBDB(2)-005A-07-P-04a	(+)	(+)	(–)
14.	NBDB(2)-005A-07-P-04b	(+)	(+)	(–)
15.	NBDB(2)-005A-07-P-04c	(+)	(+)	(–)
16.	NBDB(2)-055D-11-P02oil	(+)	(+)	(–)

OTHER STUDIES

Operational Evaluation of the Stability of Iodine in Double Fortified Salt – A Multicentric Study (ICMR – NIN – MRC – RMRC, Dibrugarh – RMRC, Bhubaneswar–TRC – IRR)

The stability of iodine in double fortified and iodised salt during storage for a period of one year under programmatic condition was determined as a part of a multicentric study under ICMR. Four types of coded salt samples (one thousand kilogram each, 25 kg x 40 bags) double fortified with iron and iodine (DFS) and fortified with iodine alone (IS) in the two forms—refined common salt (RCS) and common salt (CS), produced from the factory were received at MRC in October 2001 and stored at three places— inside room, in verandah and outside conditions as per protocol. Within a month of the date of arrival of the salt samples at MRC, one packet from each bag was taken out randomly and repacked in three packets of 100 g each after remixing. One of these samples was sent to NIN and second sample was stored at MRC. The third sample was analyzed for iodine content at MRC laboratory. The same process was repeated after 3 months, 6 months, 9 months and 12 months of the date of production of salt samples. Results of iodine content in the coded salt samples are given in Table 11. It was observed that the iodine content in the salt samples with yellow and green colour packing remained stable over a period of one year even

Table 11. Summarized results of iodine estimation in different types of salt samples RCS/ CS/ DFS/ IS* stored under different conditions over a period of one year at Malaria Research Centre

S. No.	Mean ± S.D.				
	Initial stage (0)	3 Months	6 Months	9 Months	12 Months
Y-1 to 15	44.01 ± 11.83	46.7 ± 9.48	37.21 ± 15.73	41.53 ± 11.87	41.5 ± 11.4
Y-16 to 20	43.92 ± 12.08	41.05 ± 10.11	35.01 ± 11.87	35.56 ± 11.28	39.48 ± 12.8
Y-21 to 40	51.69 ± 9.68	47.95 ± 8.36	45.10 ± 10.24	40.07 ± 10.16	40.6 ± 7.1
B-1 to 15	23.61 ± 10.54	26.06 ± 11.33	20.77 ± 8.31	20.06 ± 5.25	13.8 ± 6.6
B-16 to 20	18.52 ± 5.27	15.63 ± 4.79	15.33 ± 8.96	17.99 ± 3.71	10.57 ± 6.2
B-21 to 40	22.60 ± 11.15	25.59 ± 11.12	19.43 ± 6.07	18.10 + 5.62	14.75 ± 7.1
R-1 to 15	3.87 ± 0.62	4.85 ± 1.63	4.93 ± 0.92	7.64 ± 3.00	7.93 ± 1.8
R-16 to 20	2.66 ± 1.43	5.49 ± 1.18	3.71 ± 0.73	5.48 ± 0.82	7.73 ± 1.5
R-21 to 40	2.43 ± 1.47	4.82 ± 1.18	3.77 ± 1.16	6.71 ± 1.64	7.8 ± 1.6
G-1 to 15	50.01 ± 4.36	52.51 ± 8.10	48.24 ± 8.73	41.31 ± 9.82	45.2 ± 5.7
G-16 to 20	44.04 ± 2.50	48.04 ± 3.42	47.18 ± 8.53	46.24 ± 6.89	43.49 ± 4.88
G-21 to 40	47.66 ± 7.23	46.72 ± 8.25	44.69 ± 6.31	45.60 ± 4.46	43.17 ± 6.16

*RCS—Refined common salt; CS— Common salt; DFS— Double fortified salt; IS— Iodized salt.
Note: Sample No. 1 to 15 were stored in verandah, 16 to 20 stored in outdoor condition and 21 to 40 stored in a room/godown. Y—yellow; R— red; B—blue and G—green.

under different storage conditions, while in red and blue colour packing the iodine contents were not stable. The samples in red colour packing degraded very fast, even at the initial stage the contents were almost completely degraded. These results will be correlated with different colour code of iodised/double fortified salt.

PARASITE BIOLOGY

Studies on Drug Susceptibility

Twenty isolates characterized for their chloroquine sensitivity status were revived from cryopreserved condition and cultivated *in vitro* to retest their CQ sensitivity status and two third of 20 samples showed variation in their chloroquine sensitivity status.

P. falciparum isolates (sensitive and resistant to chloroquine) were cloned and the clones were tested for their CQ sensitivity. These clones would be characterized for genotypes of MSP-1 and MSP-2.

Molecular Characterization of *P. falciparum* Isolates

Parasite Bank

P. falciparum isolates available in the parasite bank of the Centre have been analyzed for polymorphism of MSP-1, MSP-2 and GLURP using nested PCR with family-specific primers. In MSP-1 system, all the three families namely K-1, MAD20 and R033 were observed with prevalence of first two (K1 and MAD20). In MSP-2 system, both the families namely FC27 and 3D7 were observed with approximately similar prevalence. In addition to MSP-1 and MSP-2, GLURP system was standardized and isolates were analyzed. GLURP exhibited polymorphism with 11 different size fragments. Most of the isolates were observed to be multiclonal. The results of family grouping analysis of MSP-1 and 2 of *P. falciparum* isolates are shown in Fig. 1.

HIGHLIGHTS

- Parasite bank has well characterized *P. falciparum* isolates
- Characterization of *P. falciparum* isolates in the Parasite Bank for MSP-1, MSP-2 and GLURP has revealed that each analyzed isolate has a different genotype.
- Genotyping of *P. falciparum* and *P. vivax* isolates from different regions of the country has shown higly polymorphic nature of the Indian isolates in respect of family grouping analysis of MSP-1 and MSP-2 in *P. falciparum* isolates and MSP-3? and GAM1 of *P. vivax*.
- Genotyping of recrudescence infection in five *P. falciparum* patients revealed different genotypes of MSP-1/2 in 3 patients on Day 14
- Inhibitory effect of Nitric Oxide on plasmepsin activity of *P. vivax* was observed suggesting to design strategies to selectively upregulate NO production.
- Analysis of blood samples from different endemic zones has shown high anti-GPL antibody in case of *P. falciparum* and *P. vivax* infections but not with normal or nonmalarial sera.
- HRP-II antigen studies has shown that antimalaria treatment neutralizes the antigen persistence by antibodies
- A multiplex PCR to differentiate *P. falciparum* and *P.vivax* in a single assay was standardized.

Field Isolates

Bloodspots of *P. falciparum* positive patients collected from the Jarawas tribe of Andaman and Car Nicobar islands were analyzed by nested PCR assay for MSP-1 and MSP-2 size variations in variable repeat regions. Results have shown presence of only one allele of each—450 bp for MSP-1 and 500 bp for MSP-2. Blood spots were further assayed using family specific primers. Results revealed presence of all the three families of MSP-1 with higher proportion of R033 family. In MSP-2, both the families were present with greater

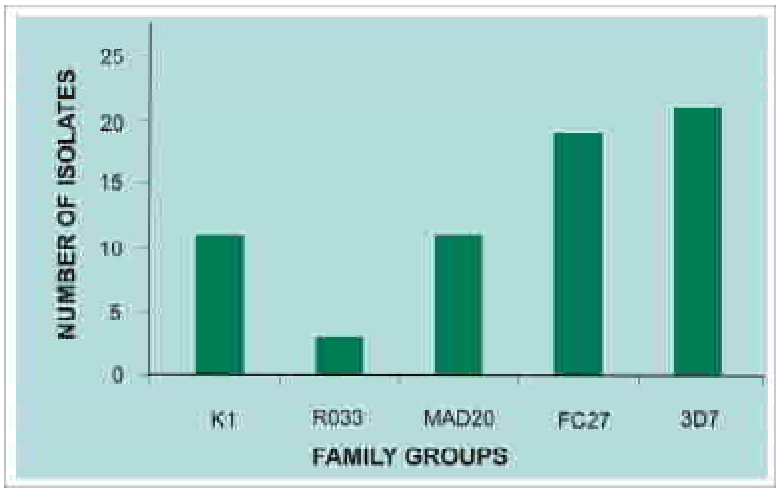

Fig.1: Distribution of family groups of MSP-1&2 of *P. falciparum* isolates of parasite bank

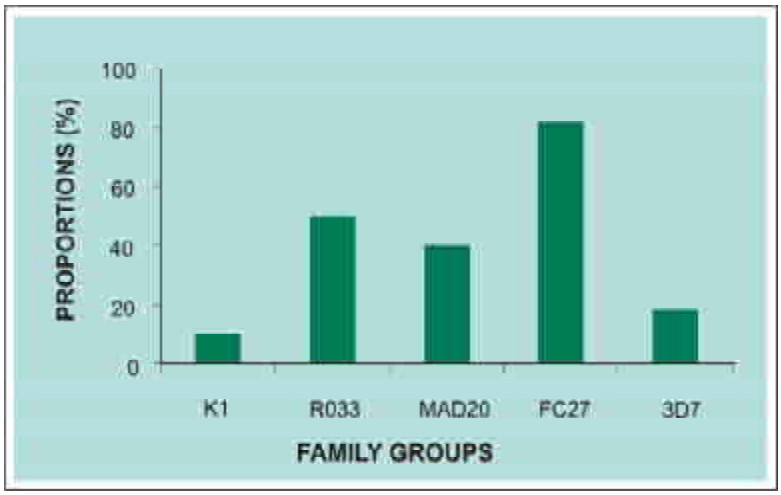

Fig. 2: Distribution of family groups of MSP-1&2 in *P. falciparum* isolates (Jarawas tribes of Andaman & Nicobar Islands)

proportion being that of FC27. The proportion of isolates with different families of MSP-1 and MSP-2 is shown in Fig. 2.

Genetic Diversity Studies of *P. falciparum* and *P. vivax* in India using Molecular Markers (ICMR Funded Project under Genomics)

P. falciparum

Field isolates collected from Assam and Orissa have been analyzed for polymorphism of MSP-1 & MSP-2. Results revealed polymorphism of both the systems among the isolates of the areas.

Twenty isolates of *P. falciparum* from Assam area have been analyzed for family grouping and observations revealed presence of all the three families of MSP-1 (K1, MAD20 and R033) and both of MSP-2 (FC27 and 3D7) (Table 1). Proportion prevalence of family-specific markers is 51% for MAD20, 31% for K1 and 17% for R033 of MSP-1 and 50% for FC27 and 3D7 of MSP-2. Out of twenty samples analyzed 50% (out of these 50% of isolates with all the three families of MSP-1) were multiclonal. Only two isolates were categorized as single clonal based on the genotyping of MSP-1 and MSP-2.

Twenty-two samples from Assam were also analyzed for allelic polymorphism using nested protocol. Among the isolates analyzed size variants of MSP-1 ranging from 400 to 600 bp were observed and in MSP-2 variants ranging between 400 and 750 bp were observed. Twenty-eight isolates of *P. falciparum* from District Sundargarh, Orissa, have been analyzed for MSP-1 and MSP-2 family grouping. All the three families of MSP-1 and two of MSP-2 were observed with a prevalence of 40% for K1, 38% for MAD20, 22% for R033 of MSP-1 and 58% for FC27 and 42% for 3D7 of MSP-2 (Table 1). About 57% of the isolates were multiclonal based on genotypes of both MSP-1 and MSP-2.

Table 1. Family grouping analysis of MSP-1 and MSP-2 among Assam and Orissa samples

Area	MSP-1		MSP-2			% of multiclonal isolates
	K1	MAD20	R033	FC27	3D7	
Assam	11 (31.4)	18 (51.4)	6 (17.1)	10 (50.0)	10 (50.0)	50
Orissa	15 (40.5)	14 (37.8)	8 (21.6)	36 (58.1)	26 (41.9)	57

Figures in parentheses are per cent proportions.

Differentiation of Recrudescence from Fresh Infection

Isolates collected from three patients on Day 0 and Day 14 (day of recrudescence) showed different genotypes of MSP-1/MSP-2 suggesting new infection. However, in another two patients, isolates collected on Day 0 and Day 14 showed same genotype of both MSP-1 and MSP-2. To get more conclusive results, analysis of these samples using GLURP is in progress. Fig. 3 shows the genotypes of paired samples.

P. vivax

Twenty *P. vivax* isolates collected from Delhi have been analyzed for GAM1 and MSP-3?. GAM1 system is observed to be polymorphic with alleles in the range of 400 bp to1.2 kb. In MSP-3? PCR product after restriction digestion has shown different RFLP patterns with Hha I and Alu I enzymes. Study is being continued to analyze more isolates from Delhi and Chennai.

Biochemical Characterization and Expression of *P. vivax* Aspartic proteases

Haemoglobin is an important nutrient source for intraerythrocytic malaria organisms. Aspartic proteases play a key role in the degradatve process. Most of the work on aspartic proteases has been carried out in *P. falciparum*, however, no work has till date been reported on the isolation and characterization, and role of aspartic proteases in *P. vivax*. Inhibition of hemoglobin catabolism or other essential functions catalyzed by aspartic proteases in *P. vivax* offers attractive targets for therapeutic interventions.

In continuation of earlier studies, samples from Chennai have been processed for the aspartic protease activity in *P. vivax* parasites. Aspartic protease activity was isolated from a pooled extract of *P. vivax*

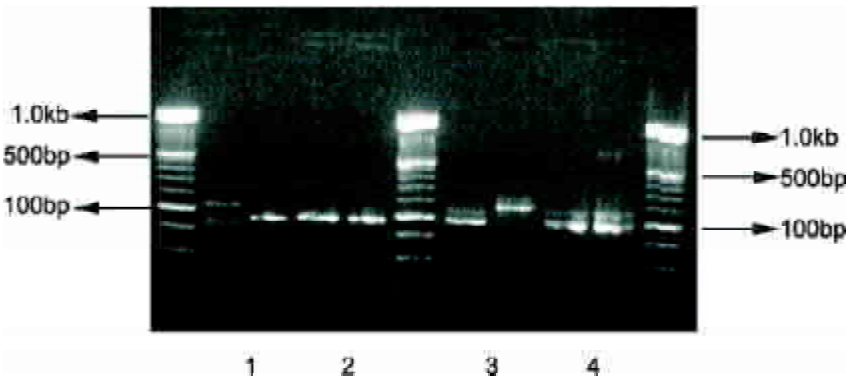

Fig. 3: Gel electrophoretogram showing genotypes of paired samples. Pairs 1 and 3 are different size variants and pairs 2 and 4 are with same size

Table 2. Purification of *P. vivax* aspartic protease

	Protein (mg)	Activity (nmoles/min)	Sp. activity (nmoles/min/mg)	Purity	Yield %
Triton extract	0.758	5.56	0.142	1	100
DEAE extract	0.139	6.92	3.250	23	124
pH 4.5 cut	0.034	2.61	6.960	49	47
Hydroxylapatite	0.013	0.97	23.660	167	17
HPLC	0.006	0.44	40.150	282	8

samples by conventional chromatography on DEAE, hydroxylapatite and gel filtration columns. The resultant peak activity was 282 fold purified over the starting material. The pH optima of the purified enzyme was found to be 4.5–5.0. The IC_{50} for the inhibitor pepstatin was 5nM. PMSF, leupeptin inhibitors of serine and cysteine proteases had no effect on the reaction. For NH_2 terminal sequencing partially purified enzyme was subjected to electrophoresis blotted on PVDF membrane and the 40kD band excised and sequenced. Tentative sequence had revealed 9 of 22 residues identical to most specific mammalian aspartic protease—renin (Table 2). Now a study is being planned to identify and characterize aspartic protease gene in *P. vivax*, using degenerate primers synthesized from the N-terminal amino acid sequences of the plasmepsins from *P. vivax*; to clone and express it in *E. coli*; and to purify and refold the recombinant protein for further studies on its role in parasite functions.

Nitric Oxide Inhibits Plasmepsin, a *P. vivax* Aspartic Protease involved in Haemoglobin Degradation

Nitric oxide (NO) has been known to possess antiparasitic activity in *Plasmodium* species. Parasite proteases are promising targets for antimalarial chemotherapy. In the present study, we have studied the inhibitory effect of NO on the plasmepsin activity, the pepsin like aspartic protease involved in the cleavage of haemoglobin degradation in *P. vivax*. NO donors (±) (E)-4-ethyl-2-[(E) hydroxyimino]-5-nitro-3-hexenamide (NOR-3), S-nitrosoglutathione (GSNO) and sodium nitroprusside (SNP) and activators of nitric oxide production IL-6, IFN-? and TNF-?, were found to inhibit the plasmepsin activity in a dose dependent manner in *P. vivax* extracts, an effect attributable to the nitrosylation of the cysteine residue at the catalytic site. However, the inhibitor of aspartic protease activity namely, pepstatin A was also found to suppress the enzyme activity. These results, therefore, represent new insights into the pathophysiological mechanisms and help in designing strategies to selectively upregulate NO production in *P. vivax* infections for antimalarial chemotherapy.

