ENTOMOLOGY IN DRDO AND DEVELOPMENT OF COMMERCIALLY VIABLE PRODUCTS FOR PROTECTION TO INDIAN ARMED FORCES

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Armed forces are exposed to different climatic regions and so to a variety of pests and disease vectors during their field operations, thus likely getting affected with various arthropod-borne hazards as well as diseases. Therefore, a proper protection mechanism is required for soldiers. In our continuous efforts for safe and effective methods of protection from pests and vectors, various products have been developed for personal and area protection for the armed forces especially during field operations. These products are also useful for the civilian.

Recently a chemical known as N’ N’ diethyl phenyl acetamide (DEPA) has been developed as a multi-insect repellent for protection from mosquitoes and other biting insects. DEPA is now commercially available in various formulations such as spray, lotion, wet wipe (tissue paper), cream, and vaporizer. Attracticide traps based on an oviposition pheromone for control of Aedes aegypti, vector of dengue and chikungunya, and other similar traps are in advanced stage of development. These traps are highly safe and economical. Herbal based new products are also developed for mosquito and leech control which are found quite effective against these creatures. They are in the form of spray, cream and vaporizer and are at various stages of commercialization.

DRDO has also developed long lasting insecticidal net (LLIN or LN) named as ‘Defender Net’ which is effective even after 20 washing against adult mosquitoes of Anopheles, Culex and Aedes groups. Besides, DRDO has also developed other products of public health importance such as Roachtox for cockroach control and Roachline for control of crawling insects particularly in kitchens, bakeries, restaurants etc. A slow release insecticidal paints (named SRIP) is also developed for control of pests in various structures.
Rodents cause serious damages to a variety of material in armed forces particularly in the Indian Navy; having thus notorious earned the sobriquet of ‘High tech pest’ as they damage important electronic equipments in ships, vessels etc. For rodent control, very effective bait, Ratox is developed. This bait is based on ‘lure & kill’ principle to overcome the problem of bait shyness (avoidance of poisonous food due to associated learning) and neophobia (avoidance of novel stimuli) among rodents and often responsible for failure of rodents’ control.

Woollen textiles are frequently damaged by the insects during their storage and sometimes cause colossal losses. A new formulation developed is named ‘Wool Care’ which can be used on the woolen articles before their storage.
SERO-PREVALENCE OF CERTAIN VECTOR-BORNE AND ZOONOTIC DISEASES IN AND AROUND VELLORE DISTRICT OF TAMIL NADU, INDIA

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Vector Borne Diseases such as Malaria, Filaria, Dengue, Chikungunya, and Japanese encephalitis are highly prevalent in many Tropical and Subtropical Countries. They cause great threat among the public in the developing Countries in terms of high morbidity and mortality which would imbalance the socioeconomic status of the Country. In general, early detection of the Cases and prompt treatment is the current strategy to prevent the Disease spread and prevent the outbreaks.

The laboratory based surveillance system has been proved to be an effective and supportive tool for forecasting the Disease outbreaks on several occasions. In India, until 1990s, the laboratory diagnostic facilities were available only in National Research Laboratories, hence several outbreaks were not reported in time. Thereafter during late 1990s, the Sero-diagnostic facilities were available only at the state referral laboratories such as King Institute of Preventive Medicine, Chennai and the Institute of Vector Control and Zoonoses (IVCZ), Hosur in Tamil Nadu.

Considering the need for establishing the diagnostic facilities to expedite the investigations of vector-borne Disease outbreaks such as Dengue, Chikungunya and JE, besides Sentinel Surveillance Hospitals in Govt. Medical Colleges, 9 Zonal Entomological Teams (ZET), have been equipped with Serodiagnostic facilities where the National Institute of Virology (NIV)-Pune kits are used.

In this paper, it is aimed to document the occurrence of vector-borne disease pattern in and around Vellore district of Tamil Nadu.
During 2010 – 2012, serum samples collected from clinically suspected patients were received from Vellore and Thiruvanamalai Districts for diagnosis of Dengue, Chikungunya, Leptospirosis, and Rickettsial infections. Results of the serodiagnosis of the samples received from the above Districts during 2010 to 2012 (up to August) showed that there were differences among the trend of the disease Pattern.

The analysis of the Sero Positivity data at Zonal level showed that the prevalence of Dengue is in decreasing trend in Vellore, Thiruvanamalai, HUDs whereas, prevalence of Chikungunya Cases was found to be in increasing trend in Vellore and Cheyyar districts, and decreasing in Thirupattur and Thiruvannamalai Health districts.

The sero prevalence of Zoonotic Disease, Leptospirosis, showed decreasing trend in Vellore, Thiruvanamalai, Cheyyar and increased in Thirupattur. The rickettsial Diseases showed an increasing trend in Thiruvanamalai and Cheyyar HUDs and decreasing trend in Vellore.

Besides these serodiagnostic facilities developed at the Zonal Entomological Team level, the flow of results to various levels facilitates immediate action for controlling the outbreaks and enabling the District health authorities to prevent any possible outbreaks in future by mapping out their areas with prevalence of these diseases.

The vector-borne disease pattern along with the networking of various health functionaries in and around Vellore District is discussed in detail in the context of establishment of effective surveillance system in Tamil Nadu. The four Health District wise last three years results were discussed in this paper.
DEVELOPMENT OF SPATIAL INFORMATION SYSTEM TO CONTROL VECTOR BORNE DISEASES (VBDs) – A CASE STUDY

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The present study investigates the vector habitat distribution and surveillance of vector borne diseases in Sitheri hills and develops a control strategy using remote sensing and GIS. The study area is situated in Dharmapuri district of Tamil Nadu in Eastern Ghats. The area is remotely located with poor basic amenities in transportation, public health care facility, lack of health awareness etc. The studies of vector-borne diseases (VBDs) have emerged as an important issue in the present context. Epidemiological and entomological data were collected from 16 villages for spatial analysis of vector habitat distribution using the IDW (Inverse Distance Weighted) and Kriging Interpolation techniques. The spectral analysis was carried out using multispectral IRS P6 LISS satellite 23.5 m resolution imageries for mapping of Normalised Vegetation Index (NDVI) and vector density. The thematic layers like vegetal density, road network, settlement, drainage, population density and primary health centres, water quality and mosquito distribution were prepared using Arc Map 11 and ERDAS IMAGINE 11. Based on the entomological survey and spatial analysis, the abundance of predominant species are Cx. tritaeniorhynchus (JE), An. Subpictus (malaria), Cx. vishnui (JE), Cx. intula (JE), Cx. fuscocephala (JE), Cx. quinquefasciatus (Filaria), St. albopicta and St. aegypti (chikungunya & dengue). Out of 16 villages, Ammapalayam, Perperi, Pudur and Selur, are sensitive areas in Sitheri hills. Using Remote Sensing and GIS, we are able to identified the risk area and also forecast the onset of the disease by correlating the epidemiological and entomological data with satellite data. The study highlight the importance of spatial technology can be very useful in remote areas.

Key Words: Vector habitat, ArcMap 11, ERDAS IMAGINE 11, Interpolation, satellite data, NDVI
LABORATORY DIAGNOSIS – THE ONLY TOOL FOR OUTBREAK INVESTIGATIONS

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Outbreak investigation has become a great challenge for public health professionals, due to the emergence and reemergence of diseases of public health importance which are dynamic and complex because of overlapping clinical presentations of several vector borne and zoonotic infections in the community. Concurrent infections make the outbreaks more complex. An outbreak occurred in Nagapattinam District is being reported here.

Increase of fever cases was reported in three villages around Vedaranyam, a town 40 Kms away from the District Headquarters during the month of November-2010. The initial impression of the fever outbreak was suspected as Dengue, as 11 patients were suspected to have suffered from Dengue based on Thrombocytopaenia and other clinical presentations observed by a private medical practitioner. As the clinical profile was varied, intravenous blood samples were collected from fever patients to confirm the aetiology. 61 clinical samples collected from one of the three villages affected, and from the government hospital Vedaranyam which is the nearest institution for referral were screened for Dengue, Chikungunya, Leptospirosis, Rickettsial infections and Typhoid.

Considering the clinical presentations, 55 samples were screened for Dengue and Leptospirosis, of which 5 (9.1%) were positive for Dengue and 16 (29.1%) for Leptospirosis. 28 samples were screened for Chikungunya and Scrub typhus. One sample each (3.6%) was positive for chikungunya and scrub typhus. Typhoid was screened for 16 samples and all were negative. All the blood slides collected (110) were negative for malaria parasite.

Dengue positives detected (5 Nos) were distributed in 3 villages. Sixteen Leptospirosis cases reported were distributed in 9 villages. Chikungunya
and Scrub Typhus positives (one Each) were distributed in two different villages. Of these 14 villages one village reported concurrent infections of Dengue (2), Leptospirosis (5) and Scrub Typhus (1).

The outbreak emphasizes the need for employing laboratory diagnostic tests irrespective of clinical presentations to screen diseases like Dengue, Chikungunya, Scrub typhus, Leptospirosis, Typhoid and Malaria to establish the etiology to prevent mortality and to reduce the morbidity.
OUTBREAK OF URTICARIAL RASH IN TIRUVARUR DISTRICT, TAMILNADU

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During the second week of August 2012, an outbreak of urticarial rash was reported from Tiruvarur District of Tamilnadu. Approximately 200 residents of village kankoduthavanitham seeking care for itching and varied rashes of unknown etiology on the first day of outbreak. The outbreak was explosive affecting 1,083 from 8 villages of two Health subcentres with a total population of 12,625 in PHC - Kankoduthavanitham, a rural pocket near the District Headquarters, Tiruvarur. The outbreak lasted for a week with a decline of cases from day 5 as the urticarial rash responded to antihistamines and in some it was self limiting.

The age group of patients affected ranged from infants (6 months) to 50 years of age and the rash did not clearly associate with, sex or occupation among affected persons. But definitely, there was an association with the increase of affected persons with an increase of age group. (0-6 = 24 // 1-5 = 126 // 6-8 = 180 // 9-14 = 322 // 15 & above = 431). The outpatients attended the hospital had exanthematous and maculo papular rashes, with severe itching without fever for 2-3 days.

After initial investigation of case reports and consultation with a Dermatologist and Paediatricians, the cases were suspected as a cause of either insect bite or viral exanthema of enterovirus group (coxasackie virus) as the clinical presentation was exanthematous rash and itching, in addition to macules and papules, resembling also the foot and mouth disease.

The insect bite was ruled out, as none of the patients reported insect bite, on enquiry. And other common skin rashes like Diaper rash, cradle cap, prickly heat, Roseola, Erythema, chickenpox, scarlet fever, Impetigo, Hives,
Lyme disease, and scabies were ruled out clinically by the clinicians who attended the patients.

Though the reported history of the patients helped to characterize the duration, onset, relationship to various environmental factors, (Rainfall) skin symptoms (such as itching, rash and pain), the clinical profile was without fever for the first 3 days among the patients. But 158 out of 1083 (14.6%) had fever on 4th day after the itching and rash started settling. All the 158 fever patients were negative for malaria parasite.

Hence the possibility of infectious agents that can cause a rash which include virus, bacteria, fungi and parasites, were also considered. Clinical samples (serum) sent to Institute of Vector Control & Zoonoses, Hosur were Negative for Dengue, chikungunya, leptospirosis and scrub typhus, (7 samples). All the samples (10) sent to King institute, Guindy, were negative for enteroviruses. But 2 serum samples out of 5 (40%) were positive for Leptospirosis sent for screening of Leptospirosis. The only positive result from the laboratory diagnosis strongly suggests, the fever outbreak might be due to Leptospirosis, though cannot be conclusive due to smaller sample size. But as reported by S.Faine, et al., in Leptospira and leptospirosis “A transient, petechial or punctate red palatal enanthem occurs in the first day or two and exanthematous rashes, are recorded in 10-30% of patients in the first week lasting 1-2 days. The appearance of the rashes is usually either morbilliform or macular, or urticarial, and occasionally purpuric and confluent.”. So Leptospirosis may also be considered among other serological tests when patients present with rashes for a day or two followed by fever.
The larvae of *Aedes aegypti* were collected from field and reared under laboratory conditions having temperature 25 ± 2°C and relative humidity 85 ± 5%. Mosquito larvae collected from the culture, washed several times with sterile distilled water, surface sterilized larvae were placed on nutrient agar plates. The inoculated plates were incubated at 37°C and after 24 hours, colonies obtained on the plates were identified by their morphological characteristics, using gram staining method. The Bacterial species obtained from gut as the mosquito larvae was characterized using partial 16S rRNA as *Bacillus thuringiensis* (Bt) strain IAM 12077 with zero (0.0) E-value. The effect of Bt on the larval development have been studied. And the results indicated that mosquito took 12 days to complete their life cycle and turn into adult. Whereas low dose and high dose treated larvae were not able to form adult. Adult 100% mortality after 192 h.p.i obtains with high dose and 95% with low dose, compared with 55% in control. The result therefore suggest that the bacterium isolated from the gut of *A. aegypti* is pathogenic and could be used in the field to control vector of disease after further screening in the laboratory.

Keywords: - *Aedes aegypti*, *Bacillus thuringiensis*, 16S rRNA
A QUALITATIVE STUDY TO IDENTIFY BARRIERS TO MALARIA CONTROL AMONG TRIBAL POPULATION OF MALARIA-ENDEMIC GADCHIROLI DISTRICT, INDIA: IMPLICATIONS FOR NATIONAL VECTOR BORNE DISEASE CONTROL PROGRAM (NVBDCP)

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Malaria is a major public health problem in India. Tribal people, constituting nearly 10% of Indians account for 50% of all malaria deaths in India. Our study sought to identify barriers to malaria control as perceived by the government health workers and officers implementing NVBDCP in tribal areas of malarial-endemic Gadchiroli district. This district is heavily forested and 37% of the population is tribal. We conducted focused group discussions and in-depth interviews with community health workers, medical officers stationed at primary health centers in tribal areas, and district malaria and health officers. Although programme components were perceived to be comprehensive, many operational gaps and cultural barriers were identified. For malaria prevention, lack of knowledge about malaria, lack of culturally appropriate education material, limited use of bed nets and a perceived ineffectiveness of insecticides against malaria by villagers were the major barriers. Active surveillance was limited by remote location of villages and inadequate frequency of village visits by the health workers. For curative care, delay in diagnosis due to initial care seeking from traditional healers, medication non-compliance, poor follow up of malaria treatment by ASHAs due to inadequate remuneration were the major barriers. Improving tribal community’s knowledge about malaria by providing culturally appropriate health education to village adults as well as children in ashram schools, making traditional healers a partner in malaria control, improving treatment follow up by ASHAs, promoting use of bed nets could be some of the strategies to decrease malaria prevalence in this region.
ROLE OF ASYMPTOMATIC CARRIERS AND WEATHER FACTORS IN PERSISTENT TRANSMISSION OF MALARIA IN ASSAM - ARUNACHAL PRADESH BORDER, INDIA

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A yearlong study was carried out on the incidence of symptomatic and asymptomatic malaria carriers and the role of asymptomatic carriers in persistence transmission was studied. Relationships between malaria incidence and weather factors were also investigated. Blood surveys were conducted on monthly basis in Bengenajuli, Sapairawmari Pathar and Nigam areas near the Assam-Arunachal Pradesh border. Epidemiological indices were estimated for symptomatic and asymptomatic malaria. Multiple linear regressions between malaria incidence and temperature and relative humidity along with rainfall were carried out. Known malaria vectors collected in light traps were identified. SPR and Pf% for symptomatic malaria were 26.10 and 79.77 respectively. Prevalence of malaria vectors was observed throughout the year with varying densities. An. philippinensis/nivipes and An. annularis were predominant among the seven malaria vector species recorded. Monthly total rainfall with one-month lag was found to have the highest correlation (r = 0.919) with SPR. The relationship between SPR and weather factors was established as SPR = -114.219 + 0.579 T_min + 1.380 RH + 0.032 RF (R^2 = 0.888 p = 0.000). Low and relatively constant levels of asymptomatic malaria carriers were observed throughout the year. High vector density and presence of asymptomatic carriers of malaria parasite in the community are the cause of persistent malaria transmission in this region. Therefore, detection and prompt treatment of symptomatic and asymptomatic carriers of malaria parasites are essential to interrupt the disease transmission. Weather factors such as rainfall, temperature and relative humidity can be used for forecasting malaria incidence in this region.

