# Long-lasting Insecticidal Nets and Materials

Insecticide treated nets (ITNs) have been proved efficacious in several parts of the world in reducing the mosquito densities and also reduction in malaria incidence. However, periodic re-impregnation, erratic dose of the insecticide, and other factors diluted the efficacy of ITNs in long-term. To overcome these problems, long-lasting insecticidal nets (LN) have been developed and are being promoted in many malaria endemic areas.

Long-lasting insecticidal net is 'A net treated at factory level with insecticide either incorporated into or coated around fibers resisting to multiple washes and whose biological activity last as long as the net itself (3–4 years for polyester nets, 4–5 years for polyethylene ones)'.

The LN is the most technologically advanced form of treated net available today. LNs, which maintain efficacy without re-treatment for 4-5 years, represent an important innovation that could facilitate sustainable scale-up of malaria prevention. They are constructed of special synthetic fibers (polyester and polyethylene) that have been compounded with an insecticide. The net both kills and repels mosquitoes and it provides a physical barrier to them. Tests have proven that the presence of a LN or long-lasting net material hung as curtain, also discourages mosquitoes from remaining in the surroundings. A few brands of LNs are already in use in some countries. World Health Organization (WHO) has given full recommendation to Olyset® Net and interim recommendation to PermaNet® 2.0, Duranet®, Net Protect<sup>®</sup> and Interceptor<sup>®</sup>. K-O Tab 1-2-3<sup>®</sup> and ICON<sup>®</sup> MAXX (long-lasting insecticide treatment kits were also approved by WHO.

#### LN Trials carried out by NIMR

NIMR has carried out Phase II and Phase III trials of three long lasting insecticidal nets, namely Olyset net, PermaNet, Interceptor and K-O Tab1-2-3. Trials on few LNs are underway. The results of laboratory and field trials on the above mentioned LNs are presented below.

# **Olyset® Nets**

#### **Bio-efficacy and Wash Resistance**

Results of cone bioassay tests for determining discriminatory exposure time and bio-efficacy revealed that Olyset Net produced 100% mortality in An. culicifacies, An. subpictus, An. stephensi within three minutes of exposure and after 24 hours holding. In Ae. aegypti, 100% mortality was observed at 5 min exposure, while in Cx. quinquefasciatus at 30 min exposure time and 24 hours holding. From the results 3 min was established as discriminatory exposure time for An. culicifacies, An. stephensi and An. subpictus; 5 min for Ae. aegypti and 30 min for Cx. quinquefasciatus. Cone bioassay tests on washed Olyset Nets at an interval of 24 hours up to 20 washes showed that washing did not dilute the efficacy of Olyset net as evidenced by the fact that 100% mortality was observed in An. culicifacies even



Cone bioassay on LN



Ring net bioassay on LN

after 20 washes, whereas in *Cx. quinquefasciatus* 100% mortality was observed only up to 12 washes. The results clearly shows that Olyset<sup>®</sup> Net is highly effective in killing anopheline mosquitoes even after repeated washings, which did not dilute the efficacy (Ansari *et al* 2006).

Bio-efficacy and wash resistance studies in Orissa on *An. culicifacies* and *An. fluviatilis* showed 100% mortality in *An. culicifacies* up to 11 washings, whereas 100% mortality was observed in *An. fluviatilis* even after 20 washings. The median knock down time for these species ranged between 4.55 - 6.00 min and 4.45 and 5.45 min respectively against *An. culicifacies* and *An. fluviatilis*during one year of intervention (Sharma *et al* 2007). Bio-efficacy and wash resistance studies in Assam indicated that the bio-availability of insecticide on Olyset net fibre was consistent (100% kill effect) up to 10 months of monitoring, and nets were observed to be washresistant even after 20th wash at fortnightly intervals.

#### Phase II Evaluation of Olyset Nets

Beel Akbarpur village located in the PHC Dadri, Distt. Ghaziabad, Uttar Pradesh, India was selected for the field trial. Indoor resting density of of An. culicifacies mosquitoes (MHD) in Olyset Net, untreated net and without net structures was 53.75 ± 11.2, 59 ± 20.6 and 55.5 ±23.1, respectively (Fig. 42). However, the density of An. culicifacies was reduced drastically in Olyset Net used structures. The percent reductions in An. culicifacies density in the Olyset Net used structures when compared with structures, where nets were not used during pre- and post-experiment were 94 and 95.2% based on density in untreated net and without net structures respectively. In case of Cx. quinquefasciatus and total mosquitoes the percent reduction observed over untreated net and without net structures were 47.2 and 54.1, and 50 and 57.7 respectively.