Glycophospholipid Antigen from *P. falciparum* Culture Supernatant: Isolation, Chemical Analysis and Detection of *Pf* Infection

A *P. falciparum* malaria blood stage antigen was isolated from *in vitro* parasite culture supernatant. The antigen was identified as a mixture of glycophospholipids (GPL). The serological activities of the GPL from *P. falciparum* culture were examined by ELISA against *P. vivax* and *P. falciparum* infected patient's serum. Cross-reactivity of GPL was tested against serum from

patients suffering from other diseases. High anti-GPL antibody was found in case of *P. falciparum* and *P. vivax* infection but not with normal or nonmalarial sera. Blood sera of people from different malaria endemic zones with different history of malaria exposure in recent past were examined by ELISA against both the GPL and RESA Pf/155 (AR1) antigen. The GPL antigen was found to be a glycophospholipid having galactose, mannose, xylose and glucose moiety in the glycogen part and also contain phosphate group and lipid (Tables 3–4).

Table 3. Comparison of ELISA O.D. value between glycophospholipid isolated from parasitized and nonparasitized *Pf* culture supernatant

Ag	Pooled serum	Dilutions		
		100	200	400
GPL_1(Parasitized)	*Pv*	0.57	0.50	0.36
	Pf	0.66	0.54	0.23
	Neg	0.34	0.28	0.19
GPL_2(Nonparasitized)	*Pv*	0.48	0.36	0.28
	Pf	0.47	0.35	0.32
	Neg	0.42	0.35	0.32

Circulating Histidine Rich Protein II Antigen and Specific Antibody Responses in *P. falciparum* Patients during Acute Infection and after Treatment

P. falciparum synthesizes three histidine rich proteins, HRP-1, HRP-2 and HRP-3. HRP-1 was identified as parasite knob associated antigen, whereas HRP-2 was identified as surface exposed protein complex available in all natural isolates irrespective of knob phenotype. HRP-3 is available on parasite surface at the lowest abundance compared to HRP-1 and HRP-2. *P. falciparum* HRP-2 plays a crucial role in parasite development and growth. *Pf*-HRP-2 circulates in blood even after 14 days post infection. Its C-terminal half induces a partially protective response. It is proposed that HRP-2 may facilitate transport of haemoglobin to the food vacuole and catalyze the reaction. HRP-2 is a structurally well-characterized molecule present in all natural isolates of *P. falciparum*. It shows potential effects on the host immune system and has proven its worth as an antigen for specific diagnosis of malaria.

The study was conducted in a group of patients suffering from uncomplicated falciparum malaria to monitor how long circulating HRP-2 antigen could be detected in blood by sandwich ELISA and to determine the profile of antigen-specific antibodies following antimalarial treatment. Finger prick blood samples from a group of 40 malaria

Table 4. Antibody against glycolipid antigen and its response under different malariogenic conditions

Area	ELISA O.D.				Known status
	No.	R1	*Pf*	Glycolipid	
Raigarh	44	0.44 ± 0.18	0.42 ± 0.20	0.59 ± 0.29	Appearing
Haldwani	42	0.30 ± 0.09	0.34 ± 0.12	0.27 ± 0.09	Disappearing
Rajasthan	46	0.35 ± 0.15	0.47 ± 0.20	0.47 ± 0.17	Epidemic
Control	10	0.21 ± 0.14	0.22 ± 0.13	0.06 ± 0.05	Nonendemic

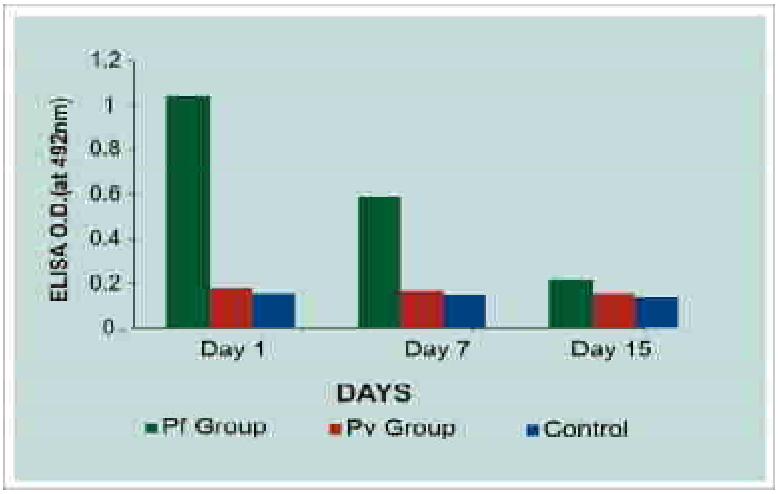

Fig. 4: HRP-2 antigen profile in three study groups

patients (36 with *P. falciparum* and 4 with *P. vivax* infection) were tested for histidine rich protein–2 (HRP-2) antigen by antigen capture assay and for parasite-specific antibodies, both IgM and IgG isotypes by indirect ELISA against HRP-2 peptide and *P. falciparum* infected erythrocyte lysate. These patients were treated with recommended doses of chloroquine and they were followed-up for antigenaemia and antibody level.

HRP-2 antigen detected in whole blood was quite high on Day 1, thereafter a significant decrease in the HRP-2 antigen level was observed on Day 7 and Day 15 in all patients. Similar trend in both anti-HRP-2 and anti-*Pf* IgM was observed. Anti-HRP-2 IgG levels increased moderately in 31 out of 36 on Day 7 and Day 15 as compared to Day 1. Immunoglobulin-G detected in patients against *Pf* lysate showed an increasing trend as compared to Day 1. Significant positive correlation was observed between HRP-2 antigenaemia and IgM in sera of individuals from *P. falciparum* group, whereas negative correlation was achieved between HRP-2 antigen and IgG levels. Blood samples of four *P. vivax* patients had no detectable amount of HRP-2 antigen and anti-HRP-2 antibody, but measurable amounts of IgM and IgG were detected against *Pf* crude antigen due to serological cross-reactivity. Five healthy normal individuals exhibited negativity in three sets of assays (Figs. 4 and 5).

This study provides information that during natural course of infection a high level of circulating free HRP-2 antigen could be detected and at least a measurable or detectable amount could be found even up to seven days after antimalarial treatment. Persistence of antigen was found to be associated with development of antigen-specific antibodies and its antibody mediated neutralization after successful response to antimalarial treatment.

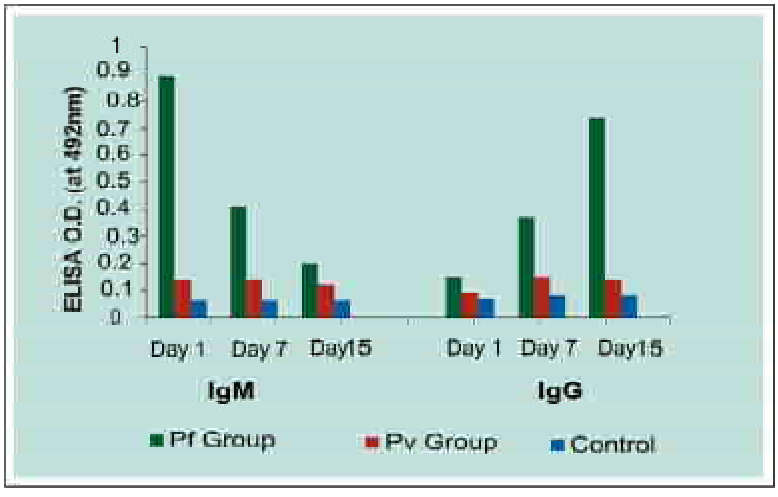

Fig. 5: Anti HRP-2 IgM and IgG profile in three study groups

Studies on Monoclonal Antibodies against *Plasmodium vivax* Erythrocytic Stages

In continuation of the earlier work, monoclonal antibodies derived from two hybridomas MAb1B3C6 and MAb4E6 were tested for their reactivity in *P. vivax* infected erythrocytes by immunofluorescence and

in *P. vivax* crude lysates transferred c nitrocellulose membrane after electr phoresis. Both the antibodies reacte with early and late trophozoites and wi schizont by IFA test (Fig. 6). By imm noblotting, MAb1B3C6 antibody reac ed with nearly 42kDa protein ar MAb4E6 antibody reacted with 30kI proteins of a pooled preparation *P.vivax* parasites collected from diffe ent geographic areas of the country. These two proteins were named as P1 and P2.

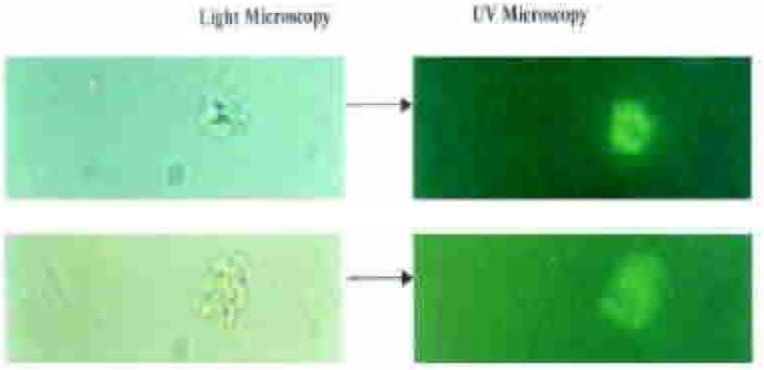

Fig. 6: Indirect immunofluoroscence of acetone fixed *P. vivax* schizont reacted with MAb1B3C6 and MAb4E6

P. vivax protein from crude parasitized erythrocyte lysate was purified by affinity chromatography using two sets of columns of 6MB-Sepharose coupled with immunoglobulin fraction of MAb1B3C6 and MAb4E6. Purified proteins (P1 and P2) were transferred on PVDF membrane and were put in the sequencer. Sequencing of the proteins, P1 and P2 showed significant alignment with erythrocyte membrane associated antigen of one *P. falciparum* clone: p*Pf* and the sequence of P2 showed significant alignment with serine protease inhibitor of *Rattus norvegicus*. Work is continued on the isolation of high affinity parasite antibodies from existing panel of clones to develop diagnostic reagents.

PCR-based Identification of Malaria Parasites

Simple PCR assay using small subunit ribosomal DNA primers was employed. In a total of 79 *P. falciparum* positive bloodspots tested, 98.7% gave amplification in PCR assay, however, in 11 microscopically *P. falciparum* negative bloodspots, none of the samples showed amplification in PCR assay. A multiplex polymerase chain reaction to differentiate *P. falciparum* and *P. vivax* in a single assay was standardized using a mixture of specific primers for each of the two species. The two species could be identified on the basis of size corresponding to 183 and 206 bp respectively for *P. falciparum* and *P. vivax* (Fig. 7).

Studies on Morphological changes in Cerebellar Purkinje cells

A study has been initiated to see the morphological changes taking place in cerebellar purkinje cells and in the surrounding parenchymal cells during cerebral malaria. The study aims to see the effect of plant extracts with antimalarial properties in cerebral malaria in the experimental animal model using Swiss albino mice and *P. berghei ANKA* strain.

Primary Screening of Herbal Products for their Antimalarial Activity

There is an urgent need for new antimalarials particularly to those parasites, which are showing resistance to the existing antimalarials. The medicinal plants or parts of the plants used for the treatment of fever in rural/tribal areas collected from different geographical regions of India were tested for their antimalarial properties. In addition to 13 medicinal plants tested *in vitro* for their antimalarial activity, two new plants collected from Kerala and Uttaranchal have been tested *in vitro* for their antiplasmodial activity. The *in vitro* test was done using both chloroquine sensitive and resistant isolates of *P. falciparum* using schizont maturation inhibition assay. The IC_{50} values were 1.5 ? g/ml and 6 ? g/ml respectively.

In vivo schizontocidal activity was tested for one of the above extracts which was showing good effect. Based on the results of the schizontocidal activity of the earlier studies a compound was prepared by mixing two 50% ethanol extracts and was given to *P. berghei* infected Swiss albino mice. Three groups of five animals each were taken. One group received no drug, which served as control. The other two received 100 mg and 50 mg/kg body weight each respectively. From the control group all the fiveanimals died by Day 9 of experiment where as from the first experimental group out of five animals three survived and in the second group only one animal survived the infection and the average survival rate of other four animals survived was 13 days.

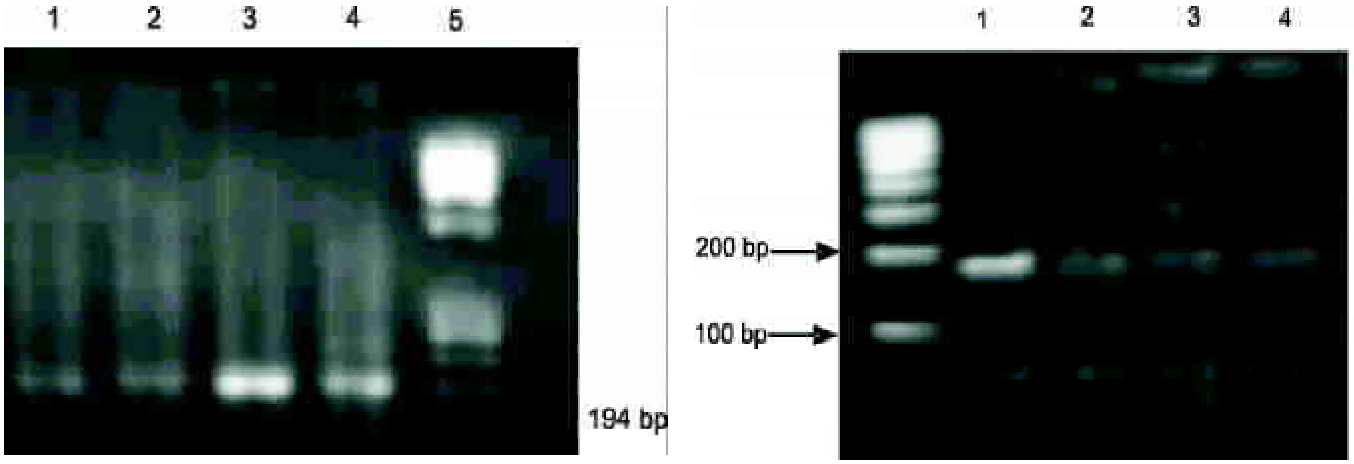

Fig. 7: Gel electrophoretograms showing differentiation of *P. falciparum* and *P. vivax*

EPIDEMIOLOGY

Therapeutic Efficacy of Chloroquine and Sulphapyrimethamine

(i) In Uncomplicated falciparum Malaria (WHO Funded Project under RBM-TSN/DRP)

A standardized protocol has been developed by WHO to assess the therapeutic efficacy of antimalarial drugs against clinically manifested infection with *P. falciparum* in individuals of various age groups. The therapeutic efficacy protocol is based on clinical and parasitological responses of the patients and it has the purpose of determining the practical efficacy of the drug regimen in study areas with the ultimate objective of ascertaining its continued usefulness or the necessity for replacing it in the routine treatment.

Present study has been conducted at seven sites— Kathiatali and Simonabasti of District Nowgong, Assam; Sonapur and Boko of District Kamrup, Assam; Keonjhar Town, Padmapur and Basudepur of District Keonjhar, Orissa. In order to reduce the patient recruitment time, health centre close to well-defined community was identified to conduct the activities at peak malaria season by selecting local pockets and organizing mobile clinics. Microscopically confirmed cases of *P. falciparum* were enrolled according to the inclusion and exclusion criteria. Treatment with recommended drug was given under supervision and the test schedule to follow-up the patients at various intervals for 28 days was maintained. Assessment of efficacy of both chloroquine (CQ) and sulphapyrimethamine (SP) was conducted at five sites each.

HIGHLIGHTS

- Therapeutic efficacy studies of chloroquine in falciparum malaria have shown up to 30% treatment failure of CQ in northeast states and 3% in Orissa
- Therapeutic efficacy studies of chloroquine in uncomplicated vivax malaria in Navi Mumbai, Gautam Budh Nagar and Chennai City showed no treatment failure of CQ
- Evaluation of *Pf* diagnostic kit— ICT Binax showed 100% sensitivity and specificity in detection of *P. falciparum*
- Malaria trend, spatio-temporal dynamics and epidemic cycle in Mewat region of Haryana were worked out using GIS
- Situation analysis of malaria in Gadchiroli district, Maharashtra emphasized the need of health education and community involvement for malaria control
- Impact of climate on malaria was studied in Tumkur (Karnataka) and Bikaner (Rajasthan)
- Analysis of correlationship between malaria cases and meteorological indicators indicated that there is positive as well as negative relationship with meteorological indicators in different areas
- A field trial site for malaria vaccine is being developed

In CQ study areas, wherever patients showed treatment failure to CQ were treated with second line drug— SP and they were then followed-up as per study protocol. It is observed that 30% cases showed treatment failure to CQ in District Nowgong (NK), where revised drug policy has already been introduced. In Kamrup district (KS and KB), treatment failure with CQ was less than 25%, which denotes the said regimen is still effective. Almost all the patients from Padmapur and Basudepur of District Keonjhar responded to CQ, treatment failure was noticed only in two patients (3%). Treatment with SP showed adequate clinical and parasitological responses in all the patients except one in KS, Kamrup.