Key words: Malaria, asymptomatic carriers, weather factors, transmission.
ASSESSMENT OF COVERAGE AND COMPLIANCE OF MASS DRUG ADMINISTRATION UNDER ELIMINATION OF LYMPHATIC FILARIASIS PROGRAMME IN THIRUVARUR, NAGAPATTINAM AND THIRUVANNAMALAI DISTRICTS, TAMIL NADU, SOUTH INDIA

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Lymphatic filariasis (LF) is one of the neglected tropical diseases and is caused mainly by the filarial worm, Wuchereria bancrofti, through the bites of mosquito, Culex quinquefasciatus. Globally, over 120 million people are infected, with 40 million incapacitated. In India, the National Programme for Elimination of Lymphatic Filariasis (NPELF) aims to achieve the target by 2015. In view with the global elimination of LF through Mass Drug Administration (MDA), a single annual dose of diethylcarbamazine (DEC) and albendazole was given for the all eligible population in endemic areas of Tamil Nadu by department of public health, Tamil Nadu during May 2012. It is essential to have a good coverage and compliance by the people for the success of MDA program. Hence, coverage and compliance of MDA was assessed in three endemic districts namely, Thiruvarur, Nagapattinam and Thiruvannamalai. A cross-sectional coverage evaluation survey was carried out. Based on the drug coverage, one urban and three rural clusters were selected in each district. A pre-tested semi-structured proforma was used to collect data to assess the coverage and compliance of MDA along with possible determinants for non-
attaining the expected coverage. The survey revealed out of 2074 eligible for MDA but the drug was received by only 1547 individuals. It was observed that the overall coverage was 69.74%, 80.13% and 73.53% in Thiruvarur, Nagapattinam and Thiruvannamalai districts respectively, while the compliance was 71.60%, 66.16% and 86.22%, in these three districts. Fear of side-reactions and non-availability of the drug-receiver in town were identified as the major reasons for non-compliance. Since it is very much essential to improve compliance in future rounds of MDA to achieve targets of control and eventual elimination of LF, we suggest that more attention needs to be given to the drug distributors, through training and by providing them with IEC (Information Education Communication) materials, in order to ensure a high coverage and compliance within a reasonable time frame.
IN SILICO PREDICTION OF HIGHLY INTERACTING PEPTIDES OF GANODERMIN WITH THE ENVELOPE PROTEIN OF DENGUE VIRUS SEROTYPE 2

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Currently 40% of the world’s population are at risk from dengue. WHO estimates that the global burden of dengue infections may be 50–100 millions every year. Dengue virus serotype 2 (DENV2) is more likely to cause severe disease compared to other closely related yet antigenically distinct serotypes (DEN-1, DEN-2, DEN-3 and DEN-4). Though, many antiviral drugs (small molecular drugs) are available most of them have their own adverse effects. So there is a great need of novel and more efficient antiviral agents. Ganodermin, a 15 kDa antifungal oligopeptide of Ganoderma lucidum, was selected and 20 octapeptides has been derived randomly. An in silico approach has been made to predict the potential use of ganodermin derived octapeptides against dengue serotype 2 envelope protein in the present study. Molecular docking was performed using GLIDE Tool (Schrodinger Maestro). The interaction between protein-protein is mediated mainly by hot-spots, regions that account for 80% of the binding energy which plays a vital role in the disruption of functional protein-protein complexes. Better interaction between the envelope protein (DEN-2) and peptide (THTVMINH) was obtained with the energy value of 9.958 kcal/mol. These findings suggest that the peptide fragment “THTVMINH” can be used as an alternative antiviral drug for DEN-2 infection. An in vitro study may help to improvise the efficacy of the identified fragment through cell line analysis.
TEMPERATURE EFFECTS AND MALARIA RISK;
OCCUPATIONAL EXPOSURE IMPLICATIONS FOR WOMEN
IN KEONJHAR DISTRICT, ODISHA, INDIA.

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An estimated 2.7 million people die of malaria and in pregnancy causes
200,000 newborn deaths each year worldwide. District surveillance data of
Keonjhar showed Plasmodium falciparum (pf) > 93%, Annual parasite incidence
(API) >17 and deaths 10% of the state (Keonjhar 23 and Orissa 239) in
2008.

The district consist of total 17,84,000 population and out this 7,93,884
(44.5%) belong to tribal community with 53% male and 47 % female. The
study showed, major vectors of malaria in Keonjhar district were An.culicifacies,
An.fluviatilis & An.minimus. There are many other factors which favor malaria
transmission in Keonjhar such as high temperature 16-40°C (cause of
shortening of gonotrophic cycle of the vectors and extrinsic incubation period
(EIC) of the parasites), excess rainfall (1500 mm) & rise of humidity
(60-80%), spread of drug resistant parasites, prevalence of insecticide resistant
mosquitoes, large forest coverage (34%), hilly terrain with rivulets, huge mining
activities, lack of treatment seeking attitude in people, existence of myth and
miss-conceptions, low referral from the inaccessible areas and migration of
population. Those at highest risk biologically are infants and young children,
pregnant women, non-immune people from non-endemic areas. Particularly
pregnant women are four times more vulnerable. Further the occupational
exposure of women in dawn and dusk is more risky as they work outside
and perform household, mining, forest and agricultural activities which is the
peak biting time of many mosquito vector species.
Thus it addresses study on climate resistant vectors and their bionomics which will help to build an entomological model for different geophysiographic situations to predict the malaria risk through climate change analysis, gender disaggregated data on human epidemiology and it needs collaboration across ministries of health, environment and finance at local, national and regional level.
AN INSIGHT INTO GLOBAL MALARIA CONTROL:
PROGRESS IN RESPECT TO ACHIEVING MILLENNIUM
DEVELOPMENT GOALS (MDG6)

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In the year 2000 world leaders committed to eight Millennium Development Goals (MDGs) ranging from halving extreme poverty and hunger to rolling back killer diseases like AIDS and Malaria. The deadline of 2015 to achieve these ambitious targets is now rapidly approaching, hence it is an opportunity to have a look on progress made in this front and devise future control strategies. MDG 6 specifically calls for halting and reversal of the incidence of malaria by 2015. As per the recently released UN MDG Report (2012) and World Malaria Report (2011), there were 216 million malaria episodes of malaria with 0.65 million deaths around the Globe in 2010 of which 81% were in Africa. It is evident that the estimated global incidence of malaria has decreased by 17% since 2000 and malaria specific mortality rates have declined by 25%. Reported malaria cases fell by more than 50% between 2000 and 2010 in 43 of the 99 countries with ongoing malaria transmission. The decline in Global malaria cases was due to escalation in efforts and intensify the key interventions of malaria control like increase in the use of insecticide treated bed nets (ITN), artemisinin based combination therapy (ACT), Indoor Residual Spray (IRS) and intermittent preventive treatment (IPT) in some areas. It was noted that percent of children sleeping under ITN in sub-saharan Africa is estimated to have grow from 2% in 2000 to 39% in 2010. Person protected from IRS rose from less than 5% in 2000 to 39% in 2010 and there was increase in the use of ACT as well. In India also there has been significant improvement as number of malaria cases declined from 2.08 to 1.56 million during 2001-2009 and Plasmodium falciparum (Pf) cases from 1.04 to 0.84 million. Annual Parasite Incidence (API) also declined from 2.12 per 1000 to 1.36 in 2009 and Slide Positivity Rate (SPR) from 3.22 in 1995 to 1.41 in 2010. However, there is a belief that cases are highly under reported and there is a need to measure the true burden of the disease. In
Sri Lanka, malaria incidence has declined by 99.9% since 1999 and malaria is now at low levels and only 124 indigenous cases were reported in 2011 and Sri Lanka has set a goal of eliminating malaria by the end of 2014. Malaria map continues to shrink and in 2011 one more country Armenia was certified to be free from malaria. Despite, of all these successes still there is a challenge in terms of emerging threat of resistance against ACTs in Thai-Cambodia border, vector resistance to the commonly used insecticides including Pyrethroids and the climate change related factors. Though there is a ray of hope with the development of new drug molecule like Synriam launched recently and the work being done in the area of parasite and vector biology & genomics. The need of the hour is to intensify the efforts to sustain the gains and keep the momentum going to reach final destination.
BIOLARVICIDAL EFFECT OF PHYTO-SYNTHESIZED SILVER NANOPARTICLES USING *PEDILANTHUS TITHYMAILOIDES* LEAF EXTRACT AGAINST *Aedes aegypti* L. (DIPTERA; CULICIDAE)

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Mosquitoes transmit dreadful diseases to human beings wherein biological control of these vectors using plant-derived molecules would be an alternative to reduce mosquito population. Aqueous leaf extract and green synthesized silver nanoparticles (Ag NPs) from *Pedilanthus tithymaloides* (L.) Poit were investigated for their efficacy against the developmental stages of dengue vector *Aedes aegypti* L. (Diptera; Culicidae). The biologically synthesized Ag NPs were characterized by UV-Vis spectrum where responsible peak were observed at 460 nm and X-ray diffraction (XRD) analysis showed the crystalline nature of Ag NPs were produced in the reaction mixture. Responsible compounds were recorded using Fourier Transform Infrared (FTIR) and 15-30 nm size produced Ag NPs were characterized by Atomic Force Microscopy. Further, on exposure of the larvae and pupa to varying concentrations of aqueous leaf and Ag NPs for 24 h, these Ag NPs showed 100% mortality from first to fourth instars and pupae of *Aedes aegypti* at 0.25%, which is the highest concentration, tested; wherein, it was the lowest concentration of aqueous leaf extract alone which showed only 10-18% of mortality. Lethal concentration (LC50) values of Ag NPs against the larval and pupal stages were 0.029, 0.027, 0.047, 0.086 and 0.018%, respectively with no mortality in control. These results suggest that the use of *Pedilanthus tithymaloides* silver nanoparticles can be a rapid, cost effective, recyclable and environmentally safer bio-larvicides which can form a novel approach to develop effective bio-pesticides for controlling the target vector.
TITLE: DENGUE EPIDEMIC IN KOLKATA: CRITICAL BUT NEGLECTED BREEDING SITES PLAYING AN IMPORTANT ROLE.

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Background: Dengue continues to remain a disease of concern in West Bengal. This year Kolkata and its’ adjoining areas suffered from a massive clutch of the disease. The case burden increased more than double of the expected number.

Aims: 1. To study the vector habit and habitat. 2. To search for the cause of sudden increase of cases.

Method: 1. Search for the vector and the breeding sites by entomological survey. 2. Interviewing the residents and the health workers in the affected region.

Result: Breeding sources were abundant throughout the affected region. The residents of the housing complex were careful to remove the indoor breeding spots but they overlooked the immediate surrounding environment, as most of them were unaware of the flight distance of the vector. The breeding spots were found in the common shared areas e.g garbage vat, gardens, plastic cover over shanties etc.

HI, BI and CI were found to be high. Breeding spots were found also at various construction sites e.g metro rail project site, and public places e.g bus terminus, temples courtyard etc. The identified vector was Aedes albopictus. Unexpected breeding places such as polluted water, place with plenty of sunlight (not shaded, as usual breeding habitat) were also observed.

Conclusion: Many of the breeding sites were too reclusive to be covered by vector control workers. Strong community participation is warranted at such sites. Awareness generation activities should be accordingly planned. Control measures are necessary in major public places. Authorities of construction sites should be made responsible.

Key Point: Dengue, Vector, Aedes, Breeding habitat.
URBAN DENGUE SURVEILLANCE IN SURAT CITY, INDIA 2005-2011:– ENTOMOLOGICAL AND VIROLOGICAL TRENDS AND THE IMPACT OF VECTOR INTERVENTION.

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Dengue is the fastest reemerging arbovirus having the potential causing large scale outbreak caused by a bite of infected mosquito of Aedes Aegypti in urban, peri urban and rural areas of tropical and subtropical countries. There has been a theatrical increase in disease since last three decades. This study is retrospectively analysis of disease burden, restraints and impact assessment of integrated vector management in urban area of Surat city.

A structure engendered in Surat city that 488 surveillance worker deployed for vector control along with these activities on source reduction campaigns and administrative expenditure charges (Rs.48.5 lakhs in 2011) were recovered from defaulters who creating the mosquitogenic condition in their own premises. School children are made aware including their teachers about the life cycle of mosquitoes by arranging demonstration of it and they are taught to identify the mosquitoes breeding and how to destroy them in the area of their vicinity. For the duration of 2005 to 2011 of mean highest Aedes aegypti per ten man hour density 2.02 (10PMD) were observed in August; whenever, during the period 2005 to 2011 mean highest density showed in 0.92 (10PMD). Additionally, passive surveillance activities were carried out by 39 Urban Health Centre (UHC) in conjunction with three major Government hospitals working along with 105 private hospitals.

Gujarat had witnessed first dengue outbreak from Surat city during 1967, presence of DEN-2 virus was detected. As city with fast growth rate, had 4.8 million(2011) of dense population and favorable climatic
condition like temperature, humidity and rainfall, dengue fever has been reported throughout the year. A total of 1445 cases including 2 deaths were reported within 2005 to 2011, giving an incidence of highest found in October and a case fatality rate of 0.14%. There was no significance ratio in sero-positivity rate among male compared to female, however, contribution of dengue incidence predominant in male 990 (68.5%) and 445 (31.5%) were found in female. The overall Aedes premises index were not correlate with the incidence of dengue, although the density of Aedes was significant. The predominant dengue serotype during 2005 to 2011 was dengue virus DEN-2 which remerged with a clade replacement in the early 1967.

Key contributing factors to the worldwide resurgence of dengue rise in number and size of densely populated urban cities that are conducive for the spread of the disease and the adaptation and proliferation of dengue vectors.

Key words: Dengue, incidence, intervention, impact and vectors.
CURRENT SCENARIO OF JE INFECTION


Japanese B Encephalitis, a mosquito borne viral infection, commonly prevalent in areas, where pigs which act as amplifier host are intensively raised. It has been observed that JE infection occurs primarily among children in various studies throughout the world. To know about the incidence of JE infection in Madurai, the CSF and serum samples of clinically suspected patients admitted in GRH, Madurai and confirmed by JE IgM capture ELISA were studied retrospectively, with respect to age, sex and immunization.

By analyzing the study samples, the incidence of JE infection was found to be 1% [6/678] in 2009, 1% [5/556] in 2010, 0.4% [2/482] in 2011 and 3.4% [10/296] in 2012 (up to June). Further analysis revealed that in 2009, 100% of cases belong to paediatric age group and in 2010 only 50% were in paediatric age group. In 2011 both the positive cases were adults and in 2012 (up to June) out of 10 JE positives, 60% belonged to paediatric age and only 40% belonged to adult age group.