It was observed that landing rate on Olyset Net was drastically reduced. However, 100% mortality was observed in those mosquitoes which landed on the Olyset Net. Mortality was also observed in indoor resting collected mosquitoes within 24 hours, which were exposed to the net for few seconds and settled to rest on walls. The average mortality was 50.8% in *An. culicifacies*, 41.5% in total anophelines, 33.6% in *Cx. quinquefasciatus* and 44.8% in total mosquitoes. This clearly emphasizes that the Olyset<sup>®</sup> Net not only kills the landed mosquitoes but also kills the mosquitoes which enters in to the room having Olyset Net and are accidentally exposed to the net.

The results also revealed that Olyset net produced strong repellent action. Repellent action of Olyset Net was more pronounced in *An. culicifacies* as compared to total anophelines and *Cx. quinquefasciatus*. The repellent action of the Olyset Net was 55.2% in *An. culicifacies* as against 38.6% in *Cx. quinquefasciatus*. Results also revealed that excito repellency action (ERA) was almost 100% against all the mosquito species over a period of six months (Ansari *et al* 2006)

### Phase III Evaluation of Olyset Nets

Large-scale field evaluation of Olyset nets to study the entomological and epidemiological impact was undertaken in Uttar Pradesh, Orissa and Assam states. The results of these trials are furnished below.

#### **Uttar Pradesh**

A village-scale trial was undertaken from August 2003 to August 2007 in Khandera, Beel Akbarpur and Anandpur villages in Dadri PHC, Distt. Gautam Budh Nagar, Uttar Pradesh, India. Olyset nets were distributed in Khandera village, untreated nets in Beel Akbarpur and Anandpur village was kept as control, where nets were not used. Results of entomological evaluation revealed a marked difference in the indoor



Fig. 42: Mean man hour density of *An. culicifacies* and *Cx. quinquefasciatus* in structures with Olyset nets, untreated nets and without nets





resting density of the major malaria vector *An. culicifacies* in the Olyset-net village, when compared to the untreated net and without net villages (Fig. 43). The use of Olyset nets not only reduced the indoor resting density of *An. culicifacies* but also reduced mosquito entry into the Olyset net used structures.

There was no landing on the Olyset net resulting complete protection of users. Results of epidemiological evaluation in the three villages revealed that parasite incidence (Cases per thousand population) in village with Olyset net during 2003-04 (pre intervention year) was 39.5, which declined to 0.12, 0 and 2.5 during 2004-05, 2005-06 and 2006-07 in the post intervention years respectively. In the untreated net and without net villages, the cases/1000 during 2003-04 (pre-intervention) was 44 and 19 and as compared to 6.1, 3.8 and 8.8 during 2004-05, 2005-06 and 2006-07 in the untreated net village and 19.5,11.5 and 10 in without-net village (Fig. 44). Similar trend in Pf/1000 in the three villages was also observed. Random checking for the usage pattern of Olyset net during the peak transmission



Fig. 44: Malaria cases/1000 population in Olyset, untreated and without net villages before and after intervention periods

season revealed that more than 80% people are still using the Olyset nets even after three years of distribution of these nets. Results of the present study revealed that Olyset nets are highly effective in reducing the indoor resting density of *An. culicifacies*, man-vector contact and malaria incidence even after three years of use in field. Inhabitants expressed overwhelming response to Olyset nets and they did not report any adverse effects due to the use of Olyset nets. Follow-up trials on Olyset nets are still in progress.

#### Rourkela ,Orissa

A village scale-trial was conducted in Bisra block of Sundargarh district. The study area comprised 22 villages which were randomized into three clusters and designated as Olyset net cluster (8 villages, 1953 pop.), untreated net cluster (9 villages, 2019 pop.) and no net cluster (5 villages, 1863 pop.). Baseline pre-intervention data were collected from November 2005 to January 2006 followed by one year of intervention phase from February 2006 to January 2007. In the Olyset net study area, there was a significant reduction of 80.6, 94.1 and 76.7% in the entry rate of An. culicifacies, An. fluviatilis, other anopheline species respectively with an over all reduction of 63.5% in total mosquitoes. Floor sheet collections in houses with Olyset net indicated 39% immediate mortality in total mosquitoes. The personhour density of An. culicifacies and An. fluviatilis in houses with Olyset net, untreated net and no net are shown in Figs. 45 and 46 respectively.