Table 1. Baseline characteristics in chloroquine treated patients

Parameters	Navi Mumbai	G.B. Nagar
Drug dose (over 3 days)	25 mg/kg	25 mg/kg
No. of cases	85	48
Males/Females	68/17	31/17
Age (range)	1–65yrs	3–59 yrs
% patients with fever (Day 0)	55	45.8
H/o use of antimalarials	None	None
Adverse effects		
Vomiting	8.2%	—
Giddiness	2.3%	—
Purities	—	6.9%
Parasitaemia (range)	—	280–10,960 ?l
Fever clearance time		
24 h	87%	95%
48 h	100%	95%
72 h	100%	100%
Parasite clearance time		
48 h	98.8%	100%
72 h	100%	—

(ii) In Uncomplicated vivax Malaria

Navi Mumbai: Study was conducted in collaboration with Navi-Mumbai Municipal Corporation at six urban health posts—CBD, Karave, Nerul, Sanpada, Koparkhirne and Airoli. Up to December 2002, 85 patients of *P. vivax* malaria were enrolled and 28 day follow-up was conducted. The baseline characteristics of patients are listed in Table 1. All patients responded to treatment with chloroquine.

Gautam Budh Nagar (Uttar Pradesh): The district has seasonal transmission of malaria and is epidemic prone. A total of 48 (31 males + 17 females) patients of vivax malaria in the age range of 3–59 years were enrolled from PHC Dadri, Distt. Gautam Budh Nagar. There was no treatment failure up to Day 28 in this group of patients (Table 1).

Chennai City (Tamil Nadu): The study was undertaken in Sowcarpet, which is situated in the northern coastal belt of Chennai. The area is hyper endemic for malaria and the transmission is perennial. Study was conducted from the Central Malaria Laboratory of the Chennai Corporation located at Basin Water Works street (Div. 38) in Sowcarpet area. All fever cases reporting at the clinic were screened for malaria positivity by the Chennai Corporation staff. Patients with vivax malaria were sent to the MRC clinic established in the same building for enrollment in the study. Up to December 2002, 143 (130 males + 13 females) cases were enrolled of which 136 (95.1%) cases were adults. No recrudescence was observed in any patient in 28 day period. The parasite clearance time was 2 days in 87.8% patients, 3 days in 8.7% and >3 days in 3.5% (Table 2).

Table 2. Analysis of parasite count in patients

Day	No. of cases	Mean parasites/?l	SD	Minimum	Maximum
Day 0	115	7237.7	6499.03	352	39680
Day 2	115	32.14	163.46	0	1360
Day 3	115	13.2	18.0	0	120
Day 7,14, 21 and 28	115	Nil	Nil	Nil	Nil

Evaluation of Diagnostic Kit— ICT Binax

In continuation of earlier studies evaluation of ICT binax diagnostic kit based on detection of HRP-2 was done. The results are shown in Table 3. The study showed that ICT binax kit is highly sensitive and specific for the detection of *P. falciparum.*

Table 3. Results of ICT Binax diagnosis

Microscopic diagnosis	No. tested	Test kit result	
		+ve	–ve
P. falciparum	77	77	0
P. vivax	40	0	40
–ve	32	0	32
Total	149	77	72

Association of Leptospirosis in Patients of Severe falciparum Malaria

Severe and complicated falciparum malaria presents with many complications like cerebral malaria, jaundice, acute renal failure, etc. Among these, acute renal failure occurs in < 1% cases but the mortality from these cases are reported up to 45%. Data from Ispat General Hospital, Rourkela shows nearly two fold increase in the number of patients with severe complications like acute renal failure and jaundice over a period of five years. Leptospirosis is an acute anthropozoonotic infection prevalent worldwide and is emerging as an important public health problem in India. The clinical picture of leptospirosis mimicks severe and complicated malaria especially that of acute renal failure and jaundice. The recent increase of acute renal failure and jaundice among malaria patients at Rourkela may be due to leptospirosis alone or in combination with malaria. Hence, a collaborative study was undertaken to rule out the presence and/or association of leptospirosis among these patients. A detailed clinical, hematological and biochemical examination was done in these patients. There were 13 severe malaria patients (microscopy/ICT +ve 8; microscopy/ICT –ve 5) and 8 uncomplicated malaria (microscopy/ICT +ve). Serum samples were tested for leptospirosis by Leptospira IgM specific Agglutination Test and Lepto Tek Dri Dot Test at NICD, Delhi. None of these were positive by serology for leptospirosis. The study will be continued with larger sample size representing all age groups.

Spatio-temporal Dynamics of Malaria in Mewat

Malaria Trend and Epidemic Cycle

In Mewat, API during 1991-2001 revealed that from 1991–1993 there was a declining trend and the API reached below 2, later during 1994 and 1995 API increased to ~ 5. In 1996, the API was around 33, statistically more than average ±2 S.D.) and was declared as an epidemic year. Thereafter a decline was observed and the API in 1998 reached below 2, subsequently by 2001 it was < 0.5 (API < 2 is considered as very low risk area and need not ot be covered by indoor residual spraying as per NAMP norms) (Fig. 1). Statistically malaria incidence in the

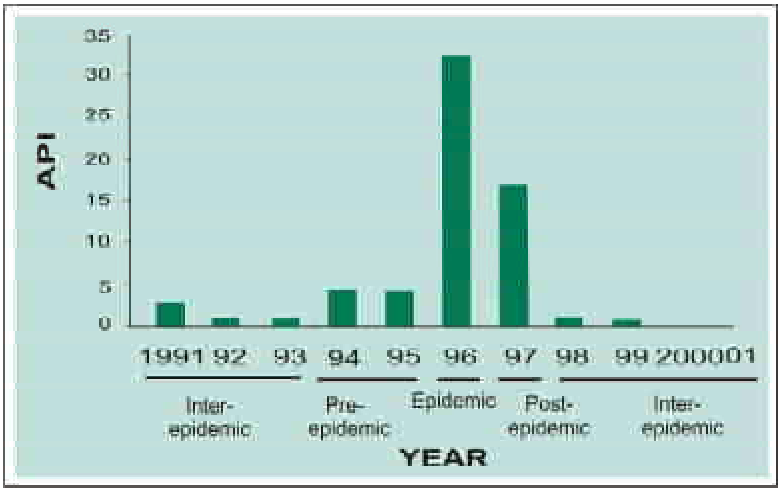

Fig. 1: Annual parasite incidence in Mewat from 1991 to 2001 showing different epidemic phases

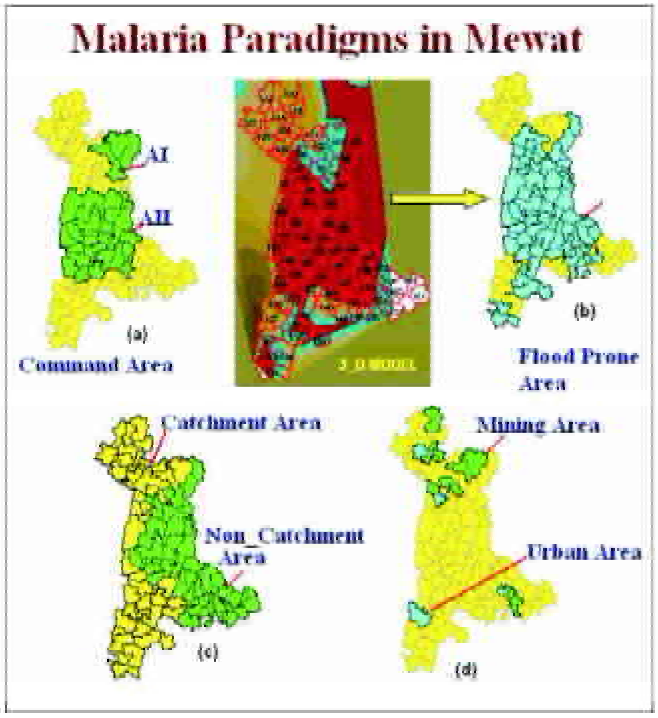

Fig. 2: (a) Two command areas AI and AII in Mewat; (b) Flood prone areas; (c) Catchment and non-catchment areas; and (d) Urban and mining areas

years 1993 and 1998 were found similar. Thus the entire cycle, 1991 to 2001, a span of 11 years was classified into different epidemic phases, such as inter-epidemic (1991–93), pre-epidemic (1994–95), epidemic (1996), post-epidemic (1997) and once again as the inter-epidemic phase (1998–2001). A post-epidemic investigation in 1996, revealed that *P. vivax* was the prominent species of malaria but *P. falciparum* was also prevalent in some pockets.

Identification of Paradigms

Using GIS, based on geographic reconnaissance, ecological and socioeconomic profile initially five malaria paradigms—irrigation command, catchment, mining, urban and flood prone areas were identified (Fig. 2). Section-wise map of the area was overlaid on thematic maps to delineate the sections falling in 5 malaria paradigms. Each paradigm exhibited its own eco-epidemiological characteristics and potential for maintaining malaria transmission of varying intensity.

Malaria Receptivity by Paradigm

It may be seen from the Fig. 3 that during 1991 overall API in all the paradigms was around 5, then declined to around 2 by 1993, again increased during 1994 and 95 representing pre-epidemic phase when it touched around 7 API in all paradigms. Epidemic phase started in 1996, 1997 was the post-epidemic year and 1998–1999 once again represent the inter-epidemic period. During 1996 different paradigms responded differently, maximum amplification occurred in urban/semi urban paradigms with API about 45. This was followed by flood prone, command area A-II and noncatchment paradigms which exhibited the same amount of amplification and the API reached around 40. Mining paradigms showed about 20. The lowest malaria incidence was observed in command area AI (API about 10). By 1998 malaria incidence reached below 2 in all the paradigms. Further decline continued to reach API below 1 by 1999 subsequently in 2000 and 2001 API reached below 0.5 in all paradigms.

Spatio-temporal distribution map of malaria for the years 1991 to 2001 depicted spatial spread of various epidemic phases (Fig. 4). The sections other than < 2 API were extracted for the years 1993 and 1998 and overlaid on paradigm maps to study eco-epidemiological profile of malaria during inter-epidemic phase. It revealed that API in 1993 and 1998, the years of similar malaria situation in the last two inter-epidemic periods, flood prone area, irrigation

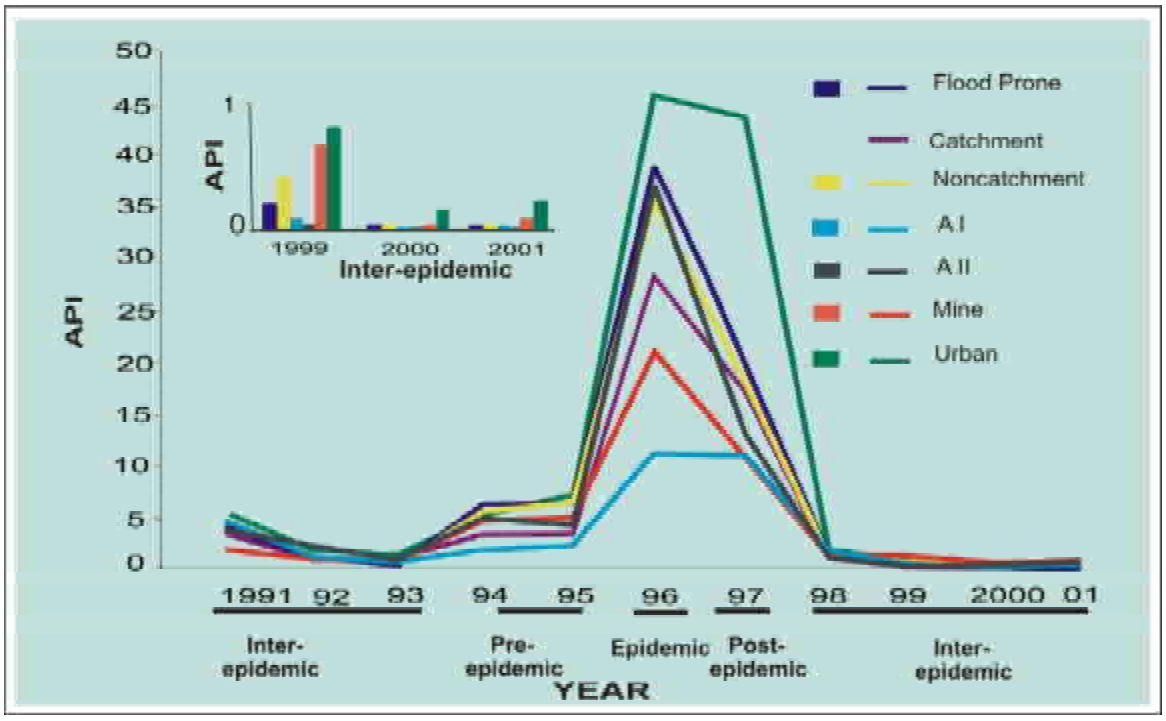

Fig. 3: Paradigm-wise different epidemic phases in Mewat (1991–2001).

command area AII and noncatchment area retained as active pockets of malaria transmission. Amplification started during 1994–1995 (pre-epidemic phase) and engulfed all paradigms by 1996 (epidemic year). Therefore, there is a need for quantitative assessment through field surveys in these paradigms to explain above phenomenon of residual malaria and to identify 'epidemic risk factors' to prevent future epidemics.

Serological Profile following Malaria Outbreak in Mewat Region of Haryana

Mewat region of Gurgaon district in Haryana experienced heavy rain and inundation in 1996, followed by a severe malaria outbreak. High incidence of *P. falciparum* infection and deaths were reported though control measures were taken at a large-scale to combat the epidemic situation of the area by government and local agencies. Assessment of the efficacy of intervention measures to control malaria transmission, however, received little attention.

In addition to this nothing was known about the antimalarial immune status and parasitological exposure level of the population. In an attempt to understand

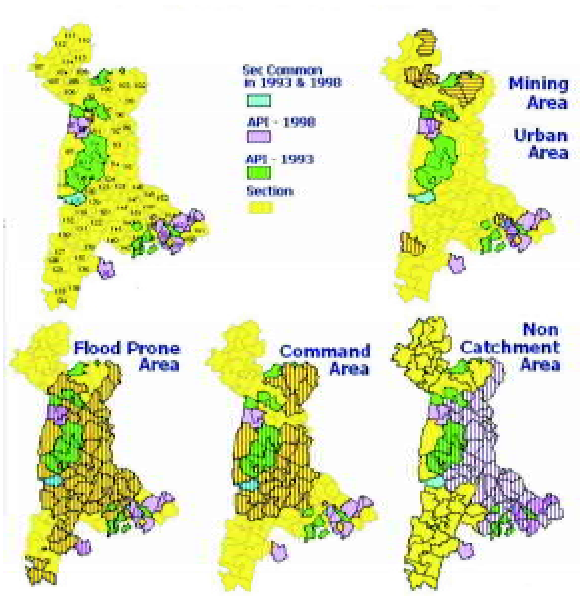

Fig. 4: GIS identifies malaria active sections during inter-epidemic period and their related paradigms. It revealed that active pockets confined to three paradigms—flood prone, non-catchment and command area AII.

Table 4. Anti AR1 and anti *Pf* antibody (IgG) levels in the finger prick blood samples collected from residents of CHCs Nuh and Firozepur Jhirka in the aftermath of malaria outbreak (1997 and 2001)

Year	Village	Total no. of blood samples	Mean±SD ELISA OD_{490}		% Mean seropositivity	
			AR1	*Pf*	AR1	*Pf*
1997	FN	211	0.96±0.15	0.90±0.09	98.8	98.2
	SL	195	0.66±0.25	0.74±0.28	82.0	82.2
	ML	261	0.79±0.20	0.95±0.16	96.0	95.6
	KH	179	0.83±0.17	0.80±0.15	96.4	95.8
2001	FN	47	0.65±0.24	0.56±0.22	69.8	68.9
	SL	83	1.01±0.23	0.93±0.22	98.8	98.6
	ML	70	0.95±0.26	0.75±0.19	96.4	94.8
	KH	48	0.77±0.19	0.68±0.18	89.6	85.5
	FN (School children)	45	0.436±0.064	0.53±0.07	80.0	17.0

FN—Firozepur Namak; SL—Salmbha; ML—Malabh; KH—Khedla.

the epidemic situation and subsequent consequences, cross-sectional surveys were conducted during February–March 1997 and April 2001 to study the antimalaria immune status of the residents of four villages of community health centres (CHCs) Nuh and Firozepur Jhirka, using ELISA as a tool and parasitological exposure level by microscopic blood slide examination.

Seroepidemiological observation in the aftermath of outbreak showed high titers of IgG antibody directed against AR1 synthetic peptide (EENVEHDA–C) and *P. falciparum* crude antigens. Parasitological results—slide positivity rate (SPR), slide falciparum rate (SfR) and per cent *Pf* were observed to be low during these surveys. A negative relationship was noticed between the levels of anti AR1, *Pf* antibody and parasitological results in the residents (Table 4).