Vaccination for JE with JE SV 14-14-2 vaccine had been carried out in and around Madurai in 2009 for all children less than 15 years of age. This had resulted in decrease in the incidence and shift from pediatric to adult age group in 2010&11. The analysis of statistics for 2012 up to the month of June revealed that the incidence is on the rise and paediatric group are more affected among the positives. Vaccination will be an effective tool to control the resurgence of JE infection in view of increase in incidence and more number of positives in pediatric age group.
RAPID DEVELOPMENTAL ACTIVITIES AND RURAL LABOUR MIGRATION; PREVALENCE OF MALARIA IN BHUBANESWAR CITY, ODISHA, INDIA

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The relationship between migration and malaria are complex. In the malaria equation parasite/vector/people; parasite and vector have received much greater attention than have people. The everyday aspects of people and their lives are fundamental importance in malaria and its control, among these aspects migration is on great significance.

A study was under taken on labour migration in Bhubaneswar [the capital of odisha and an up-coming city (since 1960)] showed that 94% migrants were from high endemic (SPR>5) areas (Ganjam, Gajpati, Keonjhar, Angul and Dhenkanal) and highest percentage of migrants were from Ganjam (51%); Only 6% of migrants were from low endemic areas (Khurda, Nayagarh and Mayurbhanj). 90% of migrant labours visited their native places once in every three months. Rapid developmental and industrialization activity in capital attracts the labours in one hand and in other hand causes warming trend of the city and is an alarming state for it on the point of malaria transmission and risk.

The to and fro population moment between Bhubaneswar and known high malaria endemic areas may be a reason for prevailing malaria in the city. There should be some surveillance mechanism to access the malaria parasite infection load in migratory labour population and emphasis should also be led to know the infection load in women labours, as gender differences in access to and control over resources limit women’s ability to access health care for themselves and simultaneously, vector population and effect of warming trend of environment on parasite sporogony should be taken into account for urban malaria control programme.
CO-CIRCULATION OF DENGUE AND CHIKUNGUNYA VIRUS IN TAMIL NADU – A REPORT ON THE RECENT TREND

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Dengue and Chikungunya have caused severe epidemics in several tropical countries over the past 2 decades. The two viral infections are currently a global scale threat. Dengue fever caused by flavivirus in the family flaviviridae, is the most prevalent arboviral disease in tropical and subtropical regions of Asia. Chikungunya caused by an alphavirus in the family Togaviridae and both diseases have similar clinical symptoms including fever, rash and joint pain as well as head ache, nausea, vomiting, arthralgia and myalgia. The main vectors are mosquitoes of the genus Aedes usually Aedes aegypti. Both dengue and chikungunya virus infections has been frequently noted in the 4 southern states of India [Tamil nadu, Kerala, Karnataka and Andhra Pradesh]. All the four serotypes of dengue virus have been circulating in Tamil nadu. As the circulation of more than one serotype of dengue viruses in an area are said to be a risk factor for the appearance of the life threatening DHF, monitoring of viruses is very essential in order to implement control strategies. During May 2012, a sudden increase in fever cases was reported in the government hospital and adjoin private clinic in Tirunelveli and Kanyakumari districts, the patients admitted with fever, myalgia and arthralgia. To find out the etiology of the fever in the outbreak, a rapid sero-epidemiological study was carried out.

A total of 106 serum specimens were collected from Tirunelveli [58] and Kanyakumari [48]. These samples were analysed for dengue and chikungunya specific IgM antibodies separately by using MAC ELISA kits supplied by National Institute of Virology, Pune. Out of 106 sera tested 32 [30.18%] had dengue IgM antibodies and 16 patients [15.10%] had chikungunya IgM antibodies. Among this two patients showed dual infection had both dengue IgM and chikungunya IgM antibodies. Further a routine surveillance network is necessary and that will probably answer many questions in this area.
PURIFICATION AND CHARACTERIZATION OF KERATINASE FROM CHICKEN FEATHER DEGRADING BACTERIUM (BACILLUS THURINGIENSIS SEROVAR ISRAELENSIS) USEFUL FOR MOSQUITO CONTROL IN PUBLIC HEALTH PROGRAM

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Human industrial activities, inevitably, generate industrial wastes, consisting of, inorganic and organic materials, discharged from factories and food processing industries. Degrading or handling these wastes, as unused disposals, without acquiring any additional benefits has led to an idea to develop a suitable technology to utilize bio-organic wastes. Chicken feathers have been discarded in bulk as waste from poultry industries, globally. We have reported earlier that Bacillus thuringiensis serovar israelensis (Bti) can be used for biodegradation of feather waste for biopesticide production. In the present study, purification and characterization of keratinase from feather degrading bacterium (Bti) was carried out. Protein precipitate obtained at 60% ammonium sulfate saturation and Sephacryl S-200 column purification showed the highest activity of 2.3 and 11.68-fold respectively. The purity of the enzyme was revealed by SDS-PAGE as a single band of molecular weight of 40 kDa and it was further characterized bio-chemically. The optimum pH of the enzyme shifted to a more neutral range (6.0-8.0) with the highest activity at pH 7.0. The optimum temperature of the activity was determined to be 30 °C. The keratinase enzyme retained 51% residual activity (303 U/mg protein) at 70 °C (60 min) and the half-life of the enzyme was 130 minutes at 40 °C, 90 min at 50 °C and of 60 min at 70 °C, respectively. Keratinase activity was enhanced by calcium and magnesium ions while EDTA, PMSF, β-mercaptoethanol and manganese inhibited the activity. This is the first report of investigating the keratinase produced from Bti that degraded chicken feathers for the synthesis of mosquitocidal toxins.
BIODIVERSITY OF ANTI MALARIAL PLANT IN NORTH-EASTERN INDIA: A CASE STUDY

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Disease Control Programmes in India have shown considerable improvements in the country but Malaria continues to be a major challenge. Malaria is one of the most serious human diseases in the tropics and subtropics, with around 225 million cases occurring worldwide each year (WHO, 2010). According to a 2006 health report, over 54 percent malaria deaths in the country were reported from the northeast region with an area of 255,511 km², about 7 percent of India's total area. The area has a total population of 44.98 million (census 2011) about 3.7 percent of India's total population. North eastern India, which is rich in forests and wildlife, has an especially high malarial incidence, with over five reported malaria cases per thousand people. This high case is due to continued political unrest and ethnic violence, and difficult terrains make essential health and development services inaccessible. The present study highlight the important use of plant base for treatment of malaria as it is the only source for treatment in remote areas where there is no medicinal facilities. A survey was carried out and 100 plant species belonging to 48 families are used by the people of Northeast India for the treatment of malaria. Species including Polygala persicariaefolia, Vitex peduncularis, Alstonia scholaris, Coptis teeta, Crotolaria occulta, and Ocimum sanctum have been commonly used in these states. Leaves, Roots, bark and whole plant are the most frequently use part for the treatment. The study will help in planning conservation, sustainable use and future pharmacological research work for treatment against malaria.

Keywords: Anti malaria plant, Malaria, North eastern India, survey, pharmacological research
NOTES OF LESSONS FROM DENGUE OUT BREAK IN THIRUNELVELI, TAMIL NADU, INDIA

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Dengue is an acute viral febrile frequently presenting with head ache, joint or bone and muscular pain, rash and leucopenia as symptoms. It is transformed into DHF (Dengue Hemorrhagic Fever) and DSS (Dengue Shock Syndrome) with hemorrhagic phenomena and plasma leakage with volemic shock. This present study revealed the control measures undertaken during the major Dengue outbreak in Thirunelveli (Lat 80 42N Lon 77 042E), Tamilnadu, India in May 2012. Since the entire District had been reported cases, Blocks of the district was taken as unit for implementing all containment measures under the supervision of one Senior Entomologist. The outcome was a great success within its expected time in the block Papakudi, it is highlighted here.

The Block Papakudi is 81082 populations. It has two Primary Health Centers (PHC) Maruthamputhur and Papakudi. All of the sudden, fever cases had been raised among out patients during the first week of May 2012 and the laboratory confirmation was made with Ig-M ELISA in the Medical College Hospital, Thirunelveli. Since then, the following line of containment measures implemented. 1. Soon after ascertaining the Dengue vectors as Aedes aegypti and Aedes albopictus, anti larval and anti adult measures had been employed in entire block. 2. To concentrate the most vulnerable villages, daily information on fever cases from PHC, GH (Government Hospital), Private Clinics, Out Reach Medical services and sentinel centers were taken as resources and made them as baseline data collection. 3. Since lives claimed from pediatrics groups in the outbreak, serovar identification was sought and ascertained that DEN-1 and DEN-3 were circulating in the community. 4. Although all probable cases referred and clinical management was in progress, magnitude of the problem was ascertained by the Aedes indices, House Index(HI), Container Index(CI) and Breteau Index(BI) and inferred that Entomological surveillance was the promising tool to ascertain the intervention during the outbreak. More will be discussed in this research paper.
MALARIA: A GROSSLY NEGLECTED VECTOR BORNE DISEASE IN INDIA

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As on today almost 100 countries in the world are endemic for malaria. More than 2 billion population is in the risk of malaria. It is estimated that there are 300-500 million new cases in the world each year. 90% of global malaria burden is in Africa. 1.5 million to 2.7 million deaths are estimated every year in the world of which 1 million are among children under 5 years in Africa. Despite the fact that the overall malaria remained stationary, malaria continues to be a major public health problem in Bangladesh, Bhutan, India, Indonesia, Myanmar, Nepal, Sri Lanka, Thailand and DPR Korea. Almost 85% of SEA region are at risk of malaria. Even though the reported number of malaria cases is on decline from 3.6 -3.0 million the proportion of P falciparum cases has not shown any change. The reported and estimated number of malaria deaths decreased from7.1 to 4.1 thousand. Malaria is a major public health problem in the country. 95% of the population resides in malaria endemic areas. In India, epidemiology of malaria is complex because of the geo ecological diversity, multi ethnicity and wide distribution of the nine anopheline vectors. As per NVBDCP records in most of the states the API was less than 2 where as 2-5 API was in scattered regions and API more than 5 was recorded in the states of Rajasthan, Gujarat, Karnataka, Goa, MP, Chattisgarh, Jharkand and North Eastern states.
ECTOPRASITIC INFECTIONS IN HUMAN LIFE

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Tick and Mites, Bedbugs, Fleas are the arthropods which cause some harmful infections and diseases in human beings as well as in domestic animals. Ticks are close relatives of mites. Many scientists feel that ticks evolved from mites into parasitic associations with animals during the time of the large reptiles. Mites, along with ticks, are small arthropods belonging to the subclass Acari (also known as Acarina) and the class Arachnida. The study of ticks and mites is called acarology. A tick is a small, blood-sucking mite. Ticks are ectoparasites (external parasites) living by hematophagy on the blood of mammals, birds, and sometimes reptiles and amphibians. Several species of hard ticks are significant human disease vectors (or carriers) and are responsible for the spread and increase of Lyme disease and the persistence of Rocky Mountain spotted fever (RMSF). Mites are tiny arthropods, usually less than 1 mm in size. Many live freely in the soil or water, but there are also a large number of species that live as parasites on plants, animals, and some that feed on mold and they cause diseases like human itch or scabies mite (Sarcoptes scabiei), Hair Follicle mites. Fleas are small, wingless bloodsucking insects order Siphonaptera with a characteristic jumping movement. They feed mainly on mammals but also on birds. The most important species are the rat flea, the human flea and the cat flea. The rat flea is important as a vector of bubonic plague and flea-borne typhus. Cat fleas incidentally transmit tapeworms. The sand flea or jigger burrows into the skin of humans and may cause infections. The term “bug” is slang for insect. Used technically, however, it refers to the thousands of species of the order Hemiptera. Bedbugs, which can be found in beds or furniture, feed on humans to obtain blood-meals and they are not important in the transmission of diseases, although they possibly play a role as vectors of hepatitis B virus.
STUDY ON THE STATUS OF LYMPHATIC FILARIA
TRANSMISSION IN TWO VILLAGES OF WARDHA DISTRICT
IN MAHARASHTRA STATE

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A pilot study was undertaken in the month of November 2011 to find
the transmission of lymphatic filariasis and vector status in two villages, Hingni
and Gohada of Wardha district in Maharashtra state. These villages are situated
besides the Bar dam which is a part of the Bor wildlife sanctuary. The blood
smears were collected from 08:30 pm to 11:00 pm and the entomological
collection was undertaken early in the morning between 06:00 am to 8:30
am. The Hingni village is densely populated with 1010 individuals and has a
very poor sanitation infrastructure, while Gohada village which is just 2.5 km
away is sparsely populated with 335 individuals and has a decent sanitation
infrastructure. At Hingni village, 88 samples from volunteers of different age
groups were collected and screened for microfilaria carrier and from Gohada
village, 90 samples were collected and screened for the same. At Hingni
village, four samples (2 male and 2 female) were found to be positive for
W. bancrofti (4.54%) whereas in Gohada village, not a single sample was
found positive. Although, highest prevalence of vector Culex quinquefasciatus
was found in both the villages, the infection rate of the vector was found to be
10% in Hingni and 0% in Gohada village. Proper sanitation and sparse
population is found to be the main cause for the difference in the filariasis
transmission scenario of the two neighbouring villages.
BIOMEDICAL BIBLIOMETRICS ON CHIKUNGUNYA LITERATURE FOR INDIA - A TOOL FOR DETERMINING THE QUALITY OF SCHOLARLY ARTICLES FOR DISEASE INFORMATION

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Biomedical bibliometrics is a tool to measure scientific publications on biomedical subjects. By content analysis, citation and impact factor analysis, the quality of scholarly articles is determined. In practice, much of the work that falls under bibliometrics involves various types of citation analysis, which looks at how scholars cite one another in publications which link between scholars and the development of areas of knowledge over time. In the context of this toolkit, bibliometrics are also one of the key ways of measuring the impact of scholarly publications which is determined in part by the number of citations to articles. The publishing profile of the author is determined through publications through in high impact factor (IF) journals. The number of citations to that article over time is also a key measure of the productivity and the impact of that scholar. The values of the contents in articles published by authors are justified by Citation analysis.