The overall feeding success rate of mosquitoes in the trial village was only 18% in comparison to 44.2 and 79.1% in villages with untreated nets and no nets respectively. A significant reduction was also recorded in parity rate and human blood index of vector species in Olyset net area. There was a significant reduction of 65–70% in malaria incidence and 46% in malaria prevalence (parasite rate) in Olyset net area. The attack rate of *P. falciparum* in different age groups during intervention phase also showed significant reduction in Olyset net area as



Fig. 45: Density of *An. culicifacies* in houses with Olyset net, untreated net and without net during preintervention (Nov 2005 to Jan 2006) and postintervention (Feb 2006 to Jan 2007) in Orissa



Fig. 46: Density of *An. fluviatilis* in houses with Olyset net, untreated net and without net during preintervention (Nov 2005 to Jan 2006) and postintervention (Feb 2006 to Jan 2007) periods in Orissa

compared to untreated net and no net areas (Fig. 47).

The community acceptance of Olyset nets was high as there was 80–98% compliance rate of net usage in the study population during different months. The use rate of untreated nets in control villages was between 70 and 90% in different months.

#### Assam

A village-scale trial was conducted in Kamrup district of Assam from July 2005 – June 2006. It was observed that there was virtual disappearance of *An. minimus*, the mosquito vector species (Fig. 48) corroborated by data on mosquito-landing rates in Olyset net villages. A consistent decline in malaria incidence was observed in Olyset net villages, and overall impact on malaria transmission was highly pronounced and significant when compared to untreated net intervention and no net villages for the corresponding study periods (Fig. 49). The Olysetnets were safe to use for which community compliance and acceptance was high, and are operationally feasible community-based intervention for sustainable management of disease vectors.



Fig. 47: Attack rate of *P. falciparum* in different age groups using Olyset net, untreated net and no net



Fig. 48: Relative abundance of *An. minimus* in experimental villages of Sonapur PHC, Assam, India subject to introduction of Olyset<sup>®</sup> nets in August 2005

#### **PermaNets**

PermaNet<sup>®</sup> 2.0 is manufactured by M/s. Vestergaard Frandsen (India) Pvt. Ltd. The net is coated with Deltamethrin @ 55 mg a.i./m<sup>2</sup> on a polyester fabric with100 denier fiber with Warp-Knetheo weave type and 156 het/sq inch mesh size.

## Wash Resistance and Bio-efficacy

Results of cone bio-assay tests revealed that PermaNets produced 100% mortality in *An. culicifacies*, *An. fluviatilis*, *An. stephensi*, *An. minimus*, *An. philippinesis* and *Ae. aegypti* within three min of exposure and after 24 hours recovery period. In *Cx. quinquefasciatus*, 100% mortality was observed at 30 min exposure. The results clearly show that PermaNets are highly effective in producing 100% mortality in *Anopheles* mosquitoes in a short time of exposure.

Results of ringnet bioassays on unwashed and washed PermaNets showed median knock-down time (KT<sub>50</sub>) of *An. culicifacies* (Uttar Pradesh) on unwashed PermaNet was 6.5 min and it progressively increased with the number washes up to 7 min after



Fig. 49: Malaria transmission trends in experimental villages of Sonapur PHC, Kamrup district, Assam, India during pre-intervention (July 2005), and postintervention months beginning September 2005 (Olyset nets were introduced in August 2005)



Hand catch collections in LN used houses

20 washes. The KT<sub>50</sub> of An. culicifacies (Orissa) was 11 minutes on unwashed PermaNet. Similarly, KT<sub>50</sub> of An. stephensi was 8 min on unwashed PermaNet and after 20 washes, it was 16 min. The KT<sub>50</sub> of An. fluviatilis was 8 min on unwashed PermaNet. The results showed high efficacy of PermaNet against both the species as evidenced by the fact that even after 20 washes the mortality in both the species remained >80%. Further, there was no significant difference between the mortalities of the two species (p>0.05). In ring net bioassays the baseline median knock-down time against An. culicifacies is 393 sec. However, after progressive washings the median time for knockdown (MTKD) also increased. The MTKD of An. culicifacies was less than that of An. stephensi and the difference was statistically significant (p<0.05) (Sreehari et al 2007).