Seroepidemiological studies could be used to evaluate the immune status of the population, transmission pattern and assessment of efficacy of intervention measures. The data could also be utilized for effective surveillance of malaria.

Seroepidemiological Assessment of Resurgence of Malaria in Haldwani Area, after withdrawal of the Bioenvironmental Control Programme

Cross-sectional seroepidemiological studies were undertaken in Haldwani area including plain and forested areas in pre and post-bioenvironmental control programme (implemented in 1986) to assess the actual malaria situation in the area and the impact of control programme. Bioenvironmental control programme was withdrawn (in 1990's) as malaria situation was improved and decline in malaria incidence was reported. Before the implementation of bioenvironmental control programme, sero-reactivity against AR1 antigen (a nonapeptide [EENVEHDA(C)] representing an epitopes in the 3 carboxy terminal immunodominant repeat region of the ring infected erythrocyte surface antigen (RESA/*Pf* 155) of *P. facipuram* was observed to be 0.695±0.150 and 0.628±0.105 in children population, but after the implementation, seroreactivity decreased considerably. The AP1 and ELISA O.D. recorded in children population in 1991 in nontransmission and transmission periods were 0.244±0.089 and 0.134±0.064

respectively in the years 1992 and 1993, it remained low (0.145± 0.058 and 0.269±0.09) indicating improvement in malaria situation. Antimalarial antibody levels measured in 368 small children from the plain area in 1997 showed moderate levels of anti AR1 IgG antibody. High levels of anti AR1 IgG antibody were observed in adult population in the forested area in 1997. Gradual rise in malaria incidence was noticed in Haldwani area as 456 active fever cases with 11.2 SPR and 1.1 SfR in 1998 and 625 fever cases with 18.9 SPR and 9.9 SfR in 1999 were reported, supporting our observation and confirms the resurgence of malaria (Tables 5). Using linear regression analysis of ELISA O.D. values and known annual parasite index (API) of a given area a formula was developed to calculate equivalent transmission index (ETI=270.55/ELISA O.D.±7.40) from which the level of endemicity could be estimated. Five villages from plain areas were considered for estimating malaria status through serology. Each village showed moderate anti AR1 ELISA O.D. which indicates a moderate malaria transmission. On comparison of seropositivity and equivalent transmission index, the values confirms the same.

Table 5. Impact of bioenvironmental control programme on the seroreactivity against AR1 *P. falciparum* antigen and malariogenic condition of Haldwani region (1989-2000)

Year	Month	Mode of transmission	Total no. of children population	AR1 ELISA O.D.	API	SPR	ETI
1989	Oct	Peak	208	0.695 ± 0.150	52.2	20.4	196.8
1990	Jan–Feb	Non	684	0.628 ± 0.105	42.3	20.3	177
1991	Jan–Feb	Non	161	0.24 ± 0.089	17.4	17.5	72.3
1991	Nov	Peak	104	0.134 ± 0.064	5.2	5.8	43.6
1992	Sep/Oct	Peak	104	0.145 ± 0.058	4.2	7.5	46.6
1993	Jan	Non	146	0.269 ± 0.090	6	6.8	88.4
1997	Feb–Mar	Non	368	0.42 ± 0.069	–	–	118.31
2000	Feb–Mar	Non	84	0.73 ± 0.15	–	–	205.13

Whereas in two forested villages where implementation of control programme is impossible due to lack of accessibility, malaria incidence was very high. Annual parasite index collected from MRC field stations were 231.2 and 162.99 for HP and JL villages. Accordingly, mean AR1 ELISA O.D., percentage seropositivity and equivalent transmission index, all the values are very high which indicates the correlation of parasitological and serological data (Table 6).

Table 6. Village-wise malaria status in plain and forested area of Haldwani during February–March 1997 in all age groups after the withdrawal of bioenvironmental control programme

Area	Villages	No. examined	Mean AR1 O.D. ± S.D.	% Seropositivity	ETI
Plain	AR	91	0.43 ± 0.08	55.9	118.41
	SNBC	19	0.42 ± 0.09	51.0	115.90
	NVC	171	0.40 ± 0.14	56.5	113.03
	RP	58	0.35 ± 0.09	36.2	98.73
	JN	29	0.54 ± 0.15	51.0	145.48
Forest	HP	231.2	0.85 ± 0.22	89.10	221.73
	HP2	23.12	0.92 ± 0.13	96.00	238.95
	JL	162.99	0.69 ± 1.80	86.86	182.62

Cut of value for AR1 = 0.35 ELISA O.D.

Blood samples from three villages were collected from nonmalarial in-

Table 7. Anti AR1 IgG antibody titer in adult population of Haldwani (Plain area) during March 2000 (Nontransmission season)

Area	No. examined	AR1 ELISA O.D.	ETI
Daval Chaur	33	0.67±0.18	188.66
Haripur Motia	28	0.78±0.13	218.43
Kishan Pur	23	0.75±0.14	210.31

Cut off value for AR1 = 0.35 ELISA O.D.

dividuals. Each village showed moderate to high transmission as per anti ARI ELISA O.D. and equivalent transmission index. From our hypothesis, this information indicates that villagers experienced malaria in the previous year of sample collection (Table 7). In conclusion, seroepidemiological information can estimate the malaria status of a population much better compared to other classical methods.

Delineation of Breeding Habitats and Landscape Features Suitable For *An. culicifacies* Abundance using Satellite Remote Sensing (ICMR Task Force Project)

The study was continued in three selected PHCs of Tumkur district. Entomological surveys including positivity of breeding habitats, emergence of adult species and man hour density of anopheline vectors (indoors) were undertaken along with ecological changes in January and June 2002. Parasitological surveys for point prevalence were also carried out.

Satellite data (IRS 1C/D LISS III and PAN) products were procured for the dates of entomological surveys. False colour composite/hybrid colour composite images were generated from merged products of LISS III and PAN. Supervised classification was done and statistics of land use features like water bodies, barren area, rocks with vegetation, barren rocks, etc. were generated in respect of 27 villages for May 2001. Based on data of January 2002, FCCs images were generated and statistics of land use features were also generated in respect of six villages of Tovinkere PHC. Findings are given below:

(i) Tanks, streams, ponds, marshy areas and irrigation wells were found as breeding habitats of *An. culicifacies* in the decreasing order.

(ii) May/June month was found critical in differentiating entomological and ecological parameters supporting *An. culicifacies* populations.

(iii) Analysis of satellite data revealed that tanks, streams, ponds, marshy areas were detectable by LISS III and PAN merged products. Irrigation wells whose positivity for *An. culicifacies* was insignificant, were not detectable.

(iv) The difference in land use features in villages of high and low malaria categories indicated that in villages of high malaria incidence presence of water in water bodies (0.36–35.78%), more vegetation cover (24.9–85.72%), less barren area and scrub (0–12.9%), less barren rocks (0.13–9.68%) as compared to 1.44–3.2% water bodies, 15.07–34.19% vegetation cover, 8.57–30.21% barren area and scrub, and 0.35–20.33% barren rocks in least malarious area (Byalya). The presence of streams and tanks in the vicinity of human settlements were found more productive for breeding of *An. culicifacies.*

(v) The village-wise analysis of satellite image revealed that remote sensing may be used for ecological change detection at village level and for stratification of high/low malarious areas.

Impact of Climate Change on Malaria in India

As a part of Ministry of Environment and Forests, Government of India, preparation of India's Initial National Communication (NATCOM) to the UNFCCC on vulnerability assessment and adaptive measures due to climate change, a study was undertaken on the impact of climate change on malaria in India.

Based on monthly fluctuations in malaria cases in the year 2000 (NAMP data), regions vulnerable to climate change were identified. Keeping in view the minimum requirement of temperature and relative humidity (RH) for development of *P. vivax* and *P. falciparum* parasites, transmission windows (TWs) of malaria in different cities (representing concerned state) were determined. Based on the areas vulnerable to climate change were further identified.

The projected rise in temperature (T) due to climate change (1.5, 2.4 and 3.8°C by the year 2020, 2050 and 2080 respectively) and precipitation (2 $\pm$ 1% by 2020, 3 $\pm$ 1% in 2050 and 7 $\pm$ 3% by 2080) were added in monthly mean temperature and RH of baseline year 2000. The TWs were determined based on the TWs temperature alone as well as in combination of T and RH.

When we look at the projected temperature in the years 2020, the TW is likely to increase by one month in northeastern states, Rajasthan, Uttar Pradesh and Gujarat while by two months in J&K, Himachal Pradesh and Madhya Pradesh. The TW is likely to remain unaffected in Andhra Pradesh, Chhattisgarh, Haryana, Punjab, Karnataka, Kerala, Maharashtra, Orissa, Tamil Nadu, Uttaranchal and West Bengal. Since malaria transmission dynamics is complex, affected by rainfall pattern, agricultural practices, socio-economic conditions and the intervention measures undertaken, projection of malaria based on temperature alone may not hold true. If intervention measures being practiced in India are implemented effectively, the whole scenario of malaria may change (drastic reduction), let temperature or rainfall be suitable.

Longitudinal case studies were undertaken in Tumkur (Karnataka) and Bikaner (Rajasthan) districts to find out correlation of climatic factors—temperature, RH and rainfall with malaria to find out suitable adaptive measures. Surveys on socio-economic conditions prevalent in the areas were also made to assess the impact of climate change on socioeconomic conditions and thereby on malaria. The results of the case studies in Tumkur revealed that temperature fluctuation during different years is not much but the rainfall pattern at the threshold of TW helps in providing suitable RH for effective transmission of malaria. In Bikaner district, the rainfall at the threshold of TW for at least two consecutive months is important in causing increase in malaria cases. The overall analysis of correlationship between malaria cases and meteorological indicators indicate that there is positive as well as negative relationship with meteorological parameters in different areas.

SITUATION ANALYSIS

District Ghaziabad (U.P.): Study was carried out in two PHCs Garh Mukteshwar (high risk of malaria) and Dasna (low risk of malaria) in Ghaziabad district (U.P.) from 7 to 17 October

Table 8. Epidemiological indices in PHC, Garh Mukteshwar, Distt. Ghaziabad (U.P.)

Village	Population	TBS	SPR	SfR	Cases/000	*Pf*/000
Nanu Pura	5505	7	0	0	0	0
Bhagwantpur	392	3	33.3	33.3	2.5	2.5
Jireena	3415	8	12.5	0	0.29	0
Salar Pur	2741	5	0	0	0	0
Sharifabad	790	2	0	0	0	0

2002. Parasitological survey was carried out in both the PHCs for few days and was compared with the data collected by DMO, Ghaziabad In PHC, Garh Mukteshwar the slide positivity rate (SPR) ranged from 0 – 33.3 and SfR also ranged from 0–33.3 (Table 8). In PHC, Dasna no case was reported. Malaria cases recorded by the District Malaria Officer in five villages of each PHC based on active case detection at an interval of 15 days were nil in these villages. Entomological observation revealed that *An. culicifacies* was predominant species and per man hour densities ranged between 7 and 27 in Garh Mukteshwar PHC and in Dasna PHC *An. culicifacies* man hour density was in between 7 and 14 during October. Susceptibility test was carried out as per WHO procedure and found that *An. culicifacies* was resistant to DDT in both the PHCs (Table 9).

District Gadchiroli (Maharashtra)

Maharashtra government has been able to reduce malaria in most of the districts but the problem in Gadchiroli district was still persisting in spite of implementation of the main tools of intervention—indoor residual spray (IRS) by deltamethrin and fever radical treatment (FRT) to the best possible. An investigation was made to find out the reasons of persistence of malaria in Gadchiroli district during November 2002.

Annual blood examination rate was found to be ranging from 49 to 94 indicating that the health workers were reaching to community efficiently. *Pf* % during 1997 to 2001 ranged from 51 to 66%. The slide positivity rate ranged from 1 to 2.72. The coverage by two rounds of IRS (synthetic pyrethroids) was around 90% and for better compliance to radical treatment,

Table 9. Result of insecticide susceptibility test in PHC, Garh Mukteshwar, Distt. Ghaziabad (U.P.)

Insecticide	Species									
	An. culicifacies					*Cx. quinquefasciatus*				
	Doses (%)	No. exposed	Exposed period (hr)	% knock-down	Corrected mortality	Doses (%)	No. exposed	Exposed period (h)	% knock-down	Corrected mortality
Deltamethrin	0.02	15 x 2	1	100	100*	0.02	25 x 2	1	44	100*
DDT	4	15 x 2	1	1	0	4	25 x 2	1	—	0
Malathion	5	15 x 2	1	100	100	5	25 x 2	1	38	100

*100% knock-down was obtained within 15–25 min of exposure.

blister pack was introduced containing 4 tablets of chloroquine of 600 mg each and 4 tablets of 45 mg primaquine.

Detailed parasitological investigations revealed that 25% of positive cases detected during the survey were having gametocyte stage of *P. falciparum* parasite. It indicates that compliance to FRT was not satisfactory. Entomological findings revealed that the main vector, *An. culicifacies* was resting indoor and was 100% sensitive to the sprayed insecticide—deltamehthrin. *An. fluviatilis* was encountered rarely. Results of cone bioassay indicated that on the wall surface sprayed before 10 days, mortality of *An. culicifacies* was around 68%, while the mortality was only 13% on the surface sprayed before one month. It indicates that the quality of spray was not satisfactory and required close surpervision. It was learnt that just after second round of spray in October, the inhabitants got their houses smeared/white washed.

A deep insight into the reasons of persistence of malaria revealed that the intervention measures were being affected by the social factors like locked houses, not allowing spray in all the rooms and smearing of houses by mud/white wash. FRT was affected by the approach of inhabitants to seek the help of local quacks rather health services and not taking full course of antimalarials. The overall findings emphasize the importance of health education to community and their involvement in malaria control for achieving best results.

Development of a Field Site for Malaria Vaccine Trial (A Collaborative Project with International Centre for Genetic Engineering and Biotechnology, New Delhi – Funded by Department of Biotechnology, Govt. of India)

Studies were continued in hyperendemic and low endemic areas of Sundargarh district, Orissa for preparation of a site for malaria vaccine trial. There are 13 study villages with a total population of 4221 under Gurundia and Birkera PHCs of Sundargarh district out of which, 8 villages with a population of 2058 are located in deep forest close to the streams, under the influence of *An. fluviatilis* and have persistent malaria transmission. The remaining five villages with a total population of 2163 are located in a plain area close to a perennial river where *An. culicifacies* is the main vector and malaria transmission is low and seasonal. The study villages are predominantly inhabited by ethnic tribals—Oram, Munda, Khadia, etc. The geographical reconnaissance and mapping of the study villages were completed and a computer based epidemiological as well as GIS database has been developed.

Entomology and Parasitology: Longitudinal and cross-sectional parasitological and entomological surveys were conducted in all the study villages. The weekly surveillance was conducted in all the study villages through village volunteer workers. The SPR, SFR, Pf per cent and annual parasite index (API) in the forest villages were 41.8, 35.1, 84.0 and 397.5 respectively, whereas in the plain area villages these were 17.1, 12.1, 70.8 and 55.5 respectively. The malaria incidence was more in the younger age groups up to 15 years and the highest incidence was in the 0–5 age groups in the forest area but in the plain area, malaria cases were evenly distributed in all the age groups.

During the year, cross-sectional mass blood surveys were conducted in all the study villages during March, June and November covering all the transmission seasons— low, moderate and high respectively. The parasite rate in the forest and plain area during March, June and November was 11.4. 1.1; 9.8 and 0.6, 20.2, 1.84 respectively. Malaria was more prevalent in the younger age groups as compared to adults. Out of the total malaria cases in forest area, the prevalence of *P. falciparum*, *P. vivax* and *P. malariae* accounted for 82, 16 and 2 per cent respectively but in the plain area, the prevalence of *Pf* and *Pv* were 68 and 32 per cent respectively. The spleen rate in children living in forest area villages was above 80 per cent throughout the year and also in adults it was above 30 per cent, whereas, in plain area the spleen rate in children and adults was ranging from 20–35 and 2–12 respectively. A study on the therapeutic response of chloroquine in the study population is in progress.

Entomological surveys were carried out in two indicator villages each in forest and plain areas. During the year, 14 anopheline species from forest area and 11 species from plain area were recorded. *An. culicifacies* was widely prevalent in both the areas whereas *An. fluviatilis* was totally absent in the plain area. The man hour density (MHD) of *An. culicifacies* in the forest and plain area was in the range of 3.3–35.0 and 8.0–26.0 respectively. The MHD of *An. fluviatilis* in the forest area ranged from 0.2–15.3. Results of all night mosquito landing collections on human baits showed that *An. fluviatilis* prefer to bite humans and the man biting rate in forest area during low, moderate and high transmission seasons was 0.62, 6.5 and 16.5 bites per person per night respectively, whereas it was nil in the plain area. The average man biting rate of *An. culicifacies* in the forest and plain area was 0.3 and 0.5 bites per person per night respectively. The sporozoite rate in the forest area during low, moderate and high transmission seasons was 0, 1.32 and 2.1 respectively and it was nil in the plain area. The entomological inoculation rate (EIR) in the forest area during low, moderate and high malaria transmission seasons was 0, 0.085 and 0.35 infective bites per person per night respectively whereas, it was nil in the plain area. The study population is being increased to 15,000 for which new villages have been identified and baseline data collection as well as census operation is in progress.