Based on Chikungunya literature in India from the year 1964 to till date, published articles include 337 Numbers in 138 journals. The literature in recent years on this disease are all concentrating to publish in peer reviewed high IF journals. During this period, more than 6 articles published per journals are given in descending order as Indian J Med Res(38), Virol J (11) Commun Dis (10), Emerg Infect Dis(9), Am J Trop Med Hyg(8), Trans R Soc Trop Med Hyg(8), J Vector Borne Dis (7), Parasitol Res(7), Acta Virol(6), Indian Pediatr(6) and Southeast Asian J Trop Med Public Health(6). Other articles are published mainly on clinical aspects. On subject wise analysis more articles are clinical oriented. Diagnostic methods, pathogen study on chikungunya are concentrated...
in most of the journals. Very few articles are concentrating on mosquito vectors (i.e) *Aedes aegypti* (or) *Ae. albopictus*. Hence, this bibliometry study is indicating and suggesting to authors to concentrate publishing more articles on vector biology, vectors bionomics and disease transmission. Few articles are published in high IF journals like *Lancet* and *Lancet Infectious Diseases* and most of the articles are published in peer-reviewed journals with low IF. This will help the authors to concentrate publishing their articles in International high IF peer reviewed journals.
TIME SERIES ANALYSIS MODELS FOR PREDICTION OF DENGUE INCIDENCE

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Dengue fever (DF) and Dengue Hemorrhagic fever (DHF) substantial burden to a family in terms of the loss of life and economic impacts. Monitoring and predicting dengue incidence facilities early public health responses to minimize morbidity and mortality. Increasing dengue incidence has been attributed to climate change; however, contradicting reports show inconclusive relationships between dengue and climatic factors. This article discusses about the two models of time series analysis such as Poisson Regression model and Auto Regressive Integrated Moving Average (ARIMA) model for prediction dengue incidence. The Poisson Regression model based on monthly weather variables and monthly notified cases of dengue fever and Auto Regressive Integrated Moving Average (ARIMA) model based on past and present dengue cases, climate and seasonal factors (rainfall, temperature and relative humidity) as inputs to predict feature. These models are useful tools for monitoring dengue incidence. Furthermore, these models can be applied to surveillance data for predicting trends in dengue incidence.
THE SUCCESSION OF THE VECTOR BORNE DISEASES REPORTED FROM THE SMALLEST UNION TERRITORY OF INDIA LAKSHADWEEP ARCHIPELAGO

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Union Territory of India Lakshadweep archipelago is the smallest Union Territory of India which is a blessed territory comprised of 36 islands with only ten inhabited islands. The statuses of vector-borne diseases reported here have changed drastically over recent years. If the status of the vector borne diseases in the Lakshadweep islands were reviewed from the beginning, it was endemic for bancroftian filariasis and malaria. Filariasis was endemic practically in all the islands, primarily as a socioeconomic problem related to the coir industry, as coir soakage pits heavily bred Culex quinquefasciatus mosquito, the vector of Wuchereria bancrofti. Malaria in these islands was unstable and sporadic, depending upon the presence of the vector species. The filariasis infections declined due to the introduction of DEC-mediated salt therapy during 1976-79 and the large-scale introduction of mosquito larvivorous fishes, such as Gambusia affinis and Poecilia reticulata during 1980. Malaria cases also declined due to the effective implementation of the National Anti-Malaria Programme (present NVBDCP), ensuring early detection and prompt treatment of all malaria cases through active and passive surveillance. During 1997-99, only four imported cases of Plasmodium vivax were reported from these islands. The data collected during March 2000 from Agatti and Kavaratti islands revealed the presence of five mosquito species, viz. An. stephensi, An. varuna, Cx.quinquefasciatus, Ae. aegypti and Ae. albopictus. The brief survey of two islands, viz. Agatti and Kavaratti, revealed the presence of Anopheles stephensi, a known vector of urban malaria in India. Earlier on the mainland, the southward peninsular spread of An. stephensi, including in Kerala, a non-malarious state, was recorded and the species was reported to be involved in two major outbreaks of malaria. It
appears that over the years *An. stephensi* has invaded the Lakshadweep islands and gained a permanent foothold on these islands due to the availability of a large number of community and rain-harvesting cement storage tanks. Afterwards the studies conducted during 2006 in the chikunguya inflicted Andrott and Kalpeni islands of the Lakshadweep islands showed the dominant role played by the *Ae. albopictus* in this area. These diseases have changed and the primary causes of change are the ecological and environmental change which is the key impacts & primary causes for establishment of the succession of selected vectors and thus the transmission of particular mosquito borne infections in these islands. The succession pattern of the vector borne diseases recorded so far from these islands showed the epidemiological significance in a small island situation over a long time is discussed.
Japanese encephalitis is currently causing much public health concern as it has assumed epidemic proportions in several parts of the country. JE mostly affects the rural poor and underprivileged sections of the society. Recently during July-September 2011, JE cases were reported from four different districts - Jalpaiguri, Darjeeling, Dakshin Dinajpur and Cooch Behar of North West Bengal region. Thus a detailed entomological sampling of vector mosquitoes was undertaken in the affected districts to bring out the knowledge about the emergence of multi-vector activity responsible for this episode to suggest the effective vector control strategies to combat JE. A detailed review of the entomological sampling undertaken in the Northern West Bengal region was analyzed. *Culex vishnui* subgroup was recognized for many years as the major vectors and play an important role in the epidemiology of JE in India. *Culex gelidus* was the predominant mosquito in the dusk collections and profusely breed in many habitats from these areas which may have an important role in zoonotic cycle for the maintenance of this virus. *Mansonia* species, breed in ponds from these areas. More numbers of Anophelines collected from these areas added a new dimension and complexity to the natural history of the disease in India. The virus tends to spill over into human populations when infected mosquito populations build up explosively. It is also required to plan and implement measures for reducing man-vector contact at the village block level for JE prevention and control. To reduce the man-vector contact the vector control intervention methods like long lasting insecticide nets and insecticide treated curtains can be distributed and at the same time space spraying and indoor residual spraying can be undertaken to overcome this problem. A longitudinal follow up study throughout the year should be undertaken to bring to limelight the presence of few more species from Northern West Bengal region.
COMPARATIVE EVALUATION OF ANTI-MOSQUITO ACTIVITY OF ENICOSTEMMA AXILLARE AND AGERATUM CONYZOIDES EXTRACTS AGAINST CULEX SPS.

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Mosquitoes are single group of insect, which act as a vector of human diseases like Malaria, Dengue fever, Filariasis, Chikungunya, Japanese Encephalitis and yellow fever that causes millions of mortality and morbidity among the human being every year. The control of mosquito is facing a threat because of the emergence of resistance to synthetic insecticides and leading to environmental pollution. The present study aims to investigate the anti-mosquito (insecticidal) activity of petroleum ether, chloroform and methanol extracts of two selected plants Enicostemma axillare and Ageratum conyzoides against Culex species. A detailed study was done to monitor the effect of plant extracts on different instars larvae and pupae of Culex species. The insecticidal activities of different concentrations (5, 10, 15, 20mg/l) of extracts were treated to find out the median lethal concentration (LC₅₀). Both the plants under study possess improved insecticidal properties which control the vector in a green approach. The larvicidal efficacy of solvent extracts may be due to the presence of phytochemicals in the extracts. Further, isolation of active constituents from solvent based extraction to determine the insecticidal efficacy will pave way to treat mosquitoes in near future.

Key Words: Anti-mosquito, Culex, lethal concentration, Insecticides, Phytochemical.
LABORATORY EVALUATION OF FRACTIONS OF TRICHODERMA REESEI AGAINST CULEX QUINQUEFASCIATUS AND ANOPHELES STEPHENSI

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Present investigation evaluated the efficacy of fractions T2 of Trichoderma reesei against Culex quinquefasciatus and Anopheles stephensi. The LC$_{50}$ and LC$_{90}$ value of the T2 against selected mosquito vectors were 8.73, 9.87ppm and 26.61, 26.84ppm accordingly. Histopathological observations unveiled the degeneration as well as alteration of midgut epithelial cells of the Anopheles stephensi. The highly virulent fraction F3 was characterized based on the Rf value (0.57) and retention time (16.13) of the GC-FID and was identified as T2 toxin, a trichothecene derivative of Trichoderma sp.

Keywords: Trichoderma reesei, Culex quinquefasciatus, Anopheles stephensi, GC-FID, LC$_{50}$, LC$_{90}$. 
MYSTERIOUS FEVER OUTBREAK, TIRUNELVELI DISTRICT
TAMIL NADU, INDIA, 2009-2010

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Background: An increased number of cases of fever with arthralgia were reported from Kadayanallur municipal area of Tirunelveli district in Tamil Nadu during Dec 2009-Jan 2010. As the cause of the explosive outbreak was not immediately identified, the health officials labelled it as ‘Mysterious fever’. We investigated the outbreak to identify the cause of the fever, identify main vectors of the disease, describe the distribution of disease with respect to time, place and person and propose recommendations for control measures.

Methods: We sample surveyed Kadayanallur Municipality covering 2699 people in 400 households. We trained local health workers for the survey. We used a semi structured questionnaire and collected details of fever, date of onset, other associated symptoms, treatment and residual illness. We collected blood sample for antibody titer in acute case of fever. We carried out entomological survey and evaluated household, container and breteau indices. We also reviewed the data on platelet counts for fever patients attending the district hospital and one private hospitals in the area.

Results: Out of 2699 people surveyed, 461 (15.6%) had fever with arthralgia. The outbreak was started on 27.10.2009, peaked on 2nd week of January with 31 cases on 10th Jan 2010. The outbreak subsided after 21st January. Persons aged 45 years or more (20.3%) and females (18.8%) were most affected. The major symptoms included fever with arthralgia (100%), swelling of joints (7.4%), rash (4.6%) and bleeding (0.2%). Sixteen out of thirty one (52%) blood samples of acute fever cases were positive for IgM antibody for Chikungunya. All the three entomological indices were more than 5. During the initial phase of the outbreak, a large number of patients treated at public as well as private hospital were advised platelet counts.
Conclusion: The outbreak of mysterious fever with arthralgia in Kadayanallur was due to Chikungunya. It predominantly affected the adult population and females. Health authorities initiated vector control measures to contain the outbreak. We also recommended informing the etiological diagnosis of chikungunya to health providers in the area and advising them to do the platelet counts only in cases when clinically indicated.

Key words: Chikungunya, *Aedes aegypti*, Tamil Nadu,
NEW DIMENSION IN THE EMERGENCE OF A MEDICALLY IMPORTANT Aedes Species in the Context of Urban Dengue Epidemiology

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The WHO estimates that almost 2.5 billion people in the world are at risk of dengue infection. As compared to nine reporting countries in the 1950s, today the geographic distribution of dengue includes more than 100 countries. In Tropical countries, transmission of dengue infection can be perennial albeit it is increased in the monsoon. Historically, dengue has been considered predominantly as an urban disease and the primary vector of the disease is *Aedes aegypti*. *Aedes albopictus* is serving as secondary vector/maintenance vector but in the state of Kerala, this species was identified in the recent past as the primary vector. Climate change is one of the defining challenges of the 21st century. Dengue is a climate sensitive disease and so are the mosquito vectors. The overall distribution of *Aedes* mosquitoes is rapidly changing in this globalised world. *Aedes albopictus* was first described from Bengal and now has dispersed globally into new territories previously occupied by *Aedes aegypti*. Presently, Dengue is no longer a typical urban disease. Rural districts are also affected by this virus in India including in the state of West Bengal. Keeping in view the above perspectives, an attempt has been made to know the prevalence of these two potential vectors of dengue virus in different ecological zones of Calcutta and adjoining suburbs. Both the *Aedes* species were encountered on human bait in different ecological situations. In our earlier studies *Ae. albopictus* was found only in and around some urban gardens. But in a recent survey (2011), this species was found in meagre numbers (0.15 PMHL) alighting on human in the urban area, indicating its ecological plasticity and adaptability in the urban environment. Detection of *Ae. albopictus* larvae along with the *Aedes aegypti* in the containers placed in an urban housing complex having gardens also reinforces the notion of
changing behaviour and adaptability of the former species in the changing urban scenario. Interspecific competition between these two sympatric species has been observed in one ecotype in Calcutta. Studies are required to ascertain the role of *Ae. albopictus* in its newly occupied territory in the context of urban dengue epidemiology.
OUTBREAK OF DENGUE FEVER IN TIRUNELVELI DISTRICT, TAMIL NADU, INDIA, 2012

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Background: In April 2012, increased number of dengue fever cases was reported in Tirunelveli district of Tamil Nadu. We described the outbreak by time, place and person and examined potential factors supporting mosquito breeding.

Methods: We gathered data from facility-based surveillance to identify laboratory confirmed dengue fever defined as acute onset of fever with 2 or more of the following: headache, retro-orbital pain, arthralgia, myalgia, rash, hemorrhagic manifestation and leucopenia with specific IgM detected in single serum in a resident of Tirunelveli since 18 April. We analyzed the outbreak in terms of time (epi-curve), place (spot map) and person (attack rate [AR] and case fatality). We conducted environmental and entomological surveys to identify potential factors contributing to the spread of the epidemic including mosquito breeding.

Results: A total of 1377 confirmed cases (Overall AR: 13.3 / 10000) were reported between 18 April and 20 June 2012 with 29 deaths (case fatality: 2.1%). The incidence (13.3/10000) and case fatality (2.86%) were highest in 0-15 years age group and among males (AR 6.2/10000). Epi-curve had two peaks (18- 22 May; 4-8 June) with a decline thereafter. Most (86%) of deaths occurred in rural areas. High larval breeding was seen in stored water in uncovered water containers, coconut shells, banana plantations and discarded tyres. All four serotypes of dengue virus were detected in the area.

Conclusions: Late referrals and lack of standard protocol for management of patients led to high fatality. Intermittent water supply, inadequate sanitation and waste disposal services led to increase in vector
population. Health authorities need to (1) strengthen the surveillance for early detection of newly affected areas, (2) prepare standard protocol for case referral and case management and ensure that these protocols are followed (3) educate the community to modify water storage practices and environmental hygiene and (4) implement vector control measures in affected areas.
ROLE OF DUAL INFECTIONS NOTICED DURING OUTBREAK STUDIES IN SOUTHERN INDIA

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In outbreak investigation studies carried out in southern states by Centre for Research in Medical Entomology (ICMR), Madurai, we came across with few dual infections. In 2006, in Kerala there was a patient who showed dual infection with malaria and dengue. In a recent fever epidemic survey in Kerala during May 2011, we came across with two patients who showed dual infection with Japanese encephalitis (JE) virus and Chikungunya virus. During the outbreak investigation at Tirunelveli and Kanyakumari districts during May 2012, two patients showed dual infection with Dengue virus and Chikungunya virus. The vectors for Dengue and Chikungunya are same (Aedes aegypti and Aedes albopictus), whereas for JE and Malaria the vectors are different (Culex vishnui subgroup and Anophelines respectively). The abundance of different mosquito vector species is high during an outbreak because of availability of different breeding habitats in that affected area. Dual infections can possibly change the clinical spectrum of the disease; consequently, specific treatment may also be affected. Thus it requires urgent attention. Investigation of epidemiological and immunological aspects of patients with concurrent infection is worth pursuing.
CHIKUNGUNYA FEVER, AN IMPORTANT RE-EMERGING PUBLIC HEALTH PROBLEM IN TAMIL NADU

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Chikungunya virus (CHIKV), an alphavirus belongs to the family Togaviridae, causes Chikungunya fever (CHIKF) in human. CHIKV is transmitted by Aedes aegypti and Aedes albopictus mosquitoes. CHIKF is characterized by the abrupt onset of fever, nausea and severe arthralgia followed by constitutional symptoms and rash which last for 1-7 days. It is a self-limiting disease and rarely fatal. Though the clinical signs and the symptoms of CHIK fever and Dengue fever are very much similar and are difficult to distinguish without the support of laboratory investigations, the severe arthrolgia is the commonest symptom in CHIK fever cases. CHIKF is rarely fatal, however, CHIKV associated mortality has been recognized recently in the cases from India, Mauritius and in the Reunion islands. CHIKF is routinely diagnosed by the detection of virus specific IgM antibodies by MAC-ELISA test. The virus has been found circulating in different districts in Tamil Nadu. Extensive survey was conducted in Tirunelveli, Theni, Dharmapuri, Chennai etc. in Tamil Nadu. The results have indicated the active circulation of CHIKV in the community, during the dengue outbreak. Hence, the clinician should consider CHIKF in the differential diagnosis of dengue like infection appeared in the community. Laboratory diagnosis will be very useful in differentiating dengue virus infection which is hyper endemic in many parts of Tamil Nadu. The study presents the serological, virological findings of the survey which were conducted recently and discusses the importance of laboratory tests in accurately diagnosing CHIKV cases during the surveillance in Tamil Nadu.
Notes
ODONATA VS MOSQUITO VECTORS: AN INNOVATIVE, ECONOMIC AND ENVIRONMENTAL CONTROL METHODOLOGY – A REVIEW

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Odonate larvae are well-known as important components of freshwater ecosystems. They are predators of macroinvertebrates as well as small vertebrates such as fish, mosquito larvae and amphibian larvae and other microorganisms. Medium sized larvae attack the larvae of mosquitoes, midges, blackflies, small worms, tubifex worms and nymphs of aquatic insects, tadpoles, the fry of fish, and even other odonate larvae. The mosquito control intervention resulted in minimal impact on the odonate community in southern Mexico. Once hatched the odonates eggs, the larva becomes an aquatic predator, feeding on other aquatic insects, including mosquito eggs and larvae. Dragonflies eat more that mosquitoes and mosquito larvae. An overpopulation of dragonflies could increase competition and harm populations to other local rare or endangered insects. Larvae and adults of the odonata are considered efficient predators in the rice fields. The dragonflies and damselflies are true enemies of mosquitoes as the larvae as food and the adults are efficient predators of airborne adult mosquitoes. Dragonfly larvae are known to prey heavily on bottom feeder mosquitoes like Aedes larvae. A pilot field study, involving periodic augmentative release of predaceous larvae of a dragonfly to suppress Ae. aegypti during the rainy season in Yangon, Myanmar. Owing to growing resistance to commonly used organochlorine and organophosphate chemicals in the field, control of mosquitoes through eco-friendly biological means may be advantageous. Other benefits of biocontrol agents includes an ability to kill the targeted species while being safe for nontargeted ones; ease of field application; inexpensive production and the lack of infectivity and pathogenicity to mammals, including humans. Augmentative release of the
odonata larvae entails prior estimation of the numbers of natural enemies needed (within a given area and a given time) to achieve suppression to the required level, and then releasing sufficient numbers into a closed system, i.e. an environment from which they cannot disperse. The systematic release of dragonfly larvae during the monsoon season (the time when dengue fever was being transmitted by the mosquito) rapidly depressed the mosquito populations to a level lower than could have been achieved by any other known method, including treatment by chemical insecticide.
STUDIES ON CERTAIN ASPECTS OF MALARIA DEATH WERE OBSERVED IN SENTINEL SURVEILLANCE HOSPITAL WITHIN SURAT CITY, INDIA.