# **Uttar Pradesh**

### **Phase II Evaluation**

A small-scale field trail was conducted from April 2005 to November 2005 to study the efficacy of



Fig. 50: Percent reduction in per man hour density of *An. culicifacies, An. stephensi* and all other mosquitoes (collected by hand catch) in PermaNet or untreated net villages over no net village (from July to November 2005) PermaNets. Three villages, namely Nawada, Harampur and Durgawali in Loni PHC, District Ghaziabad, Uttar Pradesh, India. Depending on the prevalence of vector species PermaNets were distributed in Nawada village, untreated nets in Durgawali village and Harampur village was kept as control, where nets were not used. Nets were distributed to all the inhabitants of the two villages on 1 July 2005. The results revealed that the MHD of An. culicifacies and An. stephensi the major malaria vectors in the structures selected for PermaNet, untreated net and without net during pre distribution period was 27 and 53, 22 and 32 and, 20 and 22 respectively. After the distribution of nets the densities reduced gradually in PermaNet village. Though there was also a reduction in the village using untreated nets, the impact was less pronounced than that observed in the PermaNet village (Fig. 50).

Repellent action and excito repellent action of PermaNet against different mosquito species were calculated based on untreated nets. Results revealed that PermaNet showed high excito repellent activity (82–97%) against *An. culicifacies*, 77–97% against *An. stephensi*. Killing action of PermaNet calculated over untreated net revealed that PermaNet has high killing activities as evidenced by the fact that all the landed mosquitoes on the net died after 24 hours of observation. About 20 to 40% mortality was observed in mosquitoes entered the room and accidentally exposed to the net and rested on the walls. The results clearly demonstrate that PermaNet showed high efficacy in producing mortality in the mosquitoes, which are exposed to the net.

#### Phase III Evaluation of PermaNet

Village-scale trials were conducted with Permanets in Uttar Pradesh and Orissa states to study the efficacy of these nets on prevailing disease vectors in respective states and also on malaria incidence. Results of these trials are furnished below separately in different states.

#### Uttar Pradesh

Phase III field evaluation of Permanet 2.0 was carried out in District Gautam Budh Nagar, in western Uttar Pradesh in April 2007. Three villages with similar malaria endemicity, topography and mosquito prevalence in District Gautam Budh Nagar in Uttar Pradesh, where malaria is transmitted mainly by An. culicifacies and An. stephensi. Entomological evaluations revealed that the man hour density (MHD) of An. culicifacies during pre-intervention period in PermaNet, untreated net and no net villages was in the range of 10-13, 24-27 and 8-12 respectively. With the commencement of intervention, there was sharp decline in density of An. culicifacies in June 2007, whereas the density in the untreated and no-net villages did not decline in June 2007. However, there was an increase in the resting density of An. culicifacies in all the villages during the



Fig. 51: Impact of PermaNet on mosquito density in study villages in Distt. Gautam Budh Nagar, Uttar Pradesh during April 2007–March 2008

monsoon and post-monsoon period of August to November 2007, but the build-up of *An. culicifacies* density was much higher in the control villages as compared to PermaNet village (Fig. 51). The parity rate of *An. culicifacies* was low in PermaNet village as compared to untreated net and no net villages. The parity rate of *An. culicifacies* in June 2007 in the first month during post-intervention period in PermaNet untreated net and no net villages was 20, 66.6 and 60% respectively.

Comparison of malaria incidence data showed that during pre-intervention period of April-May, 2007, the parasite index (PI) or number of cases per 1000 population in the treated net villages was 2.5 and in the control villages with untreated nets and no nets was 1.7 and 2.9 respectively. There was no significant difference (p > 0.05) in the malaria endemicity in the trial and control villages (Fig. 52). During intervention phase, the malaria incidence in the treated net villages was much less than the untreated net and without net villages (0.84, 5.19 and 13.46 respectively). There was 85–99% compliance rate of net usage in the study population during different months.