Host Immune Responses: Study proposed on host immune responses was to determine the antibody level to vaccine candidate *P. falciparum* antigens, namely MSP-1_{19}, EBA175 and TRAP developed in the course of natural infection in different age groups in the study population.

Finger prick blood samples were collected by cross-sectional survey from individuals belonging to forest and plain areas. Indirect ELISA was done to assess the antibody level against three recombinant peptides. From the results, it was observed that overall IgG antibody profile with three antigens were higher in individuals of forest areas than those in plain areas. There was an age-wise increase in IgG level in both areas. Antigen specific IgM profile in study group was low and almost similar in two areas. Anti-TRAP antibody level found moderately high in adults of both forest and plain areas. IgG1 and IgG2 were the predominant subclass responses to all three antigens. Proportion of high responders to MSP-1_{19} and EBA175 was comparable in children and adults. A group of sera from older age group showed elevated level of IgG3 to MSP-1_{19} and EBA175. There was an association between high IgG/IgG1 antibody to MSP-1_{19} and EBA175 responses and lower prevalence of *P. falciparum* parasitaemia.

A longitudinal cohort study of parasite episodes, reinfection and antibody level against *P. falciparum* antigens of interest has been proposed.

Multiplicity of Infection: Field collected blood spots from *P. falciparum* positive patients were genotyped using PCR method. Primers of MSP-1 (block-2) and MSP-2 (central variable region) were used for PCR assay. Primary reaction primers were gene specific and nested PCR primers were family specific. About 105 samples were analyzed for two years during different transmission seasons. Multiplicity of infection ranged from 1.1 to 3.28 in low and high transmission seasons. Number of alleles observed was 22 in MSP-1 and 24 in MSP-2. A high proportion of isolates (>60%) had multiplicity of infection greater than 1.

Sequence Diversity: The sequence diversity in three malaria vaccine candidates namely MSP-1 (C-terminal 19kDa), EBA175 region II, and TRAP was determined in *P. falciparum* isolates. Primers were designed covering part of block-16 and entire block-17 of MSP-1. Sequencing of 16 field isolates has shown polymorphisms only at few amino acid positions. Eight alleles were observed in Indian field isolates, out of which three are unreported till now (from any other country study).

Primers were designed for TRAP N-terminal and C-terminal regions. Sequencing of 10 field isolates for TRAP N-terminal region showed polymorphisms at 25 amino acid positions, and three were reported for the first time. Important motifs like RGD, IQQ and the motif in thrombospondin related proteins were conserved in all the field isolates. Sequencing of 10 field isolates for TRAP C-terminal region showed variable PNP repeats in different isolates.

Sequencing of EBA175 F2 region of 16 field isolates has shown polymorphisms at 19 amino acid positions. Only five of these were reported between different strains. Only selected amino acid residues were targeted for mutations.

Malaria Clinic at 2, Nanak Enclave

A total of 1822 patients attended the malaria clinic at 2, Nanak Enclave, Delhi during January to December, of which 36 were found positive for malaria (27 positive for *P. vivax* and 9 for *P. falciparum)*. Slide positivity rate (SPR) and slide falciparum rate (SfR) are given in Table 1. Clinical examination was done and specific and symptomatic treatment was given. Blood samples were collected from volunteers with their consent for host-parasite interaction (23 *Pv* and 1 *Pf*) and genetic diversity studies (21 *Pv* and 4 *Pf*). The month-wise distribution of malaria cases is given in Table 1.

Table 1. Data from malaria clinic, MRC, 2, Nanak Enclave, Delhi

Month	BSE	Total	*Pv*	*Pf*	SPR	SfR
Jan	131	3	2	1	2.29	33.33
Feb	105	3	2	1	2.86	33.33
Mar	126	0	0	0	0	0
Apr	149	0	0	0	0	0
May	84	4	4	0	4.76	0
Jun	114	2	2	0	1.75	0
Jul	174	9	5	4	5.17	44.44
Aug	231	2	1	1	0.87	50
Sep	320	9	9	0	2.81	0
Oct	213	2	1	1	0.94	50
Nov	120	2	1	1	1.67	50
Dec	55	0	0	0	0	0

Malaria Clinic at 22, Sham Nath Marg

A total of 72 patients attended the malaria clinic at 22, Sham Nath Marg, Delhi or were referred from hospitals for blood examination and treatment of malaria during the period of January–December. Out of five patients found positive for malaria, only one patient was diagnosed as *P. vivax* and four as *P. falciparum*.

HIGHLIGHTS OF RESEARCH ACTIVITIES UNDER "INTEGRATED DISEASE VECTOR CONTROL OF MALARIA" PROJECT

Nadiad (Gujarat): A study on the health impact assessment of the Sardar Sarovar Narmada water resources development project on communicable diseases with particular emphasis on mosquito-borne diseases was initiated. A WHO-sponsored project was started in selected talukas in northern Gujarat to develop strategy for integrated control of vectors of malaria and dengue. New insecticide formulations were evaluated for malaria vector control. Support in technical, training, health education, epidemic investigation and containment was provided to antimalaria programme.

Jabalpur (Madhya Pradesh): Malaria epidemic investigation was carried out in Betul district on the request of state health department. Impact of DDT indoor residual spraying was evaluated. Situation analysis was done in Mandla, Dindori, Sagar and Damoh districts on the request of the NAMP. A study on placental malaria was done. Tolerability and efficacy of artesunate plus chloroquine or sulphapyrimethamine combination vs single agent chloroquine or sulphapyrimethamine in the treatment of uncomplicated falciparum malaria was studied. A study on integrated control of malaria in Sagar district was initiated. Insecticide susceptibility status of malaria vectors was evaluated in eight districts.

Hardwar (Uttaranchal): Work on isolation of plant origin antimalarials and mosquito larvicidal, adulticidal agents and repellents continued. Allethrin residue in the environment was examined. Concentrations of chloroquine, sulfadoxine and quinine were determined at the time of recurrence to confirm multi-drug resistant *P. falciparum* malaria in Assam. Monitoring of bioenvironmental control strategy which has been successfully demonstrated at BHEL, Hardwar, IDPL, Rishikesh and IOC, Mathura was done. Consultancy support was provided to other industrial complexes.

Rourkela (Orissa): Longitudinal epidemiological studies were continued in forest and plain areas characterized by hyper- and meso-endemic malaria situations respectively to prepare a site for malaria vaccine trial. A study on malaria transmission dynamics in tribal areas is underway. Gametocytocidal effect of compound 80/53 was studied. A study on the prevalence of G-6-PD deficiency in tribal population was undertaken. Work was initiated on the study of genetic diversity of *P. falciparum* and *P. vivax* and development of microsatellite markers. Therapeutic efficacy of chloroquine and sulphapyrimethamine in uncomplicated *P. falciparum* malaria was evaluated in Keonjhar. Situation analysis of malaria was done in three districts of Orissa. Technical, training and health education supports were provided to antimalaria programme. Field evaluation of tablet formulation of deltamethrin was done.

Sonapur (Assam): The major areas of research included evaluation of therapeutic efficacy of antimalarials, situation analysis under roll back malaria initiative, and malaria outbreak investigations in selected districts of Assam. The emergence of multiple drug resistance in *P. falciparum* has been detected in areas of stable transmission, and for their containment, intensive surveillance coupled with antivector measures with a focus on insecticide treated nets is being advocated. Other activities included GIS mapping of *An. minimus*, genetic composition of *P. falciparum* isolates, sibling species composition of *An. fluviatilis*, IEC activities during antimalaria month, and mass propagation and distribution of larvivorous fishes (guppy) in towns of Assam.

Panaji (Goa): Technology transfer on bioenvironmental control of malaria to the Mormugao Port was continued during the fifth year. A study of malaria in migrants was done. Fungal isolates with larvicidal activity were maintained for further study. A new rapid *Pf* diagnostic kit — Binax was evaluated. Malaria situation analysis was done in Panaji and Cansarvarnem PHCs as part of RBM. Therapeutic efficacy of chloroquine in *P. vixax* malaria was evaluated in Navi Mumbai. Training support to antimalaria programme was provided.

Bangalore (Karnataka): Use of larvivorous fish was scaled-up in five talukas of four districts and in Mangalore city. Geographical reconnaissance of mosquito breeding habitats was undertaken to plan integrated disease control. A remote sensing study on delineation breeding habitats of *An. culicifacies* was undertaken. Work on mosquito control plan in Bangalore city was carried forward. Monitoring of parasite sensitivity to chloroquine and susceptibility of *An. culicifacies* to malathion and deltamethrin was also done.

Haldwani (Uttaranchal): Dynamics of malaria transmission in some areas of Bhabar region, District Nainital, Uttaranchal was studied. Insecticide susceptibility of *An. culicifacies* was evaluated in four districts in Chhattisgarh (as part of situation analysis) and two districts (Nainital and Udham Singh Nagar) in Uttaranchal. Technical, training and health education supports were provided to the antimalaria programme.

Chennai (Tamil Nadu): During the year, studies on bioecology of *An. stephensi* and its role in malaria transmission were undertaken in and around Chennai city. Areas were mapped based on the ecological variants. Association of mosquito breeding with urban rainwater harvesting was studied. Study on therapeutic efficacy of chloroquine for the treatment of vivax malaria was completed, which showed that it was a fully effective drug. A study in Rameswaram revealed development of resistance to pyrethroids in *An. culicifacies*. Other activities included technical support, health education, training, and malaria diagnosis and treatment through malaria clinic in Chennai.

Shahjahanpur (Uttar Pradesh): Geographical reconnaissance of mosquito breeding habitats in Shahjahanpur district was undertaken. Malaria surveys were conducted in some high risk PHCs. Situation analysis of malaria was carried out in four districts—Lakhimpur Kheri and Sonbhadra districts in Uttar Pradesh, and East Godavari and Visakhapatnam in Andhra Pradesh. Insecticide susceptibility status of malaria vectors was evaluated. Malaria clinic was run. Health education activities were organized.

Shankargarh (Uttar Pradesh): Malaria situation analysis was undertaken in Districts Ranchi and Hazaribagh in Jharkhand state and Gadchiroli in Maharashtra. Malaria clinic at the field station provided early diagnosis and prompt treatment of malaria. Health education activities were organized. Susceptibility of *An. culicifacies* against different insecticides like DDT, malathion and deltamethrin was evaluated.

Car Nicobar (A&N Islands): Epidemiological study of malaria among primitive tribes Jarawas was continued. Role of duffy blood group in vivax malaria was studied. Health education and training activities were organized. Technical support in malaria outbreak investigation and containment was provided. Diagnostic and treatment services at malaria clinic are being provided. Use of larvivorous fish in vector control was promoted.

BIOLOGICAL MATERIAL BEING MAINTAINED AT THE CENTRE

MOSQUITO SPECIES

An. stephensi

From urban and semi-urban areas

Nehru Place, Delhi
Okhla, Delhi
Chennai, Tamil Nadu
Gurgaon, Haryana
Nanak Enclave, Delhi
Hardwar, Uttaranchal

From rural areas

Ladpur, Haryana

Morphological mutants

Red eye (r) – sex linked recessive
Black larvae (bl) – autosomal semi-dominant
Golden yellow (gy) – autosomal recessive
Creamish white eye (cw) – new mutant
Reddish brown eye (rb) – new mutant

Biochemical variants

Bahadurgarh (EST-2)

An. culicifacies Complex

Species A

Dehra, Uttar Pradesh
Burari, Delhi
Rourkela, Orissa

Species B

Acrocentric Y-chromosome lines

Ladpur, Haryana
Haldwani, Uttaranchal

Submetacentric Y-chromosome lines

Rameswaram, Tamil Nadu
Rourkela, Orissa

Insecticide resistant lines

DDT resistant – Ladpur, Haryana
Malathion resistant – Ladpur, Haryana

Species C

Submetacentric Y-chromosome line

Jabalpur, Madhya Pradesh

Insecticide resistant line

DDT resistant – Jabalpur (M.P.)

An. fluviatilis Complex

Species S and T – Rourkela,Orissa
Species T and U – Hardwar, Uttaranchal
Species T – Haldwani and Hardwar, Uttaranchal

An. sundaicus

Cyclic colonies established from Car Nicobar
Katchal, Tressa (A&N Islands)

An. annularis

Nathupura, Delhi

Aedes aegypti

Delhi

Culex quinquefasciatus

Delhi
Pondicherry
Sonepat, Haryana
Mewat, Haryana

Insecticide resistant lines

Malathion resistant – Sonepat, Haryana
Permethrin resistant – Sonepat, Haryana

Lambdacyhalothrin resistant	–	Sonepat, Haryana
Deltamethrin resistant	–	Sonepat, Haryana
Cyfluthrin resistant	–	Sonepat, Haryana
Fenthion resistant	–	Sonepat, Haryana

Morphological mutants

Red eye (re)
Scarlet eye (se)

PARASITE SPECIES

Human and Nonhuman Malaria Parasites Available at the Parasite Bank

Human Plasmodia
- Nonadapted cryopreserved isolates of *P. falciparum*, *P. vivax* and *P. malariae*
- Sera/plasma from infected patients

P. falciparum
- Adapted/characterized isolates
- Different stages of the parasite from culture
  - Merozoites (from culture supernatant)
  - Ring (by synchronization)
  - Gametocytes (by Hypoxanthine treatment)
  - Free parasites for antigen preparation (by Saponin lysis and ultrasonication)

P. vivax
- Sporozoites harvested from artificially fed mosquitoes

Nonhuman Plasmodia
- Different species of avian, simian and rodent plasmodia
- Rodent plasmodia infected rats/mice
- Sera/plasma from respective vertebrate hosts

Cell Lines
- Hepatoma cell line: Hep G2 A16 used in the *in vitro* cultivation of pre-erythrocytic stage malaria parasites
- Myeloma cell line: SP2
- Hybridomas: 2A 10 (anti-*P. falciparum* sporozoite antibody secreting cells)
  2 F2 1 A7 (anti-*P. vivax* sporozoite antibody secreting cells)

Parasite Bank is fully established as a National Resource Centre. The human and non-human parasites cryopreserved/maintained and other biological material produced in the bank are being used for collaborative studies and supplied to various organizations.

Details of *P. falciparum* Isolates Adapted/Cryopreserved

Place of collection	No. of isolates collected	Adapted/Cryopreserved
Delhi	175	70
Ghaziabad (Uttar Pradesh)	27	22
Shankargarh (Uttar Pradesh)	39	27
Baharaich (Uttar Pradesh)	21	–
Gautam Budh Nagar (Uttar Pradesh)	39	–
Shahjahanpur (Uttar Pradesh)	6	6
Mandla (Madhya Pradesh)	23	15
Jagdalpur (Chhattisgarh)	14	6
Sonapur (Assam)	25	2
Rourkela (Orissa)	33	9
Rameswaram (Tamil Nadu)	1	1
Jaisalmer (Rajasthan)	39	27
Bharatpur (Rajasthan)	35	1
Alwar (Rajasthan)	25	–
Nuh (Haryana)	25	2
Kolkata (West Bengal)	19	–
Visakhapatnam (Andhra Pradesh)	12	–
Bissam Cuttack (Orissa)	22	–
Total	**580**	**188**

Details of Characterized *P. falciparum* Isolates

Species/Strains of parasite	No. of isolates
Adapted isolates susceptible to chloroquine	54
Adapted isolates resistant to chloroquine	52
NF-54: an infective gametocyte producing strain of *P. falciparum*	1
3D 7A : a clone of NF-54	1
A-4 : a clone with binding property to CD36	1
Dd2: a clone which can invade trypsin treated erythrocytes	1
Field isolates which can invade trypsin treated erythrocytes	3
Field isolates which can invade neuraminidase treated but not trypsin treated erythrocytes	3
Field isolates which can invade normal erythrocytes but not neuraminidase or trypsin treated erythrocytes	3
Field isolates which can invade both neuraminidase treated and trypsin treated erythrocytes	5
Field isolates which can form rosettes	3
Field isolates which can bind to CSA	1
Field isolates which can bind to CD36	9
Field isolates which can bind to ICAM-1	2
Isolates with isoenzyme profile of GPI, GDH, ADA and LDH markers	22
Isolates with MSP-1, MSP-2 and GLURP markers	40

Nonhuman Malaria Parasites available at the Parasite Bank

Parasite species	Source	Susceptibility to antimalarials
Simian malaria		
P. cynomolgi bastianelli	NICD, Delhi	Not done
P. knowlesi	–do–	–do–
P. fragile	CDRI, Lucknow	–do–
Avian malaria		
P. gallinaceum	NICD, Delhi	Not done
P. relictum	Wild, Delhi	–do–
Rodent malaria		
P. berghei NK-65	PGI, Chandigarh	Not done
P. berghei NK-65*+	CDRI, Lucknow	CQ sensitive
*P. berghei**	–do–	CQ resistant
P. berghei	–do–	Quinine resistant
P. chabaudi	INSERM, Paris	Not done
P. vinckei petteri 279 BY	–do–	–do–
P. yoelii yoelii 265 BY**	–do–	–do–
*P. yoelii nigeriensis***+	LSHTM, London	–do–
P. yoelii nigeriensis	CDRI, Lucknow	Multi-resistant
P. yoelii	ICGEB, New Delhi	Not done

*Oocyst positive in *An. stephensi*; **Oocyst and sporozoite positive in *An. stephensi*; +Infective gametocyte producing strain.