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Malaria is major public health problem throughout the world it kills 2 million people every year and there are over 300 million new cases each year. Early diagnosis and complete treatment is the most important component of malaria control programme, which has a direct bearing on prevention of mortality and reduction in the morbidity, therefore follow up of the patient treatment is an important aspect of monitoring disease in community.

A system generated in Surat Municipal Corporation (SMC) that daily report of out door patient and indoor patient from 39 urban health centres, 12 major private and government hospital data were collected in report of fever and malaria. Details of patient name address, on set of illness, history of movement, treatment taken from clinic etc. and then field investigation made according to analysis and evaluate at the headquarter for further action.

During the year from 2007 to 2011 total of 250 deaths due to malaria were reported in Surat city. Out of which 31 malaria deaths were found due to P. vivax infection and 198 deaths were contributed P. Falciparum infection. However 11 deaths were found for mix (PV & PF) infection and rest of 10 deaths were found due to fever related. Out of 250 death cases reported in Surat city study group analysed for gender distribution which had revealed 182 (72.8%) for male and 68 (27.2%) for female this study had indicated male were more exposed to death as compared to female. Studies indicated high case fatality rate in the age group of 15-44 Yrs. (44.4%) 111 out of 250. Based on available of hospitals various records of Surat city showed that 163
cases have parasitemia density and co-morbidity out of 250 deaths. The essential pathologic features of severe malaria sequestration of erythrocytes that contain mature forms of the parasite in the deep vascular beds of vital organs, thus producing cerebral malaria (1.23%), renal failure (31.3%), hepatic dysfunction (37.4%) and ARDS (31.3%).

However severe anaemia (22.7%) and thrombocytopenia (66.9%) that causes bleeding diathesis is produced by haemolysis, reduced cell deformity of parasitized and nonparasitized erythrocytes increased splenic clearance, reduction platelet survival, decreased platelet production, and increased splenic update of platelets and can be produced by *P. Vivax* and *P. Falciparum* infection. In addition to above complication the parasite density observed mainly in severe vivax malaria and P. Falciparum malaria respectively showed 829-49831/µl and 234-77818/µl.

Based on the prospective observational study indicated that, *Plasmodium vivax* monoinfection was almost equally serious cause significant mortality in comparison to *P.falciparum* infection thus though the *P. vivax* infection is generally mild and does not cause much mortality to the patient our study clearly indicated from the above findings that *P. vivax* infection can cause significant severe morbidity as well as the mortality.

Key words: Malaria, death, sentinel, vivax and hospital.
FLAVIVIRUS INFECTIONS IN SCHOOL CHILDREN OF THANJAVUR DISTRICT, TAMIL NADU

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Japanese encephalitis (JE) is caused by a virus and is transmitted through mosquitoes to children below 15 years. Thanjavur district (Rice Bowl of Tamil Nadu) is the major rice growing area in Tamil Nadu, and has provided favourable breeding conditions for JE vectors. But paradoxically there are no outbreaks of JE and the encephalitis case incidence is very low. Rice is cultivated in about 47, 58, 333 hectares, in two crop seasons namely ‘Kuruvi’ (June to September) and ‘Samba’ (August to January). Blood samples were collected from school children of age 5-10 in the study villages of Thanjavur district. Athi Dravider welfare primary school in Kovilur, Panchayat union middle school in 34 Kovilur, Primary school in orantharayankudikado, Panchayat union middle school in muthur, Panchayat union middle school in Naduvakkottai and TAAAD middle school in Tiruvidaimaruth. Blood samples were collected from school children of age 5-10 in the above 6 study villages of Thanjavur district during October 2011. From each child 250µl of finger-prick blood specimen was collected using disposable sterile lancet, into a sterile vial containing 10 units of heparin. Blood samples were transported on ice to the base laboratory at Thanjavur DDHS, centrifuged at 200g for 10 minutes and the separated plasma specimens transported on ice to the Centre for Research in Medical Entomology (CRME), Madurai where they were stored at -20ºC till tested. Plasma samples was extracted with acetone, absorbed with goose erythrocytes and tested by Haemagglutination Inhibition (HI) test. A total of 314 children samples collected during October 2011 were screened for flavivirus antibodies (JE, WN and DEN antibodies) by HI test. Of 314 plasma samples tested, 129 (41.1%) were found positive for flaviviruses. Virus wise, age wise and sex wise details are discussed.
JAPANESE ENCEPHALITIS SURVEILLANCE AND 
EPIDEMIOLOGY IN TAMIL NADU

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Japanese encephalitis (JE) is a mosquito borne zoonotic viral disease caused by JE virus, involving the Central Nervous System. It is a non-contagious, mosquito-transmitted virus disease mainly affecting children below 15 years of age. Severe epidemics have occurred in the past and there is evidence to suggest that the disease is spreading to new areas. Kerala, experienced the first outbreak in 1996, and since then sporadic cases are being reported. The epidemiology of JE is complex due to a complex transmission cycle of the virus involving a variety of vertebrate and invertebrate hosts. JE case surveillance is based on the clinical and laboratory diagnosis. It is important to know whether gender difference in disease incidence is real or due to some other confounding factors. Vector abundance and vector infection are two important parameters in vector surveillance. Although considerable information on the disease is available, there are still gaps in knowledge about the epidemiology which need to be understood in order to develop sound surveillance systems for forecasting epidemics and control strategies. JE is a vaccine preventable disease but, unfortunately there has been no national policy on vaccination against JE. This is partly because there is a lack of epidemiological data from different endemic areas which can focus on the gravity of situation. Epidemics of JE continue to occur in different parts of the country and the disease is spreading to newer areas. But, epidemic-prone states have no plans to set up effective surveillance systems which are urgently needed to monitor JE activity. It is essential that appropriate control measures are taken based on sound knowledge of epidemiology and surveillance strategies, so that the disease can be controlled. In this article, it is intended to provide an update of research to understand the epidemiology of the disease in Tamil Nadu, quantify transmission, develop surveillance systems, which may serve as paradigms for other endemic areas.
SUBCLINICAL FLAVIVIRUS INFECTION IN SCHOOL CHILDREN OF TIRUNELVELI DISTRICT, TAMIL NADU

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Japanese encephalitis is the leading viral cause of Acute Encephalitis Syndrome (AES) in Asia. The disease primarily affects children under the age of fifteen years. Seventy percent of those who develop illness either die or survive with a long-term neurological disability. Since the first case of JE was documented in the late 19th century, the disease has spread beyond its early domain - traveling as far as Australia by the year 2000. Over the past 60 years, it has been estimated that JE has infected ~10 million children globally, killing 3 million and causing long-term disability in 4 million. Many individuals living in flavivirus endemic areas developed sub clinical infections in childhood and gain a degree of protective immunity. Tirunelveli district, lies on southern part of Tamil Nadu between 8°05’ and 9°30’ north latitude and 77°05’ and 78°25’ east longitude.

Finger prick blood samples were collected from school children of age 5-10 in 4 study villages of Tirunelveli district [Harijan primary school, Senthimangalam, TDTA primary school, Union primary school, manur from Kuthalaperi, Hindu middle school, union primary school, TDTA primary school from Magiladi, and Union primary school, Ariyanayagipuram] during November 2011. From each child 250µl of finger- prick blood specimen was collected using disposable sterile lancet, into a sterile vial containing 10 units of heparin. Blood specimen were transported on ice to the base laboratory at Tirunelveli DDHS, centrifuged at 200g for 10 minutes and the separated plasma specimens transported on ice to the Centre for Research in Medical Entomology (CRME), Madurai where they were stored at -20°C till tested. Plasma samples was extracted with acetone, absorbed with goose erythrocytes and tested by Haemagglutination Inhibition (HI) test for HI antibodies.

Flaviviruses
A total of 326 children samples collected during November 2011 were screened for flavivirus antibodies (JE, WN and DEN antibodies) by HI test. Of 326 plasma samples tested, 239 (73.3%) were found positive for flaviviruses. Virus wise, age wise, sex wise and village wise details are discussed. In JE, there is always a preponderance of subclinical infections and therefore, reporting only overt cases underestimates the total level of virus transmission in an endemic area. Subclinical infections can be quantified by estimating seroconversion rates in susceptible age-groups of children.
ELIMINATION OF BAMBOO STUMP FENCING TO PREVENT DENGUE MOSQUITOES BREEDING IN THE DISTRICTS OF WEST BENGAL

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It is well known that West Bengal is famous for growing Jute and bamboos. In the recent entomological survey for Japanese encephalitis / dengue study in May 2012 throughout districts of West Bengal, it is noticed that every house is having fence around their houses for their gardens made-up of bamboo straps and stumps in order to prevent cattles from entering their garden. The vast growing bamboos cheaply available in West Bengal are the reason for the utilization for fencing in the houses. The full grown bamboo will be holding several nodes at the gap of one foot in between the bamboo and there is hallow space in between each node. The block in each node of the bamboo doesn't allow water to enter from one portion of the bamboo to another portion. The bamboo when cut into pieces, the hallow space will be seen in between each node which looks like container in form. The bamboos are buried straight in the ground for fence and the groves in the bamboos facing the sky pave way for rain water storage and attracts Aedes mosquitoes to breed heavily in each water holding stumps. The entomological study reveals that almost all the villages in the districts are having bamboo fencing in their houses. The bamboo stumps in 25 houses in each district were examined for Aedes larval breeding where 70% of immatures of Aedes found in the bamboos. The larvae brought from the field after emergence were identified as Aedes albopictus (40%), Aedes scutellaris (30%) and Armigeres (sabulbatus and magnus) (24%) Tripteroides (5%), Toxorhynchites (1%). The permanent solution to control Aedes breeding in the bamboo stumps is to remove the hallow bamboo stumps in between the strap fence and replace solid stumps which dose’nt hold water, secondly the hallow spaces can be blocked by stuffing mud or sand into the bamboo holes and thirdly making a hole above the bamboo node to remove stagnated rain water. The method said above can be done immediately to put an end to dengue and chikungunya spread further in West Bengal districts.
GENETICALLY MODIFIED MOSQUITOES IDEAL TO REDUCE DENGUE THREAT

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Dengue fever is an illness caused by an RNA flavivirus spread by the bites of Aedes aegypti mosquitoes. Dengue affects between 50 and 100 million people in the tropics and subtropics each year, causing fever, muscle and joint ache as well as potentially fatal dengue hemorrhagic fever and dengue shock syndrome. Advances in transgenic technology have allowed the development of genetically transformed insects that have reduced ability to support the development of disease pathogens. The integration of this new method within national vector control programmes is indeed the biggest challenge, notwithstanding the current weak health systems in most disease-endemic countries (DECs) to efficiently apply vector control interventions. The genetically modified OX513A mosquito was constructed using a transposon (jumping gene) piggyBac LA513 isolated from Baculovirus, a common soil dwelling DNA plant virus. Genetically modified (GM) OX513A Aedes aegypti male mosquitoes are released in an area inhabited by people. The living modified organism (LMO) released contains a fluorescent molecular marker and a ‘self-limiting’ genetic construct. When female Aedes aegypti mosquitoes mate with OX513A male mosquitoes, all offspring will die, hence preventing the emergence of the next generation. Transgenic mosquitoes are readily identified by red fluorescence due to expression of DsRed2, a red fluorescent protein from a marine microbe. If the strategy succeeds, it may be a successful vector to minimize the burden of Dengue epidemic effectively.
INSECTICIDE SUSCEPTIBILITY *ANOPHELES VAGUS* AROUND SOLMARA CANTONMENT OF ASSAM

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The widespread use of insecticides leads to the development of insecticide resistance, making insecticide use ineffective and limiting the available options for disease control. Thus, the susceptibility status of target species is monitored routinely to select the effective ones. *Anopheles vagus* in Assam cause biting nuisance and may serve as a secondary malaria vector under unusual circumstances that includes dense concentrations of humans in association with low numbers or absence of bovids and/or primates. In the present study, *An. vagus* mosquitoes were collected using CDC light traps and aspirators (hand catch) from Udmari and Balitika villages near Solmara cantonment area. The collected *An. vagus* females were assayed for diagnostic doses of DDT (4%), deltamethrin (0.05%), malathion (5%) and lambda-cyhalothrin (0.05%) using the standard WHO method.

*Anopheles vagus* was resistant to DDT by 58.4%, but tolerant to malathion by 94% and lambda-cyhalothrin 86.3%, whereas susceptible to deltamethrin by 98.7%, respectively in Udmari. In Balitika *An. vagus* was found tolerant to DDT by 91.7% and lambda-cyhalothrin 87.8%, whereas susceptible to deltamethrin by 98.8% and malathion 100%, respectively. The KDT₉₀ and KDT₅₀ values for these insecticides also showed the similar responses. In this study baseline information on insecticide resistance was generated for *An. vagus* which may act as secondary malaria vector in the study area. The trend of insecticide resistance should be monitored carefully so that the impact of existing vector control on resistance population should be assessed.

Key words: *Anopheles vagus*, Solmara, Insecticide, Susceptibility.
PUPAL INDEX (PI) AS A RELIABLE TOOL FOR ASSESSING DENGUE VECTOR ADULT POPULATION AND ITS POSSIBLE CONTRIBUTION TO THE COMMUNITY BASED VECTOR CONTROL

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Dengue fever is emerging as important public health problems in worldwide more than 120 countries were recorded and about 2.5 billion people were affected every year. The new area was extending day by day due to increase of urban population. In Chennai, since 1998 dengue has a serious public health problem, a major epidemic occurred in 2001; recorded 737 cases and all four sero-types were reported. Now, Chennai is hyper-endemic area, due to its susceptibility an urgent and important vector control measures could be needed, very particularly, concentrated on eliminating the most pupal productive containers and thus prevent future outbreaks in the metro city. The lack of knowledge about vector bionomics and preventive measures would be the major causes of dengue in the highly amalgamated urban population. The most economical way is to develop technique for the people to entrust their responsibilities to a simple approach of cleaning the high pupal productive key containers. Plastic /Metal Drum constituted (80.7%), (28.4% in CT/CC) in the Check area when compared with Intervention area (6.6%) (0.4% in CT/CC) was reduced due to IEC awareness and also organized mass cleanup campaign was conducted with help of Student community (Health Ambassador) and SHG volunteers were involved during pre monsoon seasons.