Mosquito landing catches of the LN

# Orissa

Field evaluation of PermaNets was initiated during August 2007 in malaria endemic block of Gurundia in Sundargarh district. Density of malaria vector species *An. culicifacies* and *An. fluviatilis* was monitored in houses having PermaNets, untreated nets, and no nets in trial and two control villages respectively during pre-intervention period (August to October 2007) and intervention period (November



Fig. 52: Malaria incidence in PermaNet, untreated net ant no net villages during May 2007–April 2008 in Uttar Pradesh



Fig. 53: Density of *An. culicifacies* in houses with Perma-Nets, untreated nets and without nets during pre-intervention (Aug to Oct 2007) and postintervention (Nov 2007 to Jun 2008 phases in Orissa

2006 to June 2008). The MHD of An. culicifacies during pre-intervention period in trial villages, untreated net villages and no net area ranged from 8.5-10, 5-9.5 and 4-4.5 respectively. However, with the commencement of intervention during November, 2007, there was sharp decline in the density of An. culicifacies (Fig. 53) in PermaNets area (range: 0-1) as compared to areas with untreated nets (range: 1.5-5.5) and without net (range: 2.5-6). Similarly, density of An. fluviatilis during pre-intervention period in PermaNets, untreated net and no net area was ranging between 1-2, 0.5-2, and 2.0-11.5 respectively (Fig. 54). The impact of introduction of PermaNets in the trial villages on An. fluviatilis was so remarkable that the man hour density of this species came down to zero and remained so during most part of the intervention phase, whereas in control villages with untreated nets and no nets the



Fig. 54: Density of *An. fluviatilis* in houses with PermaNets, untreated nets and without nets during preintervention (Aug to Oct 2007) and postintervention (Nov 2007 to Jun 2008 phases

density was in the range of 0.5–1.5 and 2–9 respectively during intervention period. The density of *An. culicifacies* in PermaNets area showed a significant reduction of 89% as compared to untreated net and no net area respectively. Similarly, a reduction of 100% was recorded in the density of *An. fluviatilis* in houses with PermaNets in comparison to houses with untreated nets and no net respectively.

The parasite rate or percent of individuals positive for malaria parasite in PermaNets, untreated net and no net population during pre-intervention was 6.8, 5.8 and 7 respectively and no significant difference was observed between all the thee clusters (p >0.05). During intervention phase the parasite rate in PermaNets users had come down to 2.9, whereas in untreated net and no net users, the parasite rate was 3.4 and 15.1 respectively (Fig. 55). Comparison of parasite rate in PermaNets net villages during intervention with that of pre-intervention period showed that there was 57% reduction in malaria prevalence, which was statistically significant (p<0.01). Malaria prevalence in untreated net area also showed a significant reduction of 41.4% in comparison to pre-intervention phase, whereas there was an increase of 115.7% in no net villages.

# Interceptor<sup>®</sup> Net

#### Wash Resistance and Bioefficacy

Interceptor nets have shown wash resistance up to observed 20 washes. Bioavailability of insecticide on net fibers was found effective against malaria vectors, *An. culicifacies* and *An. fluviatilis* in Orissa; *An. culicifacies* in Chhattisgarh; and *An. minimus* in Assam. Exposure of mosquitoes in Sundargarh district and Kamrup district registered 100% mortality in cone bioassays, while in Chhattisgarh, 100% mortality was registered in first 2 washes and in subsequent washes up to 20 times, the mortality



Fig. 55: Malaria prevalence in PermaNet, untreated net and without net villages during pre-intervention (Aug to Oct 2007) and post-intervention (Nov 2007 to Jun 2008) phases recorded through crosssectional surveys in Orissa



Delivering instructions on the use of LNs to the users

remained >85%. The studies on wash resistance indicated good wash resistance of interceptor nets and also bioefficacy against important malaria vectors (WHO criterion: >80% mortality in 3 min cone bioassays).

#### **Field Evaluation**

Field evaluation of Interceptor<sup>®</sup>, alphacypermethrin-treated long-lasting insecticidal nets (LNs), was undertaken to assess entomological and epidemiological impact in malaria endemic villages of Sundargarh district (Orissa), Kamrup district (Assam) and Kanker district (Chhattisgarh). Three clusters of villages in each study area were selected. In one cluster of villages Interceptor nets were distributed to all the inhabitants, in the second cluster, untreated nets were distributed and the third cluster was kept as control where nets were not distributed.