EXPERIMENTAL ANIMAL FACILITY

Rabbits, pigeons, domestic fowls, laboratory mice, etc. were procured, maintained and utilized for research purpose throughout the year as per the guidelines issued by the concerned authorities. These animals were housed at 22, Sham Nath Marg and 2, Nanak Enclave buildings and were used as blood meal source to mosquitoes of different species and strains maintained at the Centre. Laboratory mice were used in screening the antimalarials, host-parasite intraction studies and maintenance of rodent plasmodia at the parasite bank. Experiments on animals were performed with the approval of the Scientific Advisory Committee (SAC) and Institutional Animal Ethics Committee (IAEC) of the Centre.

INFORMATION, EDUCATION AND COMMUNICATION

National Science Day Celebrations

National Science Day (NSD) was celebrated in the semi-urban area located in the outskirts of Delhi by holding a health education camp. The theme of the NSD for the year 2002 was "Wealth from Waste". Lectures, live demonstrations of various stages in the life cycles of mosquitoes and malarial parasites, larvivorous fish, EPS beads, and video shows were held in the health education camp. Flyers in Hindi were also distributed.

Observance of Antimalaria Month

A workshop on malaria was organized for paramedical staff of Primary Health Centre, Chawla near Delhi. Charts and exhibitions were displayed. Popular lectures on malaria transmission, prevention and control were delivered. Demonstrations of blood slide preparation and biological control agent were given. Slide and video shows were held. Brochures and pamphlets were distributed.

Participation in Health Exhibition

Malaria Research Centre participated in Swadeshi Arogya Mela held at Jawahar Lal Nehru Stadium, New Delhi during 7 to 12 February. The significance of this mela was top level scientists and technologists explored the possible options towards national self-reliance and sustainable development. MRC displayed its research activities and achievements in the form of exhibition panels. Live demonstrations, blood examination and video shows were also held during the six-day exhibition. School children and general public visited this exhibition in large numbers. Flyers and folders were distributed to the visitors.

Distribution of VHS Cassettes

Over 40 video cassettes were reproduced and 25 were distributed to the trainees, various health departments and NGOs.

MRC WEBSITE

A website of Malaria Research Centre was designed and launched on 28 October 2002 on the occasion of the Silver Jubilee celebrations of MRC. The URL of website is **http://www.mrcindia.org.** The website contains information on:

1. Directory of all the MRC-field stations with separate page for each field station describing their activities and contact information.
2. Directory of all scientists of MRC with their address, e-mail and research interest.
3. Information about publications of MRC including year-wise research publications for last 10 years (1993–2002) and separate page for *Indian Journal of Malariology*.
4. Information about activities of MRC such as, activities of Audio-Visual Unit, research activities and services rendered by MRC.
5. Pages with 'Search' form (within website) and 'Feedback' form.
6. Links to important malaria websites.

Additionally, following documents have been uploaded on the website:

1. Chapter wise PDF files of document entitled, 'A Profile of Malaria Research Centre' published on the occasion of Silver Jubilee celebrations of MRC.
2. Abstract book of International symposium on 'Challenges in Malaria and Prospects for Research', organized by Malaria Research Centre, Delhi from 29–31 October 2002.
3. Annual Report 2001.

Database on research projects and their activities are under preparation.

The website has been included in DMOZ open directory project. The various pages of website can now be searched using Google® search engine.

PUBLICATIONS AND LIBRARY

PUBLICATIONS

***Indian Journal of Malariology*:** The *Indian Journal of Malariology* keeping up the tradition and hopes with which it was revived by *Indian Council of Medical Research* in the year 1981 has entered in its successful twenty second year of publication.

Malaria Patrika: A popular Hindi magazine is being published to create awareness in the community on malaria and its control which has been appreciated by the community.

Apart from above a special document namely " A profile of Malaria Research Centre" containing the research activities carried out by Malaria Research Centre in a nutshell, extramural and intramural projects undertaken, brochures, folders, books, pamphelts, etc. published, Ph.D students guided, training courses organized, papers published by MRC scientists, etc. was pubished on the occassion of completion of 25 years of MRC foundation. This document was applauded by the National and International community. A Souvenir was also published for the International symposium "Challenges in Malaria and Prospects for Research" which was organized by Malaria Research Centre at CGO Complex, Lodhi Road, New Delhi from 29-31 October 2002. This document contains the messages from VIPS, programme and abstract of research papers presented by eminent National and International scientists.

LIBRARY

Centre has one of the best libraries in the country in the field of malaria having 6507 books, 3005 bound journals, 3526 reprints, 18 video cassettes, 27 audio cassettes, 5 microfilms, 13 theses and 100 National and International reports. A total of 70 journals (60 International and 10 National) are being subscribed besides 10 journals which are being received on exchange or complimentary basis. Five magazines, 7 English and 5 Hindi newspapers are also being subscribed. In the financial year library has added 185 new books and 8 new journals.

Library renders its services not only to the scientists/research scholars of the Centre, but also to various National and International Universities and Organizations. In the year 2002 library has provided services to the scientists of 15 International and National Institutes and Universities. In the process of modernization during the year 2001-2002 library has purchased, MEDLINE CD (since 1966) and library software —Libsys. Data on 1200 books have been updated and cataloguing work is in progress with the Libsys software. Library is also affiliated with DELNET (Developing Library Network) to access various database like Union Catalogue of books/periodicals to provide required material to the scientists and users. Library provides abstracts, references, CAS and SDI services. MEDLINE CD search and internet facility to access on line journals is also available for the users. Library also provides science citation index services to scientists through INSDOC. Library has provided this facility to 20 scientists of the Centre during the year. Library provides inter-library loan facilities and reprographic services on demand. Library has started a half yearly *MRC Library News Letter* since 2002.

HINDI WEEK CELEBRATIONS

As per the mandate of the Govt. of India to promote Hindi as an Official Language the Malaria Research Centre celebrated Hindi Week from 16 September to 21 September. On this occassion of "Hindi Week" essay writing, noting-drafting and debate competitions were organized. On 16th September, Sh. S.C. Sharma, Admn. Officer organized the noting-drafting competion in which nine staff members participated, further on 18 September, Dr. Mantosh Malhotra, Dy. Director, organized the essay writing competition in which 14 staff members took part. The topics of essay writing competition were *Vishva ka Badhta Taapman/Bharat-Pak Sambandh/Malaria Anusandhan Kendra ke Pachhis Varsh*. A debate competition was organized by Sh. B.N. Nagpal, Asstt. Director, the topic was *CNG Sankat/Sankat Mochan* and 20 staff members participated. The judges of the competition were Dr. Vijay Srivastava, ICMR and Dr. Aruna Srivastava, Asstt. Director, MRC.

Lastly, the award ceremony took place and the winners of various competitions were given prizes by Dr. M.A. Ansari, Deputy Director (SG). The details of various award winners are as follows:

Noting-drafting competition	—	Sh. Pradip Dutta (I Prize)
	—	Sh. K.C. Sehra (II)
	—	Smt. Monika Malhotra (III)
Essay writing competition	—	Dr. Padmavati Tyagi (I Prize)
	—	Sh. D.S. Sontiyal (II)
	—	Sh. M.P. Singh (III)
Debate competition	—	Sh. V.K. Jain (I Prize)
	—	Sh. S.C. Sharma (II)
	—	Ms. Bhanukala (III)

The awards for use of Hindi promotion scheme were given by Dr. Arati Roy, Deputy Director. All these activities were coordinatd by Sh. S.C. Sharma, Admn. Officer and Dr. Vandana Sharma, Hindi Translator of MRC.

GLIMPSES OF INTERNATIONAL SYMPOSIUM ON "CHALLENGES IN MALARIA AND PROSPECTS FOR RESEARCH"

Malaria Research Centre organized an International symposium on "Challenges in Malaria and Prospects for Research" from 29–31 October 2002 at SCOPE Convention Centre, CGO Complex, Lodi Road, New Delhi to commemorate the completion of 25 years of the Centre. Eminent National and International scientists attended the symposium.

Distinguished prominent persons in the country applauded the organization of symposium. Messages and wishes were received from Dr. A.P.J. Abdul Kalam, the President of India; Shri Atal Bihari Vajpayee, the Prime Minister; Shri Bhairon Singh Shekhawat, the Vice President; Shri Shatrughan Sinha, the then Minister of Health and Family Welfare; Ms. Gro Harlem Brundtland, Director General, World Health Organization; Dr. S.P. Agarwal, Director General, Directorate General of Health Services (Govt. of India); Prof. N.K. Ganguly, Director General, Indian Council of Medical Research; Dr. G. Satyavati, Former Director General, Indian Council of Medical Research; Dr. V.P. Sharma, Former Director, Malaria Research Centre and Prof. R.C. Mahajan, Emeritus Professor, Postgraduate Institute of Medical Education and Research, Chandigarh.

Dr. Sarala K Subbarao, Director, Malaria Research Centre welcomed all the delegates and the symposium was inaugurated by lighting the lamp by Prof. P.N. Tandon, President, National Brain Research Centre, New Delhi and Meghnad Saha, Professor of the National Academy of Sciences, Prof. R.C. Mahajan, Dr. Padam Singh, Addl. Director General, ICMR, Dr. V.P. Sharma, Dr. S. Pattanayak, Dr. Sarala K. Subbarao and Dr. M.A. Ansari. Inaugural function was presided by Prof. R.C. Mahajan and Prof. P.N. Tandon was the Chief Guest. MRC Foundation Day lecture was delivered by Prof. V.S. Chauhan, Director, International Centre for Genetic Engineering and Biotechnol-

ogy, New Delhi. Prof. P.N. Tandon released the book entitled, " A Profile of Malaria Research Centre" specially published for this august occassion which contains the research activities and achievements of Malaria Research Centre during the last 25 years. Mementoes were distributed to those staff members who have completed 25 years of service in MRC on this occasion. Dr. V.P. Sharma inaugurated the exhibition on important research activities and achievements, and publications brought out by Malaria Research Centre.

Technical sessions were started from 1400 hours on 29 October. The first technical session on " Malaria Disease Burden" was chaired by Dr. P.R. Arbani, WHO (SEARO). Dr. Arvind Pandey acted as Co-chairperson. Dr. R.C. Dhiman was the Rapporteur and Dr. Neena Valecha coordinated the session. In this session lead papers were presented by Dr. Jotna Sokhey, NAMP, Delhi, Dr. P. Mahapatra, National Institute of Health Systems, Hyderabad, Dr. B.S. Das, Ispat General Hospital, Rourkela and Dr. A.P. Mitra, National Physical Laboratory, New Delhi. Dr. Neeru Singh, Dr. S.K. Sharma, Dr. T. Adak, Dr. S. Biswas, MRC, Delhi, Ms. Gertrude N. Kiwanuka, Mbarara, University of Science & Technology, Mbarara, Uganda and Dr. Raja Ratnam Abel, CMC&H, Vellore presented their research papers in Resources and Research Inputs session which was chaired by Prof. R.C. Mahajan and co-chaired by Dr. J. Mahanta, Regional Medical Research Centre, Dibrugarh, Assam. Dr. P.R. Bhattacharya, MRC, Delhi and Dr. B. Shahi, MRC, Shankargarh were the Rapporteurs for this session.

Session II commenced at 0900 hours on 30 October with the theme " Understanding of Vectors: The Targets for Effective Malaria Control" was chaired by Prof. K.S. Rai, Jalandhar and co-chaired by Dr. A.P. Dash, Life Sciences Institute, Orissa. Dr. Vas Dev, MRC, Sonapur and Dr. K. Raghavendra, MRC, Delhi acted as Rapporteurs and Dr. Neeru Singh, MRC, Jabalpur coordinated the session. Dr. Mohammad Shahabuddin, National Institute of Health, Bethesda, USA, Prof. Marcelo Jacobs

Lorena, Case Western Reserve University, Cleveland, USA, Prof. Dilip Deobagkar, Pune University, Pune, Dr. Sarala K. Subbarao, MRC Delhi presented their lead papers. Dr. B.N. Nagpal, Dr. Aruna Srivastava, Dr. R.C. Dhiman, Dr. Nutan Nanda and Mr. O.P. Singh, MRC, Delhi presented their research papers in Resources and Research Inputs session which was chaired by Shri N.L. Kalra, former Deputy Director, NAMP, Delhi and co-chaired by Professor N.J. Shetty, Bangalore University, Bangalore. Dr. Hema Joshi, MRC, Delhi and Dr. Alex Eapen, MRC, Chennai were the Rapporteurs.

Session III started at 1400 hours on 30 October with the theme "Vector Control Options" was chaired by Dr. P.K. Das, Vector Control Research Centre, Pondicherry and co-chaired by Dr. S.C. Das, Defence Research Laboratory, Tejpur. Dr. Ashwani Kumar, MRC, Goa and Dr. C.P. Batra, MRC, Delhi acted as Rapporteurs and Dr. M.A. Ansari, Deputy Director (SG.) coordinated the session. Dr. Chusak Prasittisuk (WHO, SEARO), Dr. V.P. Sharma, former Director, MRC, Delhi and WHO Consultant, Dr. Chris Curtis, London School of Hygiene and Tropical Medicine, London, presented their lead papers. Dr. K. Raghavendra, Dr. S.K. Ghosh, Dr. P.K. Mittal, Dr. R.S. Yadav and Dr. R.M. Bhatt, MRC presented their research papers in Resources and Research Inputs session which was chaired by Dr. S.J. Rehman, former Head, Deptt. of Entomology, National Institute of Communicable Diseases, Delhi. Dr. M.S. Malhotra, MRC, Delhi and Dr. M.K. Das, MRC, Car Nicobar were the Rapporteurs.

Session IV commenced at 0900 hours on 31 October with the theme "Parasite Biology/Vaccines/Drugs" was chaired by Dr. V.P. Sharma, Former Director, MRC, Delhi. Dr. Arun Sharma acted as Rapporteur and Dr. T. Adak coordinated the session. Dr. Chetan Chitnis, International Centre for Genetic Engineering and Biotechnology, New Delhi, Dr. Nirbhay Kumar, Johns Hopkins University, USA, Dr. Altaf Lal, CDC Atlanta, USA, Dr. Shobhna Sharma, Mumbai, Dr. G. Padmanabhan, Indian Institute of Science, Bangalore, Dr. Namita Surolia, Jawaharlal Nehru Centre for Advanced Scientific Research, Bangalore, Dr. Amit Sharma, ICGEB,New Delhi, Prof. Y.D. Sharma, All India Institute of Medical Sciences, New Delhi and Dr. Robert G. Ridley, WHO, Geneva, presented their lead papers. Dr. C.R. Pillai, Dr. Arati Roy, Dr. Hema Joshi, Dr. V.K. Dua, Dr. Neena Valecha, MRC and Dr. V. Venugopal Malaria Medicines Venture, Geneva, Dr. Pawan Malhotra, ICGEB, presented their research papers in Resources and Research Inputs session which was chaired by Prof. Dilip Deobagkar, Pune. Dr. Sukla Biswas, MRC, Delhi and Dr. R.P. Shukla, MRC, Haldwani were the Rapporteurs.

All the sessions were attended by a large number of delegates and the deliberations were of high quality. The presentations and interaction between the scientists during these sessions provided valuable information and new leads to the persons engaged in research on various aspects of malaria.

Valedictory session was organized at 1600 hours on 31 October. Dr. S.P. Tripathi, former Director General, ICMR in his valedictory address lauded the organization of symposium and shared his experiences with MRC. Dr. Chetan Chitnis, Prof. Marcelo Jacobs Lorena and Prof. C.F. Curtis enlightened the august gathering by sharing their experiences in malaria research and control. Dr.

M.A. Ansari presented the vote of thanks.

TRAININGS IMPARTED

Dr. R.M. Bhatt

1. Participated as faculty in the training of trainer's at the Regional Training Institute, Bavla, Gujarat.

Dr. Vas Dev

1. Served as a resource person on malaria training workshop for doctors of Assam State Health Services held at Guwahati from 7–11 January under EMCP programme of NAMP.
2. Served as a resource person for the workshop on "Clinical management of malaria for Medical Officers of Assam" under EMCP of NAMP held at Guwahati from 16 to 18 January.
3. Served as a resource person for "Re-orientation training of the Medical Officers of the State of Meghalaya" for *Pf* monitoring held at Shillong from 11–15 March.
4. Delivered a lecture on 'Rapid diagnosis of malaria and its prevention' in the Refresher's course for practicing medical professionals of the northeastern region held at Down-town Hospital, Guwahati from 19–21 December.