In absence of dengue vaccine, presently, the only available method to decrease the disease burden of dengue fever and its severe manifestations of DHF, DSS is by reducing vector population through an inexpensive simple
method of cleaning the major *Aedes aegypti* pupal producing key containers like cement tank and cement cisterns were identified in the study area. House Index (HI), Container Index (CI) and Breateau Index (BI) were not directly related to the adult densities. Pupal production was directly involved with adult mosquitoes. Hence, pupal index (PI) of *Aedes aegypti* in the containers will be proxy to the number of adults. Therefore, *Aedes aegypti* pupal producing containers with its abundance were identified. Very less chance of mortality for pupae in the large water holding containers, when compared with any of the larval stages, obviously it shows that the container with water was more than 7 days and not properly cleaned. Results were recorded as a measure of pupal production in both the area. This pupal index (PI) tool was accurate, dependable and comparable would help for public health policy makers for assessing disease burden in the community.
IN VITRO ANTIPLASMODIAL EVALUATION OF EXTRACTS OF MARINE CYANOBACTERIA FROM COASTAL REGIONS OF TAMIL NADU

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To identify novel antimalarial compounds from marine cyanobacteria, 25 cyanobacterial species have been isolated from various coastal regions of Tamil Nadu and identified by microscopic methods. Extracts of in vitro mass cultivated cyanobacteria using Methanol- chloroform mixture (1:1 v/v) were evaluated for their antiplasmodial activity against \textit{Plasmodium falciparum} by fluorescence based SYBR Green I assay. Extracts of \textit{Lyngbya} sp. and \textit{Oscillatoria} sp. showed promising antiplasmodial activity (IC\textsubscript{50} 18 µg/ml) against chloroquine (Cq) sensitive Pf 3D7 while their IC\textsubscript{50} against Cq resistant Pf Indo were 34 and 62 µg/ml respectively. Preparative TLC was used to obtain three fractions that showed anti Pf 3D7 IC\textsubscript{50} values of 100, 19 and 4 µg/ml respectively. Further the lack of cytotoxic effects (IC\textsubscript{50} >200 µg/ml) of active crude extracts against HeLa cell line corroborated that these extracts were nontoxic and specific for \textit{P. falciparum}. These exploratory studies suggest the possibilities of development of novel antimalarial compounds from cyanobacterial species.
In India Aedes aegypti is the recognised vector for dengue fever (DF) including its severe forms, dengue haemorrhagic fever (DHF) and dengue shock syndrome (DSS). Dengue is a disease linked to the domestic environment because the vector Aedes aegypti breeds in household water storage containers as well as in artificial, discarded containers both in domestic and peridomestic areas. Scarce water provision leads to storing water in containers which favours the breeding of Ae. aegypti. Besides, rainfall is a major parameter involved in the bionomics of Ae. aegypti. It provides abundant sites for vector oviposition. Thus an ovitrap which is a device consists of a black cylinder with a piece of cardboard is mainly used to monitor, control and detect aedes mosquito populations and thus acting as an early warning signal to preempt any impending dengue outbreaks. The black ovitrap attracts female mosquitoes to lay their eggs. Ovitraps had long been used in two major Ae. aegypti eradication programmes in the United States of America. Its effectiveness was also demonstrated in the eradication of Ae. aegypti at the Singapore Airport. In Thiruvananthapuram dt the dengue incidence per one lakh population was more than two and about 65% of dengue cases in Kerala were reported from Thiruvananthapuram dt during the year 2006. An ovitrap surveillance of dengue vectors was initiated in four localities viz. Kottapuram-Vizhinjam Panchayat(rural), Puthukulangara-Vellanad CHC(rural), Rajiv Gandhi Nagar-Medical College area -Ward 12(urban) and Karikkamandapam-Nemum PHC-Ward 52(urban) where higher number of cases were recorded when compared to other localities of rural and urban set up. The ovitrap surveillance was carried out from November 2007 to March 2010. Ovitraps survey showed only Aedes aegypti from Villinjam coastal rural area. Aedes albopictus could be obtained from Vellanad, Nemum & Medical college areas. Ovitrap breeding data can be used to identify mosquito breeding hotspots and risk areas when there is a danger of high Aedes mosquitoes infestation. These results can be used in future to plan effective vector surveillance and control operations.
ADULTICIDAL AND REPELLENT ACTIVITY OF 
ELAEAGNUS INDICA LEAF EXTRACTS AGAINST CULEX 
QUINQUEFASCIATUS ADULTS (DIPTERA: CULICIDE)

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Culex quinquefasciatus is a primary vector of lymphatic filariasis and Japanese encephalitis in India. Vector control methods involving use of chemical insecticides are becoming less effective due to the development of insecticide resistance among mosquitoes. Alternative approach of using botanicals has now become increasingly popular. In present study adulticidal and knockdown potential of leaf extract of Elaeagnus indica was investigated. Freshly emerged adult mosquitoes tested as per WHO guidelines. The result show methanol extract to be a potent knockdown agent with 80% efficiency in CDC bottle bioassay and 90% efficiency in smoke toxicity test. The present study shows that E.indica can be used as mosquito repellent with high degree of efficiency.

Keywords: Elaeagnus indica, Culex quinquefasciatus, Repellent activity, Smoke toxicity, Phytochemical
REPRODUCTIVE REGULATION IN Aedes albopictus MOSQUITOES BY Wolbachia ENDOSYMBIONTS

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Wolbachia are cytoplasmically inherited alpha-proteobacteria well known for inducing a variety of reproductive abnormalities in the diverse arthropod host they infect. The most common reproductive anomaly induced by Wolbachia in mosquito host is Cytoplasmic Incompatibility. During our survey of Aedes albopictus from populations across Karnataka and Kerala we found that all the populations were doubly infected with Wolbachia A (wAlbA) and B (wAlbB) supergroups. Further exploration of the crossing studies gave interesting insights into the dynamics of CI. It was determined that the Wolbachia density directly influence the ‘reproductive fitness’ in Aedes albopictus mosquitoes. The study found that Bangalore Aedes male and female mosquitoes have varying higher levels of A and B bacterial densities, compared to the mosquitoes of Kerala. The Bangalore female mosquito populations have the bacterial density of A very high compared to B supergroup. In Aedes mosquitoes from Kerala populations the density of A supergroup in females was high compared to that of B supergroup. The findings suggest that during multiple infections, most possibly the A supergroups dominates the B supergroup in terms of its density. When the Bangalore male mosquitoes were crossed with Kerala mosquitoes, an increased CI was observed (54.60%) in comparison with the reciprocal cross between the Kerala males and Bangalore females. It is hypothesized that there is a variation in the ‘modifying factors’ and ‘rescuing factors’ in Bangalore and Kerala populations. Wolbachia densities are likely to influence the rate of maternal transmission and could also affect the penetrance of CI in incompatible crosses.
Notes
LARVICIDAL EFFICACY OF CALLISTEMON VIMINALIS AND ALSTONIA MACROPHYLLA AGAINST Aedes Albopictus, VECTOR OF DENGUE AND CHIKUNGUNYA

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Mosquito transmits a number of diseases and remains a major source of morbidity and mortality worldwide. Use of synthetic insecticides is the first line of action for controlling the mosquitoes in the integrated vector management. However the continuous use of these insecticides may lead to the development of resistance and permanent residual effect on the bioenvironment which can be detrimental to animals including human. These factors have created a need for search of easily biodegradable alternative insecticides. The present study assessed the larvicidal activity of Callistemon viminalis and Alstonia macrophylla from M.P. region (India). Leaf extracts of these plants in isopropanol was used in the range of 10-200 ppm in the laboratory bioassays against third instar larvae of Aedes albopictus. The mortality data was recorded after 24 hrs and subjected to Probit analysis to determine the lethal concentrations (LC$_{50}$ and LC$_{90}$). C. viminalis leaf extract showed highest efficacy with LC$_{50}$ 110.67 ppm as compared to A. macrophylla which showed lower mortality with the LC$_{50}$ value 233.14 ppm. The results indicate that C. viminalis leaf extract have the potential to be used as an ecofriendly approach for the control of Ae. albopictus mosquito. Further studies on the screening, isolation and purification of bioactive phytochemical constituents/compounds followed by in-depth laboratory and field bioassays are needed.
Prevention of mosquito biting is one of the major problems in the world. Mosquitoes transmitting many diseases and causing millions of death every year. The repeated use of synthetic insecticides to control mosquitoes has created resistance and adverse environmental effects in addition to high operational cost. Many natural products have been reported as an insect antifeedants or repellents. *Azadirachta indica*, *Ocimum tenuiflorum*, *Mangifera indica* and *Citrus lemon* contains several active ingredients that are toxic to insects and safety to the environment. The safety evaluation of different parts of neem, tulsi, mango, and lemon leaf preparations were made and used as an alternative insecticide in many parts of the world. The present study was carried out to determine the efficacy of multi leaf extract against *Culex* mosquitoes in laboratory and filed studies, resulted that the aqueous crude mixed leaf extract shows 45% and 90% mortality rate of mosquitoes for 6hrs and 12hrs respectively.
PFCRT AND PFMDR MUTATIONS MAY NOT CORRESPOND TO CHLOROQUINE RESISTANCE IN PLASMODIUM FALCIPARUM

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Drug resistance in malaria parasite, Plasmodium falciparum is an important aspect in malaria control. Chloroquine has been reliable, safe and cost effective in malaria treatment, however resistance to this drug has been reported from many places throughout India. Northeastern states are endemic to malaria and cases due to falciparum contribute majority of the infection. The association of chloroquine resistance with molecular markers has not been studied well in this region. We have assessed the chloroquine resistance correlation with pfcrt and pfmdr mutation in three malaria prone areas of northeastern India. Total 77 patients with confirmed uncomplicated malaria were enrolled for the study. The cases were treated with chloroquine by the local medical authority as per NVBDCP guidelines and resistance study was carried out following WHO procedure. Treatment success rate after 15 days was 55.8 (43) percent, while remaining patients did not respond to the treatment and showed treatment failures. Pfcrt K76T mutation was found in 50.6 % cases, however, all the treatment failure cases were positive for this mutation. Pfmdr N86Y mutation could be recorded only in 31.2 % of the cases. Treatment failure was positively correlated with pfcrt mutation but not with pfmdr. Sequence analysis revealed that all the three type of amino acid haplotype of pfcrt gene are prevalent in the region. Present findings suggest that chloroquine resistance may not necessarily be contributed by pfcrt K76T and pfmdr N86Y mutation in the northeastern region and these markers may not be considered reliable in determining the chloroquine resistance in P. falciparum.

Key words: Malaria, chloroquine, resistance, pfcrt, pfmdr.
BAGAZA VIRUS, AN INSECT FLAVIVIRUS ALTERS REPLICATION OF JAPANESE ENCEPHALITIS VIRUS IN CULEX TRITAENIORHYNCHUS MOSQUITOES

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Abstract: Bagaza virus (BAGV), an arbovirus synonymous with the economically important Israel turkey meningoencephalitis virus, belonging to flaviviridae has been found to circulate in India as evidenced by virus isolation from mosquitoes and antibodies in human serum. BAGV is capable of inducing febrile illness in humans and high mortality in wild birds. A study was initiated to determine whether BAGV plays any role in the Japanese encephalitis virus (JEV) epidemiology as Culex tritaeniorhynchus mosquito, the universal vector of JEV, replicates both the viruses. Mosquitoes were inoculated with BAGV intra thoracically and were fed with JEV orally after eight days of incubation. Mosquitoes were harvested on 7th and 14th days post infection after blood meal; JEV titer was determined and compared to JEV yield in BAGV negative mosquitoes. More than three log reduction in JEV titer was observed in mosquitoes previously infected with BAGV in comparison to BAGV negative mosquitoes. However, no significant difference in virus titer was observed when both the viruses were inoculated simultaneously. The study demonstrates that persistent infection of Cx tritaeniorhynchus mosquitoes by BAGV alters the susceptibility to JEV infection. This study has great significance as the non pathogenic insect Flavivirus might be able to play an important role in the management and control of JEV outbreaks.
HOST FACTORS IN FALCIPARUM MALARIA INFECTION IN CHILDREN OF ENDEMIC AND NON-ENDEMIC AREAS

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Falciparum malaria is a dreaded disease in vulnerable children which causes high mortality and severe morbidity due to variety of complications of the disease. However, in our clinical practice we observed a difference in the prevalence, morbidity and mortality risk in tribal children of malaria endemic area as compared to nontribal children of non endemic areas.

A pilot study was undertaken on 15,000 tribal children in Bhubaneswar showed that 55% of children carried P. falciparum infection, 40% of children suffered from P. falciparum malaria out of which 34% belonged to endemic districts and rest 15 % carried the Pf parasite but did not suffer from symptomatic fever. The remarkable thing is although they suffered from malaria but they were affected by the clinical manifestations of P. falciparum malaria in a very limited way and on the other hand some of the children were asymptomatic but carried P. falciparum in them. The reason of difference in clinical complications of the disease, mortality and morbidity pattern could be due to an immunological umbrella which is protective in children of endemic region as compared to children of non endemic region.