The interceptor nets were found to be effective in controlling the indoor resting mosquitoes. There was a decline in the overall entry rate of mosquitoes in houses with interceptor nets at all the three study sites coupled with reduction in feeding success rate. In Sundargarh district, a significant reduction up to 87.5% was registered in entry rate of mosquitoes during intervention compared to pre-intervention (p<0.01). There was virtual disappearance of An. minimus in villages with Interceptor nets, but were found in untreated net and without net villages in Assam. In Kanker district, Chhattisgarh, the entry rate of mosquitoes (40.3) in interceptor used houses was comparatively lower than the untreated net (151) and without net (136) houses. The results suggested good performance of Interceptor nets in reducing the mosquito density and also preventing entry of mosquitoes into the houses with interceptor nets.

The impact of Interceptor nets on malaria incidence in Sundargarh district indicated 57–76% reduction compared to untreated net (p<0.01) and without net villages (p<0.001). Mass blood surveys in the study villages in Sundargarh district indicated similar results as that of the active surveillance. In Sundargarh, the parasites rates in the interceptor net





used villages reduced (73%) after the intervention over the pre intervention period, in the untreated net villages also 40% reduction was observed, but in the without net villages the parasite rates increased during the intervention phase than the preintervention period (Fig. 56).

In Assam, villages with interceptor nets registered drastic reduction in *P. falciparum* cases during intervention period. The total cases observed in Interceptor net used villages during the intervention period (October 2006 to April 2007) were 10, while in untreated net villages 82 cases and without net villages 233 cases were reported during the intervention period (Fig. 57).

In Kanker district, Chhattisgarh the slide positivity rate (SPR) during pre-intervention period was 8.4, 6.9, and 3.2 in Interceptor net, untreated net and without net villages respectively, whereas after intervention the SPR was 3.7, 6.5 and 11 respectively. The results indicated the impact of Interceptor nets in reducing the malaria transmission. Similarly, malaria incidence (Cases/000) reduced from 36.3 during pre-intervention to 14.7 post intervention in



Fig. 57: Malaria prevalence in different age groups in Interceptor net, untreated net and without net villages in Kamrup district, Assam



Recording of perceptions on the use of LNs from the inhabitants

Interceptor net villages, while it increased from 15 in pre-intervention to 23.1 in post-intervention in untreated net villages and from 34.2 to 42.9 in without net villages in respective periods (Fig. 58)

The interceptor nets were found to be safe for human. The community compliance and acceptance was high . No adverse health events were reported by the users. Surveys carried out in Orissa indicated 98.2% compliance, in Assam it was 100% and in Chhattisgarh it was 81.7%. All the users in the three study areas informed that there is reduction in mosquito bites (74–90%) with effect on other nontarget domestic insects like cockroaches, house flies, spiders, etc. Studies carried out to assess the human safety didn't indicate any contraindications owing to net use and the nets were found to be safe for use by inhabitants of all age groups.

The results of multicentric field evaluation in three malaria endemic states in India revealed good performance of Interceptor nets in reducing the mosquito densities and malaria incidence.

#### K-O- Tab 1-2-3

This is a deltamethrin formulation developed by M/s. Bayer Environmental Science. This is specifically meant for converting untreated nets into long-lasting insecticidal nets. The formulation consists of



Fig. 58: Malaria prevalence in Interceptor net, untreated net and without net villages in Kanker district, Chhattisgarh deltamethrin tablets and a binder; upon impregnation it converts the net into a long-lasting net.

A project on the evaluation of bioefficacy and wash resistance of K-O Tab 1-2-3 tablets supplied by M/s. Bayer India Ltd. was undertaken. Nylon mosquito nets were impregnated with the water dispersible tablet formulation of deltamethrin

(K-O Tab 1-2-3) @ 25 mg/m<sup>2</sup> dose + binder and washed as per the study protocol. Bio-efficacy of the insecticide treated mosquito nets was evaluated against *An. fluviatilis, Ae. aegypti* and *An. stephensi* using WHO cone bioassay test. The bioefficacy studies showed 100% knockdown of *An. fluviatilis* up to 30 washes. K-O Tab 1-2-3 nets were 100% effective against *An. fluviatilis* even after 15 washes. Thereafter, percent mortality recorded was 92, 92, 84.6 at 20, 25 and 30 washes, respectively.