Dr. S.K. Ghosh

1. Attended two training programmes for PHC Medical Officers, organized at Dharwad in February and at Bagalkot town in March.
2. Took part in a training programme on mosquito control as faculty for Health Inspectors of various CHCs of Karnataka.

Dr. Hema Joshi

1. Imparted training on PCR based method for diagnosis and genotyping of human malaria parasites to M/s. Getrude N. Kiwanuks, Mbarara University of Science and Technology, Mbarara, Uganda; Dr. W.M.K.T. de Alwis Wikramasinghe Kumudswamy, Scientist from Sri Lankan Antimalaria Compaign Programme, Sri Lanka and Dr. Samia Ali Omer from Sudan.

Dr. Ashwani Kumar

1. Delivered two lectures to engineers on prevention of malaria control in Mormugao Port Trust on 13 February and 6 March and also delivered a lecture on forest malaria in India and its control to Conservators of Forests from different states of India at Panaji, Goa on 19 March.

Dr. Nutan Nanda

1. Imparted training to Dr. (Mrs.) Zakkey Telmadarry, Lecturer, School of Public Health, Tehran University of Medical Sciences, in cytotaxonomic techniques for the identification of sibling species of malaria vectors for two weeks during March.

Dr. C.R. Pillai

Imparted training to the followings:

1. Dr. Md. Farahna, Ph.D. student, Anatomy Deptt., MAMC, New Delhi/UGC fellow from Sudan. Training on handling and maintenance of rodent plasmodia during February.

2. Ms. Gertrude N. Kiwanuka from Uganda. Training in *in vitro* cultivation of malaria parasites and *in vitro* drug sensitivity testing of *P. falciparum* from 12–16 August.
3. Mr. Amit Kumar c/o Dr. Pushkar Sharma, Staff Scientist-IV, National Institute of Immunology (NII), New Delhi. Training in *in vitro* cultivation of *P. falciparum* from 26 August–3 September.
4. Mr. Sivalokanathan, Research Scholar, Deptt. of Pharmacology and Environment Toxicology, Dr. ALM Institute of Basic Medical Sciences, University of Madras, Taramani, Chennai. Training in *in vitro* cultivation of malaria parasites from 23–27 September.
5. Ms. Kumudo Gunasekera, WHO fellow. Training in *in vitro* cultivation of malaria parasite and *in vitro* drug sensitivity testing of *P. falciparum* from 11–14 November and 9–16 December.
6. Ms. Samia Ali Omer, Tropical Medicine Research Institute, Sudan, Khartoum. Training in *in vitro* cultivation of malaria parasite and *in vitro* drug sensitivity testing of *P. falciparum* from 10–20 December.
7. Ms. Sumiti Vinayak and Mr. Anwar Ahmed, Ph.D. students, Deptt. of Biotechnology, AIIMS, New Delhi. Training in *in vitro* cultivation of malaria parasite *P. falciparum* from 16–27 December.

Dr. R.P. Shukla

1. Participated as faculty in a 2-day malariology training course of the malaria workers of the Kumaon region sponsored by NAMP, Delhi at J.N. Hospital, Rudrapur and organized by DMO, Rudrapur from 15 to 16 March.

Dr. S.N. Tiwari

1. Participated in a training programme in January for PHC Medical Officers of Bangalore Division and delivered a lecture on bioenvironmental control of malaria.

Dr. Neena Valecha

Imparted training to the followings:

1. Ms. Gertrude N. Kiwanuka from Uganda from 5–13August.
2. Scientists from Bhutan from 23–29 August.
3. Dr. W.M.K.T. de Alwis Wickramasinghe Gunasekera, WHO fellow from Sri Lanka. Training on diagnostics/therapeutic efficacy and drug trials from 15–18 November and 2–5 December.
4. Delivered a lecture on Rapid malaria diagnosis: Dipstick and method prinicple, merits and field application at Regional Course in Malariology for SEA region, on 15 November.

Dr. R.S. Yadav

1. Participated as faculty in the training of trainer's at the Regional Training Institute, Bavla, Gujarat.

TRAININGS RECEIVED

Dr. M.A. Ansari

1. Participated in an ICMR training programme on R & D management held at IIM, Kolkata from 8–10 February.

Dr. Hema Joshi

1. Attended the WHO/TDR sponsored South East Asian training course on Bioinformatics applied to tropical diseases at International Centre for Genetic Engineering and Biotechnology, New Delhi from 26 April to 9 May 2002.

Dr. B.N. Nagpal

1. Training on G.P.S. organized by Space Application Centre, Ahmedabad at India Habitat Centre, New Delhi on 28 May.

2. GIS training course on Introduction to ArcGIS organized by ESRI, India from 2–5 July.

Dr. A.M. Reetha

1. Training in Statistical computing and clinical trials at the Institute for Research in Medical Statistics (ICMR), New Delhi from 15–19 April.

2. ICMR–FERCAP workshop for Developing standard operating procedures for Institutional Ethics Committees at Agra from 17–19 December.

Dr. Aruna Srivastava

1. Training on G.P.S. organized by Space Application Centre, Ahmedabad at India Habitat Centre, New Delhi on 28 May.

2. GIS training course on Introduction to ArcGIS organized by ESRI, India on 2–5 July.

WORKSHOPS/SYMPOSIA/SEMINARS ATTENDED

Adak, T. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by Malaria Research Centre at New Delhi from 29–31 October and presented a paper entitled, "Relapses and *P. vivax* burden". Also acted as Coordinator for the session on "Parasite Biology/Vaccines/Drugs".

Ansari, M.A. Participated in the " X International Congress of Parasitology (ICOPA)" held at Vancouver, Canada from 4–9 August.

Ansari, M.A. Participated in an International symposium on " Malaria Control in Mekong Region", held at Siem Republic, Kingdom of Cambodia from 10–13 December.

Ansari, M.A. Participated in awareness workshop on "Sound Management of Persistent Organic Pollutants (POPs) in India : Issues and Options" organized by Confederation of Indian Indusries, New Delhi from 6–7 March.

Ansari, M.A. Participated in a workshop for annual action plan 2002 held at NAMP, Delhi from 5–6 and 12–13 February.

Ansari, M.A. Participated in a RBM workshop at Goa from 30 April–2 May.

Ansari, M.A. Participated in a workshop on " Meta Analysis of Data Pertaining to Field Trial with ITMN" held at NAIC Auditorium, NICD, Delhi on 31 May.

Ansari, M.A. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and acted as Coordinator for the session on "Vector Control Options".

Batra, C.P. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and served as Rapporteur for the session on "Vector Control Options".

Bhatt, R.M. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper on "Comprehensive management of disease vectors in Ahmedabad".

Bhattacharya, P.R. Participated in an International symposium on " Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and served as Rapporteur for the session on " Malaria Diseases Burden : Resources and Research Inputs".

Biswas, S. Attended ICMR-Ellison foundation sponsored workshop on "Immunoparasitology" at Regional Medical Research Centre (ICMR), Bhubaneswar from 11–15 February.

Biswas, S. Attended symposium on " Drug Resistance and Malaria Control Strategies", organized by Medecines Sans Frontieres at Guwahati on 28 February and 1 March and delivered a lecture on "*In vitro* drug resistance: Correlation with clinical studies in India".

Biswas, S. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, " Assessment of therapeutic efficacy of chloroquine and sulphapyrimethamine in uncomplicated falciparum malaria". Also served as Rapporteur for the session on "Parasite Biology/Vaccines/Drugs : Resources and Research Inputs".

Das, M.K. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and served as Rapporteur for the session on "Vector Control Options: Resources and Research Inputs".

Dev, V. Participated in the workshop on "Operationalization of Insecticide Treated Mosquito Nets in Assam" held at RMRC, Dibrugarh from 24–26 April, sponsored by WHO-SEARO and presented a paper on "ITNs evaluation – Sonapur experience".

Dev, V. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and served as Rapporteur for the session on "Understanding of Vectors: The Targets for Effective Malaria Control".

Dev, V. Participated in the symposium on "Drug Resistance and Malaria Control Strategies", organized by the Medicines Sans Frontieres (MSF) at Guwahati from 16–18 January.

Dhiman, R.C. Attended an International conference on "Science and Technology— Capacity Building for Climate Change" at New Delhi from 20–22 October.

Dhiman, R.C. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper on "Delineation of breeding habitats and landscape features suitable for *Anopheles culicifacies*". Also served as Rapporteur for the session on "Malaria Disease Burden".

Dhiman, R.C. Participated in "Joint Annual Conference of the Indian Society for Malaria and other Communicable Diseases and the Indian Association of Epidemiologists" at New Delhi from 9–11 November and presented a paper on "Impact of EL Nino southern oscillation events on malaria in India".

Dhiman, R.C. Participated in an International symposium on "Malaria Control in Mekong Region", held at Siem Republic (Kingdom of Cambodia) from 10–13 December and presented paper on "Reasons of malaria outbreaks in India and possible tools for early warning" .

Dhiman, R.C. Participated in a workshop on "Malaria Operational Research" jointly organized by State Malaria Control Society, State Health Resource Unit and Danida Support Unit at Korba (Chhattisgarh) from 23–25 January.

Dua, V.K. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "New compounds with antimalarial activity".

Dua, V.K. Attended a seminar on "Medicines for Malaria Venture", organized by the Confederation of Indian Industries at New Delhi on 11 February.

Dua, V.K. Attended a seminar on "Improving Productivity in Pharmaceutical Development", organized by Waters India Ltd. held at Delhi on 20 May.

Eapen, Alex. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and served as Rapporteur for the session on "Understanding of Vectors: Resources and Research Inputs".

Ghosh, S.K. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Larvivorous fish in malaria control in Karnataka".

Ghosh, S.K. Attended two workshops on "Dengue", jointly organized by BCC, MRC and NICD at Bangalore on 2 and 24 November, respectively.

Ghosh, S.K. Attended a workshop on "Biosafety Issues Related to Genetically Modified Organisms (GMOs)" jointly organized by the Ministry of Forest and Environment and Department of Biotechnology held at Bangalore on 29 November.

Haq, S. Participated in the "XVI National Congress of Parasitology" under the auspices of Indian Society for Parasitology, organized by the Department of Animal Science, MJP Rohilkhand University, Bareilly from 31 October–2 November and presented a paper entitled, "Operational trial of biolarvicides under the antimalaria programme in Surat city".

Joshi, Hema. Attended as a delegate CII/WHO sponsored seminar on "Medicine for Malaria—R&D and Improving Access to Treatment for Malaria" held at New Delhi from 11–13 February.

Joshi, Hema. Participated as a delegate in seminar on "Women Scientists and Technologists for National Development" at Vigyan Bhawan, New Delhi from 8–9 March and presented scientific achievements of women scientists of MRC by displaying posters during the conference.

Joshi, Hema. Presented a paper entitled, "*P. vivax* MSP-3 : Allelic polymorphism among field isolates in India using PCR-RFLP" in "X International Conference of Parasitology (X-ICOPA)", at Vancouver, Canada, 4–9 August.

Joshi, Hema. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Markers for the malaria parasite population structure analysis". Also served as Rapporteur for the session on "Understanding of Vectors : Resources and Research Inputs".

Kumar, Ashwani. Attended "X International Conference of Parasitology (X-ICOPA)" at Vancouver, Canada from 4–9 August and presented a paper entitled, "Three novel strains of mosquito-pathogenic strains from Goa, India".

Kumar, Ashwani. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and served as Rapporteur for the session on "Vector Control Options".

Malhotra, M.S. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and served as Rapporteur for the session on "Vector Control Options: Resources and Research Inputs".

Mittal, P.K. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Biocides in vector control : Challenges and prospects".

Mittal, P.K. Participated in the "Joint Annual Conference of the Indian Society for Malaria and other Communicable Diseases and the Indian Association of Epidemology", organized at New Delhi from 9–11 November and presented a paper entitled, " Mechanism of DDT and pyrethroid resistance in a strain of *An. culicifacies* from south India".

Mittal, P.K. Attended a workshop on "Biosafety Issues Related to Genetically Modified Organisms (GMO)", organized by Biotech Consortium India Ltd., New Delhi from 5–6 September.

Mittal, P.K.Attended a seminar on "Zero Fly, the Insecticide Incorporated Plastic Sheeting a New Tool for Control of Malaria and Vector Borne Diseases Prevention in Complex Emergences", organized by Vestergoard/Frandsen – Diseases Control Textiles at New Delhi.

Nagpal, B.N . Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Taxonomic surveys: Anopheline species diversity".

Nanda, Nutan. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Biological and ecological distinctness among sibling species".

Pillai, C.R. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organizd by MRC at New Delhi from 29–31 October and presented a paper entitled, "Parasite Bank : A national resource at MRC".

Raghavendra, K. Attended a workshop on "Sound Management of Persistent Organic Pollutants in India: Issues and Options", organized by the Confederation of Indian Industries at New Delhi from 6–7 March.

Raghavendra, K. Attended UNDP/UNIDO workshop on "Regional Network on Pesticides Production and Information for Asia and Pacific Region (RENPAP) on Production of Uses and Environmental Friendly Pesticide Formulations and Quality Assurance" held at Indian Institute of Pesticide Formulation Technology, Gurgaon, from 18–22 March and presented a paper entitled, "Insecticides in malaria control—Formulations, application and safety".

Raghavendra, K. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Insecticide resistance and biochemical mechanisms in malaria vectors". Also served as Rapporteur for the session on " Understanding of Vectors—The Targets for Effective Malaria Control".

Raghavendra, K. Participated in the "Joint Annual Conference of ISMOCD and IAE" at New Delhi from 9–11 November and presented a paper entitled, "A new focus of malathion resistance in *Anopheles culicifacies* in District Chhindwara, Madhya Pradesh".

Raghavendra, K. Attended an International symposium on " Malaria Control in Mekong Region" at Siem Republic (Kingdom of Cambodia), organized by Regional Malaria Control Programme—Europeon Commission from10–13 December and presented a paper entitled, "Case studies on insecticide resistance and its management".

Roy, Arati. Participated in an International symposium on " Challenges in Malaria and Prospects for Research", organizd by MRC at New Delhi from 29–31 October and presented a paper entitled, " Glycolipid antigen — A tool for malaria parasite diagnostics".

Shahi, B. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and served as Rapporteur for the session on "Malaria Disease Burden: Resources and Research Inputs".

Sharma, Arun. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and served as Rapporteur for the session on "Parasite Biology/Vaccines/Drugs".

Sharma, S.K. Participated in a malaria workshop at Guwahati from 28 February–1 March organized by MSF (Medecines Sans Frontieres)—An International NGO working in northeast and delivered a lecture on "Malaria epidemiology in tribal areas of Orissa".

Sharma, S.K. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Malaria morbidity in the tribal population of Orissa".

Sharma, S.K. Participated in a workshop on "ITNs Trials in India", jointly organized by MRC and NICD at Delhi on 31 May.

Sharma, S.N. Attended a symposium on "Public Health", organized by Vigyan Samwad, Nainital to commemorate centenary year of the endowment of Nobel Prize to Sir Ronald Ross at Almora from 12–14 May and delivered a lecture on "Tropical disease and their control in Uttaranchal".

Sharma, S.N. Participated in the "Joint Annual Conference of the Indian Society for Malaria and other Communicable Diseases and the Indian Association of Epidemiologists" held at Delhi from 31 October–1 November and presented a paper entitled, "Small-scale field trials of *Bacillus thuringiensis* for the control of mosquito larvae in Kumaon region of Uttaranchal".

Shukla, R.P. Attended a symposium on "Public Health", organized by Vigyan Samwad, Nainital to commemorate centenary year of the endowment of Nobel Prize to Sir Ronald Ross at Almora from 12–14 May and delivered a lecture on "Malaria in Kumaon, Uttaranchal".

Shukla, R.P. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and served as Rapporteur for the session on "Parasite Biology/Vaccines/Drugs: Resources and Research Inputs".

Singh, Neeru. Attended a workshop on "Malaria—Cause, Prevention and Control" at Mandla and presented a paper on "Malaria in Dindori and Mandla" on 13 June.

Singh, Neeru. Attended an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Malaria in morbidity in pregnant women in Madhya Pradesh". Also acted as Coordinator for the session on "Understanding of Vectors : The Targets for Effective Malaria Control".

Singh, Neeru. Attended a workshop on "Impregnated Bednet Meta Analysis" at NAMP, Delhi on 31 May.

Singh, Neeru. Attended a workshop on "Intersectoral Coordination" at Mandla on 2 July.

Singh, Neeru. Attended a workshop on "Epidemic Preparedness" at Regional Health and Family Welfare Training Centre, Jabalpur from 4–7 July.

Singh, Neeru. Attended a workshop on "Placental Malaria" at Yaounde Cameroon from 30 July–2 August.

Singh, Neeru. Attended a workshop on "Malaria Control under EMCP" as resource person for Medical Officers of Madhya Pradesh at Regional Health and Family Welfare Training Centre, Jabalpur from 25–26 November.