The observations of the study highlighted the importance of immunological and physiological aspects of host components in malaria pathogenesis. Simultaneously it is also important to identify the respective parasite strains and vectors at regional level for adding new dimensions to therapeutic interventions as well as for more specific targeted control measures to be taken up by the programme for curtailing malaria transmission. We hypothesize the marker of the immunity in these children of endemic region could be the different cytokine system compared to the children in non-endemic area. So, extensive studies are needed to find out the genetic, immunological and physiological aspects of host components in malaria pathogenesis.
IDENTIFICATION OF SEQUENCE MOTIFS OF ENVELOPE GLYCOPROTEIN INVOLVED IN DENGUE VIRUS-VECTOR INTERACTIONS

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Dengue virus (DENV) is a most common arthropod-borne tropical disease that causes 50 million infections every year. Understanding the viral entry would help in developing an efficient mosquito control strategy. It is proved that the immunoglobulin like domain III contains the receptor binding motif. Dengue virus entry into mosquito cells is poorly explored. Dengue virus uses the available surface receptor proteins of vectors and host to enter inside the host cell. Using overlay protein binding assays, proteins with the molecular mass of 67kDa, 45kDa, laminin receptors and prohibitin were claimed to be a receptor for Dengue virus in insect cells. In this work, we used computational methods to predict the putative receptors in Aedes aegypti. The receptor binding domain of Envelope glycoprotein was structurally analyzed for the presence of sequence motifs. It was identified that DENV1 and DENV3 have NGR motif at domain III which was reported to bind to various members of integrin family. Similarly, DENV2 and DENV4 were found to contain TRG_ENDOCYTIC_2 motif which is a tyrosine based sorting signal responsible for the interaction with mu subunit of adaptor protein complex. According to the prediction strategies, the integrin receptor protein family and adaptor protein complex were found to contain the complementary sequence motifs of envelope glycoprotein. It needs further confirmation through wetlab experiments.
COMPARISON OF BIOLOGICAL ATTRIBUTES OF *CULEX QUINQUEFASCIATUS* (DIPTERA: CULICIDAE) POPULATIONS FROM INDIA

D. Mangesh Gokhale*, S. Mandar Paingankar and D. Sachin Dhaigude

Biological data was compared for two populations of *Culex quinquefasciatus* Say 1823 from Gorakhpur and Pune areas from India. Development and survival of immature and adult stages of *Culex quinquefasciatus* was found significantly different in Gorakhpur and Pune populations. Principal component analysis of morphological data revealed that the two populations form significantly different clusters which can be differentiated from each other based on siphon, saddle, anal gills and pecten teeth related variables. Insecticide susceptibility results suggest that the larvae from both areas were found to be more susceptible to deltamethrin as compared to DDT and malathion. The current study provides baseline information on survivorship, morphological variation and insecticide susceptibility of *Culex quinquefasciatus* which is useful in designing effective vector control strategies.
Mosquito-borne filarial nematodes cause severe, debilitating disease of human lymphatic filariasis, which afflicts more than 128 million people worldwide. Development of an efficacious vaccine for parasitic diseases, including lymphatic filariasis, has been one of the greatest challenges in immunological research. Abundant larval transcript (ALT) is a well-established vaccine candidate for human lymphatic filariasis showing a 76% protection in jird models. However, ALT is proven to have an immunosuppressive function in vivo and specific regions in the protein have been shown to contribute to its protective as well as immuno-modulatory functions. In this study we attempt to identify the immunosuppressive and epitope regions of ALT protein to have a better understanding of this unique parasite protein to develop an efficient subunit vaccine. This can be achieved only by a complete epitope mapping of ALT to determine its functional B and T epitopes of which some may be immunodominant or immunosuppressive. Here, we attempt to map the B and T epitopes of Bm-ALT and to identify immunosuppressive regions using various B cell and T cell assays.
SIGNIFICANCE OF GENOTYPING OF LYMPHATIC FILARIAL PARASITES

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Lymphatic Filariasis (LF), one of the neglected tropical diseases causing long-term disability, is a mosquito-borne disease caused by filarial nematodes, including Wuchereria bancrofti and Brugia malayi. In the fast-moving race between host and parasite, one would predict a monomorphic species would be driven to extinction. Therefore, the genome of the parasite is likely to undergo changes to the extent of re-structuring its own populations. There are two imperatives to study genetic polymorphisms in lymphatic filarial parasites. Firstly, the development of vaccine candidate antigens cannot proceed far without the knowledge of the distribution and conservation of antigen genes among parasite populations worldwide. Whether these parasites display any form of antigenic variation also needs to be looked into. Secondly, the threat posed by potential drug resistant alleles to the success of anti-filarial therapy appears serious and draws immediate attention. Hence, it becomes necessary that we understand the population genetic structure of these parasites in order to predict how such polymorphic alleles may spread in natural populations. Here, we discuss the currently known molecular polymorphisms in filarial parasites and their potential utility in devising appropriate strategies for the control and elimination of lymphatic filariasis.
Dengue fever (DF) and its severe forms, dengue hemorrhagic fever and dengue shock syndrome, are mosquito-transmitted arboviral diseases belonging to genus Flavivirus, family Flaviviridae. DF has a major impact on public health in many tropical areas worldwide. It is currently the most important mosquito-borne, human viral disease in terms of both the number of cases and the number of deaths. The principal vector of dengue virus in urban areas is the highly domesticated *Aedes aegypti* (Linn.) mosquito, whereas *Aedes albopictus* (Skuse) is a dengue vector in Asia and the Pacific Islands. In Southeast Asian countries, dengue epidemic usually occurs during the rainy season and is correlated with an increase in breeding habitats and vector populations.

In vertical transmission by mosquitoes, the virus is transmitted by the progeny through an infected female mosquito, whereas in venereal transmission, the male transmits the virus to the female during copulation. Persistence of vertical or transovarial transmission of dengue viruses may serve to retain dengue viral pathogen in nature during inter-epidemic periods of the disease. During November 2011, dengue case incidence was reported from 4 foci in Warangal district, one from in and around Warangal municipality area (Shanthi Nagar, Kareemabad, Rangasai pet, Patakmahalla) the other foci are from Raiparthy (P) PHC area (Kowkonda) and Mogullapally PHC area (Issipet) and also from Hasanparthy PHC area (Sudhanpally). With the help of Directorate of Public Health Services, Hyderabad, the Centre for Research in Medical Entomology team carried out the larval and pupal survey as well as indoor resting collection in the affected areas. An ideal situation for *Aedes* breeding in that area with water storage containers in indoor and rain water collections in the discarded containers around the houses was observed during the survey. The collected immature were reared in the laboratory and all adult mosquitoes were identified into species level, pooled (maximum 20 mosquitoes per pool) and stored in
liquid nitrogen till testing. *Aedes* pools were screened for the presence of dengue viruses by antigen capture ELISA using dengue – specific monoclonal antibody. A total of 26 pools, consisting of 177 mosquitoes of *Aedes* mosquitoes collected from Warangal district, Andhra Pradesh were processed by dengue antigen capture ELISA. Out of 26 pools tested, 7 pools (6 pools of *Ae. aegypti* and 1 pool of *Ae. vittatus*) were found positive for dengue virus. Transovarial transmission of dengue virus by available vector species in a dengue endemic setting could be the key etiological phenomenon responsible for re-emergence of the disease from inter-epidemic to epidemic phase of disease onset. Thus monitoring natural infectivity of mosquitoes carried through vertical route of virus, may serve as an important surveillance tool for risk prediction as well as for the prevention of dengue emergence in an area and pin point vector virus foci for their elimination during non-epidemic periods of dengue. TOT of *Ae. vittatus* is reported for the first time in Warangal district. This emphasizes the need for continuous monitoring of dengue virus infections in vector mosquitoes in the study area.
MOSQUITO SALIVARY GLAND PROTEINS AND THEIR ROLE IN THE TRANSMISSION OF ARBO-VIRAL INFECTIONS

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Mosquito salivary gland (SG) plays an important role in the disease transmission because pathogens are transmitted to a host when the female mosquito takes a blood meal and mosquito SG’s are the final destination for the parasite maturation and transmission. Hence, identification and characterization of SG proteins are important in order to find out their role in the disease transmission. Mosquito SGs contain many enzymes and physiologically active proteins necessary for their blood-feeding behaviour. Several studies have shown that the D7 family of salivary proteins has been associated with binding and probably scavenging of biogenic amines such as serotonin, histamine, and nor epinephrine, which could antagonize vasoconstrictor, platelet-aggregation, and pain inducing properties. Recent studies have suggested that several proteins including small antimicrobial peptides (AMPs) like gambicin, defensins, and cecropins, presumably involved in immune surveillance, are activated in the midgut and salivary glands of the mosquito A. gambiae upon parasite infection.

In this study, SGs are dissected and collected in Phosphate Buffered Saline (PBS) with protease inhibitor on ice and stored at or below -20°C to avoid protein degradation. Studies were reported that SG is development only after 36 hours of emergence and after 5 days there is an increase in the mosquito SG proteins such as Esterase and Acid phosphatase. Poly Acrylamide Gel Electrophoresis technique is generally employed to separate the different enzymatic proteins and further characterization of such proteins are being done using 2D gel electrophoresis/Mass spectroscopy (LCMS). Cationic ferritin, lectins, α-aminase, α-glucosidase, invertase and apyrase (ATP diphospho hydrolase) are some of the proteins having significant physiological activities. Earlier studies revealed that apyrase activity starts from the second
day onwards and decreases after blood meal but not after sucrose meal. So far, little work has been carried out to find out the exact role of mosquito SG proteins in blood feeding as well as in disease transmission. Preliminary studies were carried out to determine the protein profile pattern of salivary gland proteins. Around 200 numbers of female SGs were dissected and their individual proteins are being characterized. The difference in the protein profile pattern of SG proteins of mosquitoes and their possible role in the disease transmission have been discussed.
MOLECULAR APPROACH IN DIAGNOSIS OF FILARIASIS

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Lymphatic filariasis (LF), caused by mosquito-transmitted filarial worms Wuchereria bancrofti, Brugia malayi and Brugia timori, is the second leading cause of long-term disability and seriously affects socioeconomic arena worldwide. The disease affects over 120 million people in 83 countries worldwide (Molyneux et al, 2003). Most of infected individuals are asymptomatic, while the common chronic clinical manifestations are lymphedema, hydrocele and elephantiasis. The Global Program to Eliminate Lymphatic Filariasis (GPELF) has targeted LF to be eliminated by 2020, by using selective diagnosis to identify endemic areas followed by repeated cycles of mass drug administration (MDA), to reduce both infection prevalence and transmission rates to levels below those required for sustained transmission. PCR followed by RFLP analysis may be used to differentiate filariae of different species in co-endemic areas (e.g. infective larvae in the vector). A recent multiplex PCR method allowed the simultaneous detection of Brugia and Wuchereria in blood and mosquito samples. The nested-PCR and ITS1-RFLP are potential diagnostic tools for daily routine laboratory species-specific and sensitive detection of L. loa and M. perstans filarial species. This PCR-based approach to species identification is robust, simple to perform, and easy to interpret, which makes it suitable for use in reference laboratories. The PCR-RFLP of ITS1 has potential utility in diagnosis and monitoring lymphatic filariasis control programs. Treatment of infected individuals is important to decrease the risk to other human subjects in the vicinity due to the presence of suitable mosquito vector existence.
LABORATORY BASED DENGUE FEVER SURVEILLANCE
IN AND AROUND MADURAI

N. Anuradha, Kanaka Priya Arul, Jhansi Charles,
P.A.T. Jegadeeswari and S. Karthikeyan

Dengue fever and Dengue hemorrhagic fever have become a grave public health problem in many parts of Tamilnadu in recent days. The marked increase in Dengue infection has necessitated an effective surveillance system so as to institute the control measures in a better way.

In our AES Lab, the surveillance activities are carried out by serodiagnosis for the presence of Dengue IgM antibodies in the clinical specimens received from Govt. Rajaji hospital, Madurai, nearby PHCs, GHs, EOC, Leptoclinics and private hospitals using NIV Dengue IgM Capture ELISA kit. The results are communicated to O/o DPH, NVBDCP, DD [Public health], Regional Director, SEARO EpiData team [WHO].

In 2010, the presence of Dengue IgM antibodies was tested for 1713 serum samples. Of which, 406 samples showed positivity. Only 67 samples showed positivity for Dengue IgM antibodies out of 869 serum samples tested in 2011. The positivity was found to be more during monsoon and post monsoon period. Analysis of 2012 [up to June] data shows 226 dengue positives out of 1413 serum samples being tested. The number of positives has been found to be unusually high during the pre-monsoon season (May-June) in 2012.

To summarize, the incidence of Dengue infection is on the rise in recent days and the recent epidemic has occurred during an unusual timeframe, i.e. before the monsoon involving both the rural and urban areas. Hence an effective surveillance system has to be strengthened throughout the year to foretell any major outbreak of Dengue infection.
MOLECULAR DIAGNOSIS AND ANALYSIS OF CHIKUNGUNYA VIRUS

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Chikungunya fever, caused by “Chikungunya virus,” is an arbovirus disease transmitted by the bite of infected mosquitoes belonging to the genus Aedes. Chikungunya fever epidemics have been reported from several countries around the world. Three main laboratory tests are used for diagnosing Chikungunya fevers: virus isolation, serological tests and molecular technique of Polymerase Chain Reaction (PCR). Recently, a reverse transcriptase, RT-PCR technique for diagnosing CHIK virus has been developed using nested primer pairs amplifying specific components of three structural gene regions, Capsid (C), Envelope E-2 and part of Envelope E1. PCR results are available within 1-2 days. The RT-PCR assay has many advantages over conventional reverse transcription-PCR methods, including rapidity, quantitative measurement; lower contamination rate, higher sensitivity, higher specificity, and easy standardization. Thus, quantitative RT-PCR assay might eventually replace virus isolation and conventional reverse transcription-PCR as the new gold standard for the rapid detection, quantification and diagnosis of CHIKV infection. SYBR Green DNA-based RT-PCR systems are good alternative to fluorescent probe-based RT-PCR techniques and are based on its ability to produce a 100-fold increase of fluorescence when bound to double-stranded DNA. The binding of SYBR Green to nucleic acid is not sequence-specific and the fluorescent signal produced when in complex with DNA is directly proportional to the length and amount of DNA copies synthesized during the reaction, hence making this technique very precise and sensitive. Current method can be a useful tool for rapid detection and quantification of CHIKV during natural infection, in research laboratory settings and possibly monitoring the extent of viral replication in patients for clinical diagnosis and epidemiological surveillance of possible emerging epidemic of CHIKV infection.
NS- 1 AG DETECTION – AN EARLY MARKER FOR DENGUE VIRAL INFECTION.

N. Kanaka Priya Arul, N. Anuradha, Jhansi Charles and P.A.T. Jagatheeswari

Dengue fever is one of the most common, dreadful mosquito borne arboviral illness in India. Human acquire this infection by the bite of aedes mosquitoes, which breed in artificial water sources. Dengue viral infection is characterized by fever, muscle pain, lymphadenopathy and rash. It can also cause Dengue hemorrhagic fever and Dengue shock syndrome, which are the more severe forms of illness.

Generally, serodiagnosis of Dengue fever is based on demonstration of IgM antibodies and NS1 Antigen. As IgM antibodies can be demonstrated only after 5-7 days of infection, NS1 Ag detection will be a better tool for early diagnosis to prevent complications and decrease the mortality rate.

A total of 82 serum samples were received from patients with fever of 4-7 days, from Government Rajaji Hospital, Madurai. They were all checked for NS 1 Ag [Panbio ELISA Kit]. Among them, 31 were found to be positive. The same samples were subjected to IgM antibody detection subsequently [NIV, Pune, ELISA Kit]. Out of the 82 samples, 22 were anti IgM positive. All the positive samples were then confirmed by Polymerase Chain Reaction. From the above data, it is clear that NS 1 Ag detection helped in an earlier diagnosis.

Thus the present study shows that NS 1 Ag detection helped in early diagnosis so as to initiate prompt treatment and prevent complications. This can be routinely used in outbreaks as it is also cost effective. It thereby helps to bring down the mortality rate also. NS 1 Ag detection serves as a new approach to diagnosis of Dengue Infection.
Notes
CURRENT MOLECULAR APPROACHES IN DENGUE DIAGNOSIS

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Dengue is an endemic viral disease affecting tropical and subtropical regions around the world, predominantly in urban and semi-urban areas. Dengue fever (DF) and its more serious forms, dengue hemorrhagic fever (DHF) and dengue shock syndrome (DSS), are becoming important public health problems. The World Health Organization estimates that there may be 50 million to 100 million cases of dengue virus infections worldwide every year, which result in 250,000 to 500,000 cases of DHF and 24,000 deaths each year. Dengue virus is a mosquito-borne flavivirus and the most prevalent arbovirus in tropical and subtropical regions of the world. Dengue virus is a positive-stranded encapsulated RNA virus. Laboratory diagnosis of dengue virus infection can be made by the detection of specific virus, viral antigen, genomic sequence, and/or antibodies. At present, the three basic methods used by most laboratories for the diagnosis of dengue virus infection are viral isolation and characterization, detection of the genomic sequence by a nucleic acid amplification technology assay, and detection of dengue virus-specific antibodies. After the onset of illness, the virus is found in serum or plasma, circulating blood cells, and selected tissues, especially those of the immune system, for approximately 2 to 7 days, roughly corresponding to the period of fever. Molecular diagnosis based on reverse transcription (RT)-PCR, such as one-step or nested RT-PCR, nucleic acid sequence-based amplification (NASBA), or real-time RT-PCR, has gradually replaced the virus isolation method as the new standard for the detection of dengue virus in acute-phase serum samples. The real-time PCR or RT-PCR assay has many advantages over conventional PCR or RT-PCR methods, including rapidity, the ability to provide quantitative measurements, a lower contamination rate, a higher sensitivity, a higher specificity, and easy standardization. Therefore, real-time
PCR has gradually replaced conventional PCR as the new gold standard for the rapid diagnosis of dengue virus infection with acute-phase serum samples. Present advances in molecular diagnostic methods have greatly improved the sensitivity and specificity of diagnosis of dengue virus infection. It is expected that the successful application of these assays will contribute significantly to the clinical treatment, etiologic investigation, and control of dengue virus infections.
SEPARATION AND BLOTTING OF NS1 AND NS5 DENGUE ANTIGENS FOR DIAGNOSTICS

Rohan Narayan

Background: Dengue is an important arthropod-borne Flavivirus which causes frequent outbreaks of Dengue Fever (DF) and Dengue Hemorrhagic fever (DHF) in many parts of the world. Hence, a specific and rapid serological assay for the diagnosis of Dengue is a necessity. Most of the immunoassays for diagnosis of Dengue are based on detection of antibodies against the viral structural proteins. However, since immune responses to structural proteins among Flaviviruses are cross-reactive to each other, such assays often lack specificity. Aim: The aim of this study was to develop a novel Virus Antigen Strip/Enzyme Immuno Assay (VAS/EIA) based diagnostic method for Dengue which detects IgM antibodies against the viral non-structural proteins such as NS1 and NS5.