Ring net bioassay test was also carried out to determine the median knockdown time ( $KDT_{50}$ ), 90% knockdown time ( $KDT_{90}$ ) and percent mortality. There was 100% mortality in wild caught full-fed *An. fluviatilis* in 1 hour exposure and 24 hour recovery period up to 30 washes. The KDT<sub>50</sub> and KDT<sub>90</sub> were 11, 15; 12, 18; 14, 23; 14, 25; 18, 25; 18, 26; and 20, 28 min at 0, 5, 10, 15, 20, 25, and 30 washes respectively. KDT<sub>50</sub> and KDT<sub>90</sub> value of *An. fluviatilis* against 5% deltamethrin impregnated paper were 9 and 14 min respectively.

Further studies on the bioefficacy of K-O Tab 1-2-3 were conducted to examine wash resistance against *Ae. aegypti*. There was 100% knockdown in laboratory reared *Ae. aegypti* up to 15 washes at 1 hour observation and thereafter between 85 and 92% knockdown was recorded in subsequent 20 to 30 washings. Percentage mean mortality of mosquitoes was 31% (27.2–33%) up to 15 washes at 24 hour recovery period. Thereafter, percent mean mortality was 13.8% (13.6–14.3) up to 30 washes.

# Long-lasting Insecticide Incorporated Plastic Sheets

Spraying of canvas tents with residual pyrethroid insecticide is an established method of vector control in refugee camps. In recent times plastic sheeting (polythene tarpaulins) has replaced canvas as the utilitarian shelter material for displaced populations in complex emergencies. Advances in technology enable blankets to be impregnated with pyrethroid during the process of manufacture. The efficacy of such fabrics against mosquitoes when used for protection in different ecological conditions needs evaluation. Presently the anti-malaria campaigns include insecticide based intervention measures both against larvae and adult mosquitoes. Prevention of malaria is a major technical and operational problem in displaced and mobile populations. This has necessitated a continued need by the programme managers to evaluate new efficacious strategies to control vectors in complex emergencies. Insecticide incorporated blankets is a new technology to control

mosquitoes in emergency shelter places, temporary habitations in different locations, slum settlers, tribal population, etc. The blankets can be used for covering the body for protection against mosquitoes and other insects. The results of the trial will definitely provide the usefulness of the blankets in controlling the mosquito nuisance and also disease prevalence. In India no such trial was undertaken on insecticide incorporated blankets. To tackle complex emergencies it is desirable to evaluate insecticide incorporated blanket in different ecological conditions in India to provide relief to distress people affected by natural calamities or living in remote and unhygienic environment.

#### ZeroFly

ZeroFly is an insecticide incorporated plastic sheet with deltamethrin @ 265 mg ai/ m<sup>2</sup> manufactured by Vestergaard Frandsen India Pvt. Ltd. This was field evaluated in Delhi and Orissa states for its efficacy. The results of the trial are furnished below.

# Delhi

This study was initiated in the month of Aug 2006 in Labour camps in Delhi and Noida. In both the localities ZeroFly plastic sheets were fixed at a distance of at least one km from the control localities where plastic sheets without insecticide (untreated) were fixed. In addition to the two localities the study was also carried out in RAC police camp in Delhi. Bioassay tests on ZeroFly sheets with 3-min exposure period resulted in 100% mortalities in An. culicifacies and An. stephensi. The effect of ZeroFly sheets persisted at 100% mortality against An. culicifacies even after one year of use under field conditions. Fortnightly monitoring of entomological parameters showed almost complete reduction in the indoor resting density of vector and non-vector insects in the labour camps provided with ZeroFly plastic sheeting as compared with the camp provided with untreated plastic sheetings. Entomological and epidemiological parameters were monitored at fortnightly interval. Fortnightly monitoring of entomological and epidemiological parameters have shown considerable reduction in the indoor resting density of vector and non-vector insects in the labour camps provided with ZeroFly plastic sheeting, as compared with the untreated plastic sheeting. In labour camp at a construction site in Noida, man hour density (MHD) of malaria vectors An. culicifacies and An. stephensi during post intervention period was in the range of 0-05 in the experimental area as compared to 0-12 and 0-15.5 in the control area. Similarly in JJ cluster of agricultural labor in the Jamuna belt area in Delhi, man hour density of malaria vectors An. culicifacies and An.stephensi were in the range of 0-05 in the experimental area as compared to 0-13 and 0-15 in the control area. Parasite incidence (cases per thousand population)