Singh, O.P. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Tools for identification of cryptic anopheline species".

Srivastava, Aruna. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "GIS for mapping of malaria vectors".

Srivastava, H.C. Participated in the "XVI National Congress of Parasitology" under the auspices of Indian Society for Parasitology, organized by the Department of Animal Science, MJP Rohilkhand University, Bareilly from 31 October–2 November and presented a paper entitled, "Assessment of economic burden of malaria in Raigad district, Maharashtra".

Subbarao, S.K. Attended a conference on "Entomology Research and Problems", organized by Deptt. of Zoology, Punjab University, Chandigarh and delivered a lecture on "Research progress in the last two decades on malaria vectors in India" on 20 March.

Subbarao, S.K. Attended CII-RFI workshop on "Sound Management of Persistent Organic Pollutants (POPs) in India : Issues and Options" at New Delhi from 6–7 March.

Subbarao, S.K. Participated in the National Bio Resource Development Board (DBT) workshop on "Prospecting of Botanical Pesticides" held at DBT, New Delhi on 26 July.

Subbarao, S.K. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Indian anopheline vector species population structure".

Subbarao, S.K. Attended the seminar on "Women Scientists and Technologists for National Development" at Vigyan Bhawan, New Delhi from 8–9 March.

Valecha, Neena. Attended a symposium on "Drug Resistance and Malaria Control Strategies", organized by Medicines Sans Frontiers (MSF) at Guwahati from 28 February–1 March.

Valecha, Neena. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Clinical drug trials". Also acted as Coordinator for the session on "Malaria Disease Burden".

Valecha, Neena. Delivered a lecture on "Ethical issues in clinical drug trials" at an ICMR-WHO, workshop on "Ethical Issues in Biomedical Research" from 11–13 December.

Valecha, Neena. Delivered a lecture on "Therapeutic Efficacy Protocols" at an Orientation workshop for Medical Officers and Health Assistants at Vashi Hospital, Navi Mumbai on 6 June.

Valecha, Neena. Attended a seminar on "Women Scientists and Technologists for National Development" at Vigyan Bhawan, New Delhi from 8–9 March.

Yadav, R.S. Attended a symposium on "Drug Rsistance and Malaria Control Strategies", organized by Medicines Sans Frontiers (MSF) at Guwahati and presented a paper on "Protection from malaria with insecticide treated mosquito nets" from 28 February–1 March.

Yadav, R.S. Attended an advocacy workshop on "Malaria Free Healthy Life Styles" at Ahmedabad on 10 July and presented a paper on "A model of urban malaria control—Engineering aspects".

Yadav, R.S. Participated in an International symposium on "Challenges in Malaria and Prospects for Research", organized by MRC at New Delhi from 29–31 October and presented a paper entitled, "Insecticide treated mosquito nets: Efficacy and sustainability".

IMPORTANT MEETINGS ATTENDED

Dr. M.A. Ansari

1. Participated in a meeting to discuss on IDVC issues at ICMR (HQ), New Delhi on 5 March.
2. Participated in a meeting of Course Expert Committee to design the syllabus of appllication-oriented course "Integrated Pest Management" held at Conference Room, School of Science, IGNOU, New Delhi from 18–19 March.
3. Participated in a meeting of Foundation Day Lecture on " Strategies for prevention of HIV in Asia" at ICMR, New Delhi on 17 September.
4. Participated in a India Chem. Exhibition 2002 hosted by Bayer Crop Science held at New Delhi on 20 September.
5. Participated in a Regional Priority – Setting Meeting Region VI: Indian Ocean Region at New Delhi, India from 18–21 September.

Dr. P.K. Mittal

1. Participated in the Technical Committee meeting for procurement of NAMP supplies at Nirman Bhavan, Ministry of Health under the Chairmanship of Addl. DGHS on 23 October and 30 December.
2. Participated in the meeting to discuss the progress of Project on Prospecting for Botanical Pesticides held at DBT, Delhi in July 2002.

Dr. B.N. Nagpal

1. Attended the meeting on "X Plan Proposals of the Country" organized by " Director-General, ICMR" on 23 November.
2. Attended Hindi debate competition on "Vegetarian v/s Non-vegetarian" organized by ICMR (HQ) on 16 September and received IIIrd prize from DG-ICMR.

Dr. S.K. Sharma

1. Presented the technical data generated on malaria epidemiology from vaccine trial site in a meeting chaired by Director-General, ICMR, New Delhi on 27 February.

Dr. Aruna Srivastava

1. Attended the meeting on "X Plan Proposals of the Country" organized by " Director-General, ICMR" on 23 November.

Dr. S.K. Subbarao

1. Attended Meeting on Review of Malaria Programme in Nirman Bhawan on 16 January
2. MMV Meeting on Medicine and Malaria Vaccine in New Delhi from 11–12 February.
3. MMV Meeting on Medicine and Malaria Vaccine in WHO on 13 February.
4. Meeting on Therapeutic efficacy of Drug Resistance at National Anti Malaria Programme on February.

5. Meeting of IDVC Issues at ICMR on 5 March.
6. Attended Meeting on Central Insecticide Board in Ministry of Health & Family Welfare on 2 April.
7. Participated in the Task Force Meeting on collaborative studies to evaluate operational impact of insecticide-treated bednets in malaria control in Chiang Mai, Thailand from 23–27 April.
8. Attended II Meeting of the Committee for " Hiring of Consultancy Services for social Marketing of Medicated Mosquito Bednets" under the World Bank assisted Enhanced Malaria Control Project at MOH&FW, Nirman Bhawan, New Delhi on 7 May.
9. Attended meeting regarding Introduction of Blister Packs for radical treatment of malaria in high risk areas in the Ministry on 20 June.
10. Participated in the Technical Committee for finalization of technical specifications for mosquito bednets to be used under NAMP and EMCP in the Office of Addl. DG Ministry of Health and Family Welfare on 5 August.
11. Attended the meeting regarding Review of implementation of NAMP and EMCP with world bank support in the Office of the Addl. Secy., Ministry of Health & Family Welfare on 21 August.
12. Attended the IX Meeting of Committee on Molecular Entomology (BCV) in Bangkok, Thailand from 4–8 September.
13. Attended Scientific Advisory Committee, RMRC, Bhubaneswar from 19–20 September.
14. Attended I meeting of Task Force on dengue at ICMR on 26 September.
15. Attended meeting at ICMR regarding RS/GIS on 11 October.
16. Attended SAC meeting at RMRC, Dibrugarh on 14-15 November.
17. Attended meeting on the X Plan Proposals of the country at ICMR, New Delhi on 23 November.

Dr. Neena Valecha

1. Attended WHO Informal consultation on development of South Asia Network for Malaria Drug resistance at New Delhi from 9–10 January as a Temporary Advisor.
2. Participated WHO meeting on "Improving access to treatment for malaria" at New Delhi on 13 February.
3. Attended Environmental Health Project through USAID for "Establishing a systematic frame work for the introduction and evaluation of Pf malaria Dipsticks in Nepal" from 12–19 August as a Consultant.
4. Participated for preparation of document on Drug Resistance for EHP, Nepal from 23–30 September as a Consultant.
5. Participated in a meeting "Research and Development on New Antimalarial drugs" organized by Confederation of Industry (CII) at New Delhi from 11–12 February .
6. Delivered a lecture on "Therapeutic efficacy of chloroquine in vivax malaria" at Navi Mumbai Municipal Corporation Office, Navi Mumbai on 5 May.
7. Delivered a lecture on "Therapeutic efficacy studies" at Meeting of WHO-TSN project, held at MRC, Delhi on 14 June.

8. Delivered a lecture on "Rapid malaria diagnosis: Dipstick and method principle, merits and field application" at Regional Course in Malariology for SEA region on 15 November.
9. Attended meeting of Expert group on "Drugs for Neglected Diseases—Indian initiative" at ICMR (HQ) office, New Delhi on 16 February.
10. Attended bulaquine meeting at Quest Institute of Life Sciences, Nicholas Piramal India Limited, at Mumbai on 7 June.
11. Attended a meeting to discuss therapeutic efficacy studies under WHO-TSN project at MRC, Chennai on 27 June as a Convenor.
12. Attended a Brainstorming session for identification of targets for drug development at Indian Institute of Science, Bangalore on 1 October.
13. Attended a meeting on X plan proposals of the country at ICMR (HQ) on 23 November.
14. Delivered a lecture on "New perspectives in diagnosis of malaria" at Delhi Chapter IAMM meeting, National Institute of Biologicals, Min. of Health & FW, Noida on 2 February.
15. Delivered a lecture on "Revised protocol for the assessment of Therapeutic efficacy of chloroquine for uncomplicated *P. falciparum* malaria" at Meeting of Pf monitoring activities at Directorate of National Anti Malaria Programme, Delhi from 5–6 March.

Dr. R.S.Yadav

1. Participated in WHO/SEARO Task Force meeting on collaborative studies to evaluate operational impact of insecticide-treated bednets in malaria control, Chiang Mai, Thailand as Temporary Adviser from 24–25 April.

PAPERS PUBLISHED

1. Biswas, Sukla. Enhancement of antimalarial activity of chloramphenicol against Indian *Plasmodium falciparum* isolates *in vitro* by chloroquine. *Indian J. Malariol.*, **39**(1-2): 26-33.
2. Dev, V. Micropylar apparatus of an egg of *Aedes* (Stegomyia) *aegypti* (L). *Bionature,* **22:** 13–15.
3. Dhiman, R.C. Eco-epidemiological types of malaria in India and need of research inputs for control strategies. In *Proceedings of the WHO Workshop on Strategies for Control of Kala-azar and Malaria,* December 27–28, 2001. Eds. S.K. Bhattacharya, N.K. Ganguly and C.P. Thakur (Balaji Uthan Sansthan, Patna): 149–160.
4. Dhiman, R.C., S. Bhattacharjee, T. Adak and S.K. Subbarao. Impacts of climate change on malaria in India. Souvenir of Joint Annual Conference of the Indian Society for Malaria and other Communicable Diseases and the Indian Association of Epidemiologists, New Delhi: 6–7.
5. Dhiman, R.C. and S.K. Subbarao. Reasons of malaria outbreaks in India and possible tools for early warning. In *Proceedings of Mekong Malaria Symposium,* December 10–13, 2002, Siem Reap, Angkor Wat, Kingdom of Cambodia. Ed. Frederick Gay (Mekong Malaria Forum, RMCP-EC): 121–122.
6. Dhindsa, K.S., U.M.X. Sangodkar and Ashwani Kumar. A novel method of screening soils for mosquito-pathogenic bacilli. *Lett. Appl. Microbiol.*, **35**(6): 457–461.
7. Dua, Virendra K., N.C. Gupta, P.K. Kar, G. Edwards, Neeru Singh and V.P. Sharma. Pharmacokinetics of chloroquine in Indian tribal and non-tribal healthy volunteers and patients with *Plasmodium falciparum* malaria. *Curr. Sci.*, **83**(9): 1128–1131.
8. Dua, Virendra K., Sukesh N. Sinha, Sukla Biswas, N. Valecha, S.K. Puri and V.P. Sharma. Isolation and antimalarial activity of peroxydisulfate oxidation products of primaquine. *Bioorg. Med. Chem. Lett.*, **12**(24): 3587–3589.
9. Escalante, Ananias A., Heather M. Grebert, Raul Isea, Ira F. Goldman, Leonardo Basco, Magda Magris, Sukla Biswas, Simon Kariuki and Altaf A. Lal. A study of genetic diversity in the gene encoding the circumsporozoite protein (CSP) of *Plasmodium falciparum* from different transmission areas—XVI. Asembo Bay Cohort Project. *Mol. Biochem. Parasitol.*, **125:** 83–90.
10. Escalante, Ananias A., Heather M. Grebert, Sansanee C. Chaiyaroj, Flavia Riggione, Sukla Biswas, Bernard L. Nahlen and Altaf A. Lal. Polymorphism in the gene encoding the Pfs48/45 antigen of *Plasmodium falciparum*—XI. Asembo Bay Cohort Project. *Mol. Biochem. Parasitol.*, **119:** 17–22.
11. Ghosh, S.K., S.N. Tiwari, A.K. Kulshrestha, T.S. Sathyanarayan and T.R.R. Sampath. Control of malaria transmission using larvivorous fishes. In *Trends in Malaria and Vaccine Research — The Current Indian Scenario.* Eds. D. Raghunath and R. Nayak (Tata McGraw-Hill Publishing Company Ltd., New Delhi): 154–158.
12. Ghosh, S.K., T.S. Sathyanarayana, M.V. Murugendrappa and S.K. Subbarao. Field evaluation of a rapid immunochromatographic test 'Parachek-F' in a post-monsoon

Plasmodium falciparum malaria outbreak in villages of south India. *Japanese J. Trop. Med. Hyg.*, **30**(1): 7–13.

13. Joshi, Hema, S.K. Subbarao, N. Valecha and V.P. Sharma. Ahaptoglobinemia (HpO) and malaria in India. *Indian J. Malariol.*, **39:**1–12.

14. Kapoor, Neera and M.A. Ansari. Laboratory evaluation of etofenprox treated fabrics against vector species of mosquitoes. *Intnatl. Pest Contr.*: 301–303.

15. Mittal, P.K., T. Adak and S.K. Subbarao. Relative efficacy of five synthetic pyrethroids against four vector mosquitoes *Anopheles culicifacies, Anopheles stephensi, Culex quinquefasciatus* and *Aedes aegypti*. *Indian J. Malariol.*, **39:** 34–38.

16. Mittal, P.K., T. Adak, O.P. Singh, K. Raghavendra and S.K. Subbarao. Reduced susceptibility to deltamethrin in *Anopheles culicifacies* s.l. in District Ramanathapuram, Tamil Nadu: Selection of pyrethroid resistant strain. *Curr. Sci.*, **82:** 185–188.

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18. Mya, M.M., R.K. Saxena and A. Roy. Sensitivity and specificity of isolated antigen from *Plasmodium falciparum* culuture supernatant. *Indian J. Clin. Biochem.*, **17:** 75–82.

19. Pandey, Kailash C., Sanjay Singh, C.R. Pillai, Usha Pillai, Andrew Lynn, S.K. Jain and Chetan E. Chitnis. Bacterially expressed and refolded receptor binding domain of *Plasmodium falciparum* EBA-175 elicits invasion inhibitory antibodies: Implications for malaria vaccine development. *Mol. Biochem. Parasitol.*, **123:** 23–33.

20. Pillai, C.R. and C. Usha Devi. Malaria parasite bank: A national resource for the control of malaria. In *Proceedings of the WHO Workshop on the Strategies for Control of Kala-azar and Malaria*, December 27–28, 2001. Eds S.K. Bhattacharya, N.K. Ganguly and C.P. Thakur (Balaji Uthan Sansthan, Patna): 175–185.

21. Raghavendra, K. Insecticide resistance in malaria vectors in India. In *Proceedings of the WHO Workshop on Strategies for Control of Kala-azar and Malaria*, December 27–28, 2001. Eds. S.K. Bhattacharya, N.K. Ganguly and C.P. Thakur (Balaji Uthan Sansthan, Patna): 161–173.

22. Raghavendra, K. and S.K. Subbarao. Case studies on insecticide resistance and its management. In *Proceedings of Mekong Malaria Symposium*, December 10-13, 2002, Siem Reap, Angkor Wat, Kingdom of Cambodia. Ed. Frederick Gay (Mekong Malaria Forum, RMCP-EC): 17–21.

23. Raghavendra, K. and S.K. Subbarao. Chemical insecticides in malaria control in India. *ICMR Bull.*, **32**(10): 93–99.

24. Ravikumar, K. S.K. Ghosh, T.S. Sathyanaryana, T.R.R. Sampath, G.R. Arunodaya, K.T. Shetty and M. Murugendrappa. Field evaluation on safety aspects of short-term community exposure of cyfluthrin 050 EW treated impregnated bednets for malaria control. *Pestology*, **26**(2): 6–10.

25. Ravindran, John, Alex Eapen and Indranil Kar. Evaluation of repellent action of neem oil against the filarial vector, *Culex quinquefasciatus* (Diptera: Culicidae). *Indian J. Malariol.*, **39:** 13–17.

26. Shukla, R.P., S.N. Sharma and S.K. Bhatt. Malaria outbreak in Bhojpur PHC of District Moradabad, Uttar Pradesh, India. *J. Com. Dis.*, **34**(2): 118–123.

27. Singh, Neeru. Malaria in primitive tribal population. In *Trends in Malaria and Vaccine Research—The Current Indian Scenario*. Eds D. Raghunath and R. Nayak (Tata McGraw-Hill Publishing Company Limited, New Delhi): 11–22.

28. Singh, Neeru, A.C. Nagpal and R.B. Gupta. Failure of chloroquine therapy in a splenectomized child infected with *Plasmodium vivax*. *Ann. Trop. Med. Parasitol.*, **96:**109–111.

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34. Srivastava, Aruna, and B.N. Nagpal. Mapping malaria. *GIS Dev.*, **4**(6): 28–31.

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ADVISORY COMMITTEES

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