Tent line with ZeroFly plastic sheeting

in the construction site labour camp in Noida was 37 in the experimental area as compared to 73 in the control area. Similarly, in the JJ cluster in Delhi the PI in experimental and control areas was 42 and 62.7 (Fig. 59). In RAC police camp, intervention with Zerofly sheeting revealed in the reduction of culicine density in the experimental tents as compared to the control tents. Survey about the perception of the users about side effects versus and benefits revealed a highly positive response in favour of the benefits of Zerofly sheeting.

#### Orissa

A field trial was conducted on the efficacy of ZeroFly<sup>®</sup> plastic sheeting against malaria vectors *An. culicifacies* and *An. fluviatilis* and its impact on malaria transmission in one of the highly endemic areas of Orissa—Birkera block of Sundargarh district. The study area comprised three villages which were randomized into three clusters and designated as ZeroFly plastic sheet (330 pop.), untreated plastic sheet (372 pop.) and no plastic sheet area (382 pop.). Baseline pre-intervention data were collected from July to September 2006 followed by nine months of



Fig. 59: Number of malaria cases reported in ZeroFly and untreated plastic sheets used localities in Noida, Uttar Pradesh during pre-(Jul 2006) and post-(Aug 2007–Jul 2008) intervention periods

**EPIDEMIOLOGY** 



Fig. 60: Density of *An. culicifacies* in houses with ZeroFly plastic sheets, untreated plastic sheets and without plastic sheets during pre-intervention (Jul- Sep 2006) and post-intervention (Oct 2006 to Jun 2007) periods in Sundargarh district, Orissa



Temporary shelter camp made with ZeroFly plastic sheets

intervention phase from October 2006 to June 2007. Baseline studies showed that both the vector species An. culicifacies and An. fluviatilis were 100% susceptible to deltamethrin. Cone bioassay tests were performed on An.culicifacies and house flies every month during intervention period on the ZeroFly plastic sheets while in use in the field conditions and these species were exposed for 3 and 30 minutes respectively. The results showed a knock-down effect of 95-100% and 90-100% on these species respectively during different months of intervention. 100% mortality was recorded after 24 h of recovery period in both the species. In ZeroFly plastic sheeting area, there was a significant reduction of 86.7, 100 and 84% in the entry rate of An. culicifacies, An. fluviatilis and other anopheline species respectively with an over all reduction of 84.7% in the entry rate



Fig. 61: Month-wise malaria cases/1000 in study villages during pre-intervention (Jul-Sep 2006) and postintervention (Oct 2006 to Jun 2007) periods in Sundargarh district, Orissa

of total mosquitoes in comparison to pre-intervention phase. The density of malaria vector *An. culicifacies* in houses with ZeroFly plastic sheeting, untreated plastic sheeting and no sheet is given in Fig. 60.

Floor sheet collections in houses with ZeroFly plastic sheet indicated 56.2% immediate mortality in total mosquitoes. The overall feeding success rate of mosquitoes in the trial village was only 12.5% in comparison to 49.7% and 51.1% in villages with untreated plastic sheet and no sheet respectively.

The excito-repellent rate of all the mosquitoes in houses with ZeroFly plastic sheets and untreated plastic sheets was 40.6% and 3.1% respectively. The month-wise parasite index in the trial and control areas is shown in Fig. 61. There was a significant reduction of 65% and 70.5% in malaria incidence in ZeroFly plastic sheeting area as compared to untreated plastic sheet and no sheet area respectively. The ZeroFly plastic sheeting was found to be safe for human and no adverse health events were reported. The community acceptance was high as 91% of study population was sleeping in the rooms fixed with ZeroFly plastic sheeting. The ZeroFly plastic sheeting provided relief not only from mosquitoes but also from other household pests, such as bed bugs, cockroaches, ants and houseflies as revealed by the study population. The study showed that ZeroFly plastic sheeting may be used as an effective intervention strategy that is operationally feasible to control malaria, particularly in remote high risk areas and also during complex emergency situations.