Methods: Standard Dengue virus serotype 2 (DENV-2) was raised in C6/36 and Vero cell lines for up to 5 passages and high titer virus stock was prepared by incubating the tissue culture supernatant with 7% PEG 4000. Final virus titer was estimated using Haemagglutination Assay. The viral proteins were then separated using 10% SDS PAGE and silver stained. The proteins were then blotted on to a nitrocellulose membrane using a semi-dry blotting apparatus and stained with Ponceau.

Results: The virus titer was found to be about 250 HA units. Four DENV proteins, namely NS5 (97 kD), NS1 dimer (85 kD), NS3/Envelope protein Major (71 kD/67 kD) and NS1 monomer (45 kD) were separated.

Conclusion: Previous studies have shown that the NS5 protein is a Dengue group specific antigen and that IgM responses against NS1 protein are less in primary and more in secondary infections. The virus antigen strip can be developed based on the principle used in the experiment in order to arrive at a specific diagnosis for Dengue.
DETECTION OF POLYMORPHISM OF MTDNA COI IN
*CULEX QUINQUEFASCIATUS* BY SINGLE STRAND
CONFORMATION POLYMORPHISM (SSCP)

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*Culex quinquefasciatus* is the major vector of lymphatic filariasis. Currently, there are approximately 120 million cases of lymphatic filariasis has been recorded worldwide. For the first time detection of polymorphism of mtDNA COI was carried out in *Culex quinquefasciatus* from two geographical regions separated by 105km. Three collections were made from Madurai city, and one collection from a village in Uthamapalayam. The variations of 710 bp region of cytochrome oxidase subunit I gene among 75 adult *Culex quinquefasciatus* were examined. This gene was amplified by polymerase chain reaction and tested for variation using Single strand conformation polymorphism (SSCP) analysis which is an ideal technique to detect the genetic variation among different populations of mosquitoes. Three haplotypes were identified among Madurai collection groups and only one haplotype was identified in Uthamapalayam. The variation among population is associated with climatic conditions, the availability of oviposition sites, the nature of vector control measures, and the role of human aided-dispersal. Phylogenetic analysis revealed that the Madurai mosquito populations are closely related and suggest a recent divergence from their common ancestor whereas the Uthamapalayam population is distantly related. An Understanding pattern of gene flow among *Culex quinquefasciatus* is useful to trail and predict the movement of important genetic traits such as vector competence and insecticide resistance.
Notes
DENGUE DIAGNOSIS, ADVANCES AND CHALLENGES

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Today, dengue (DF, DHF, DSS) is considered the most important arthropod-borne viral diseases in terms of morbidity and mortality. In this paper, a review is presented focusing on the main advances, problems and challenges of dengue diagnosis. IgM capture ELISA, NS1 antigen detection, virus isolation in mosquito cell lines and live mosquitoes and PCR have all represented major advances in dengue diagnosis. Dengue viremia is short, is usually observed 2-3 days before the onset of fever and lasts 4-5 days later. Therefore, samples for virus isolation, NS1 antigen detection must be taken in the first 4-5 days of the disease. For IgM antibody detection the samples must be taken on later stage i.e after 4-5 days. The time of blood collection is very important in dengue diagnosis. Proper timing of blood collection is not perfectly possible during an outbreak study. Therefore it is better to test the samples simultaneously for NS1 antigen and IgM antibody. This will help us from wrong diagnosis of false negatives / positives. Better do both and come to the conclusion. Therefore two separate tests, one for acute stage and another for convalescent stage, are needed for dengue diagnosis.

The continued development of inexpensive, sensitive, specific and easy tests that allow for early dengue diagnosis are still needed. Specifically, the following aspects require the greatest attention: To develop:

(a) Tests for early clinical diagnosis of individuals.
(b) Serological tests able to differentiate dengue from other flavivirus infections and even more specifically to determine the infecting dengue serotype.
(c) Easy and inexpensive protocols for genomic characteristion and viral load, including those that can be applied in the field.
(d) Modifications of existing protocols that simplify specimen handling and transportation.
(e) Recombinant antigens as tools for test evaluation and to produce these for serological tests.
(f) Tools that can suggest a prognosis, allowing for better management of clinical follow up.
ASSESSMENT OF VULNERABILITY OF BLOCKS FOR DENGUE IN WEST BENGAL USING GEOSTATISTICS

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Background: The re-emerging vector borne diseases like dengue cause significant morbidity in West Bengal. The altered seasonal trend of the disease indicates the necessity of vigilance throughout the year. Since resource constraint may limit regular quality surveillance in all the blocks, prioritization according to vulnerability is necessary to plan any intervention. GIS may fulfill the need of a useful tool to serve the purpose.

Objective: 1. To identify vulnerable blocks for dengue based on last three years data. 2. To compare the dengue affected blocks in 2012 with those identified as vulnerable

Methodology: In this surveillance-based retrospective cum prospective study, vulnerable blocks were identified using data of lab confirmed dengue cases of 2009-2011, applying SQL query through GIS software (MapInfo).

Level of vulnerability was set by criteria like cases more than 3rd quartile in any year or more than median in any two years or adjacent (sharing borders of minimum three quadrants of a block) to the blocks satisfying the first two criteria.

Proportion of blocks already identified as vulnerable, out of the blocks affected in current year (till August 2012), was determined.

Result: Out of 98 blocks ever affected in last three years, we identified 65 blocks as vulnerable. Those 65 blocks corresponds to 10 districts having a total of 220 blocks. Around 62% (13 out of 21) of the affected blocks in the current year, were successfully predicted from the study An urban to semi-urban spread of disease has been observed.
Conclusion: Identification of such vulnerable districts will help the program officials to some extent in order to prioritize their point of intervention. Also other factors have to be searched for to enhance the sensitivity of prediction.

Key Words: Dengue, GIS, Vulnerable
COMMUNICATION IN BEHAVIOUR CHANGE IS AN EFFECTIVE TOOL TO CONTROL DENGUE /CHIKUNGUNIYA VECTOR *Aedes aegypti* BREEDING CONTROL IN THE URBAN AGGLOMERATION OF CHENNAI METROPOLIS.

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Dengue cause serious public health problem in the world wide. Dengue hemorrhagic fever (DHF) and Dengue shock syndrome (DSS) are severe forms of dengue. Globally, 100 countries have reported dengue with more than 2.5 billion people at risk and an estimated 50 million dengue infection. In India, dengue fever has been reported in since 19th century and dengue infection reported all parts of India. In India, Tamil Nadu state is dengue endemic area and highest rate of dengue infection incidence was recorded in Chennai city during 2001. There are different kinds of social, behavioural, cultural, biological and ecological factors influences the dengue vector mosquito breeding in the ecosystem. In absence of vaccine and specific medicine, vector control through source reduction is the only method which is available universally for the control and prevention of dengue fever. The source reduction of *Aedes* will be implemented effectively by promoting community participation through the behavioral change communication methods. In Chennai, due to water scarcity, majority of householders stored water in cemented tank( CT) and cemented cistern(CC) of >100 liter storage capacity. The objective of the present study was to develop evidence based tool which promote community behavior on control of Aedes mosquito breeding in the potential cemented containers. Four cluster of areas were selected which mainly constitute of multistory building where the people used cemented tank largely for water storage. A total of 2819 Households (HHs) were surveyed among the four clusters. Out of this total, 559 HHS were using either CT / CC for water storage. Group meetings were conducted among the women members to educate them about dengue fever, its cause, mode of transmission and methods to control vector mosquito breeding. Among the total 559 HHs, 517 HHs were selected and issued Vector Control Monitoring (VCM) cards which contains the respective
family head’s name and address. In a table printed in the card, the respondents note down the date of cleaning, checking the presence of larvae and pupae in the CT/CC and material used for cleaning (scrubber/bleaching powder etc.). The picture of mosquito life cycle (egg – larvae- pupae-adult) was drawn on the VCM card. A separate picture of larvae and pupae was shown on near the table of entry for easy identification by the community. In each HHs the respective HH members were asked to clean every five days to avoid *Ae.aegypti* breeding. IEC meetings were conducted to women members to explain the method cleaning CT/CC. The researcher monitored VCM card entries made by the householders at monthly once. During the initial period the container breeding status was 24% to 58%. After six months intervention the containers breeding status was recorded as 15 %. The householders continued cleaning practice of CT/CC. Continues monitoring and supervision by the volunteers may enhance the sustainability of community cleaning practice.
A SOCIOLOGICAL STUDY TO UNDERSTAND PEOPLE’S KNOWLEDGE, ATTITUDE AND PRACTICES ON DENGUE FEVER AMONG THE URBAN COMMUNITY IN CHENNAI, INDIA

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Dengue is a viral borne disease which is the leading cause of illness and death in the tropics and subtropics. Globally 100 million people are infected yearly and one-third of population is under the risk of dengue infection. There are four types dengue virus which spread through infective bites of Aedes mosquitoes. There is no specific treatment for dengue and early recognition and prompted diagnosis will lower the severity of dengue infection. Aedes aegypti, the principal vector of dengue is anthropophilic species, breeds in and around the human dwellings and feeds on human blood. There are different kinds of environmental, social, economical and biological factors influence the Aedes breeding in the urban and rural agglomeration. Since no vaccine/specific treatment available for dengue, vector control is the only available method in the country. The community involvement is needed for the effective implementation of dengue vector control. The increasing of people knowledge will help to promote good behavior on mosquito breeding control by the community. The present study explored the knowledge, attitudes and practices on dengue fever and its mosquito breeding control among the selected adult population of study area. The study was conducted in two clusters namely Pallavan Nagar, Kasima Nagar from North Chennai, Tamil Nadu. A total of 200 households were selected for this study. One respondent who is more than 18 years in the selected household was selected for the interview. A well pre tested structured questionnaire was used for the data collection. The details of people’s socio demographic characters, their knowledge on dengue fever, mode transmission, method of prevention and water storing practices were collected. A composite scoring system followed, based on the answers given in the questionnaire, was used to establish the level of awareness in the
population. The division of the higher and lower socio-economic groups were based on their income and locality; both these variables were determined as a part of our survey. Among the total 200 HHs, 67 were male and 133 were female. About seventy three percent of the respondents had adequate knowledge about dengue fever and its vector. The community knowledge on dengue had significantly associated with the socioeconomic status. The better dengue preventive practices were observed among the high socioeconomic groups when compare to other groups. Knowledge on dengue was inadequate in the low socioeconomic group. Better preventive practices against the vector were prevalent in the high socioeconomic group. Hence, a greater focus should be accorded to the low socioeconomic areas in future dengue health education campaigns.
A STUDY ON MIGRATION OF PEOPLE AFFECTING ELIMINATION OF LYMPHATIC FILARIASIS IN SURAT CITY, INDIA

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Lymphatic filariasis (LF) is one of the major health problems in India and Surat city of the LF endemic city in Gujarat province of India. LF is chronic disease and urges the patient to face the social, economic and physical hazards. The most practical and feasible prophylactic way to inhibit the disease is Mass drug Administration (MDA) launched by World health assembly as result of its resolution to eliminate lymphatic filariasis to assist member states to achieve this goal by 2020. India wishes to eliminate filariasis by 2015.

MDA has been or being done in 51 of the 71 eligible countries including Surat city of Gujarat state in India for achieving LF elimination, however the success has been inconsistent after completing of 8 round of MDA following the norms of national guidelines. Assessable impact of MDA through National Filaria Control Programme (NFCP) by routine Night Blood Surveys (NBS) since 2001 to 2011 showed the significant role of contributing microfilaria carrier to Surat city by highly endemic state like Odisha 436 (51%), U.P 162(19%), Bihar 48(6%) and rest of the other states. Migration habitats of the people of highly filarial endemic states highly influence the transmission of the filaria in the Surat city and fade the effect of MDA. In addition to pre MDA baseline survey was carried out since 2004 to 2011. A total of 121789 NBS have been collected prior to MDA during the year 2004 to 2011. Out of it 878 microfilaria (mf) carriers were detected. State wise contribution of carriers were revealed that Odisha predominant 435(49%), U.P. 146 (17%), Surat city 109 (12%), Bihar 63 (7%) and others 125 (15%). Entomological data from the year
2004 to 2011 represents that infection & infectivity rate are declined 95.83% (0.24 to 0.01) and 85.71% (0.07 to 0.01) respectively. 10 men hour density of *Culex quinquefasciatus* were also reduced 43.10% (93.15 to 53.00) during 2004 to 2011.

The above analysis reveals that though the overall Surat city has been showing the declining trend in microfilaria rate with increasing coverage & compliance of DEC but the group of the population migrate from endemic areas in a particular month harbouring microfilaria can be a potential for transmission and therefore the timing for their migration and the area need to be kept under vigilance so the immediately after arrival their screening can be done for microfilaria and if they have not been given DEC in their respective areas, they should be immediately administered the tablet.

Key words: Lymphatic filariasis, migration, endemic, vector and Mass Drug Administration
EFFECTIVE COMMUNITY PARTICIPATION TO REDUCE MALARIA IN MUMBAI METRO CITY (MAHARASHTRA)

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Malaria is one of the most problematic Vector Borne Disease in Public Health. Resurgence of Malaria in urban area Mumbai Metro City is seen during the years 2010 to 2011. Community participation to reduce transmission and reservoir of infections through vector control. Effective advocacy was carried out to increase awareness and commitment for control of Malaria. Supportive environment was helpful for adequate human resource through community participation and active involvement. Inter sectoral co-ordination through other Departments like Railway, Air-force, Navy, Education, Public Works Department, Non-Government Organizations and Private Sectors are actively involved for vector and parasite control. Impact of above activities resulted in reduction of Malaria Positive Cases.
DEVELOPING A COMMUNITY BASED MODEL FOR EFFECTIVE MALARIA CONTROL IN A KNOWN ENDEMIC AREA IN METROPOLITAN CITY

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A prospective interventional study on malaria was executed in the year 2010 from June to December in high prevalent area in Mumbai city through PSM Department in KEM Hospital. The present study identifies the epidemiological factors responsible for malaria control leading to development of a community-based model for malaria risk reduction. Data from the IDSP Unit, established in KEM Hospital in 2009 indicated alarming increase of malaria cases & deaths provoking need for alternative interventional strategies for malaria control.

Based on the past experience a community-based strategy was evolved & implemented in the year 2010 from June to December 2010, with preparatory planning in May 2010.

A high malaria risk area (API >2) was randomly selected in F-South ward area of Mumbai city. This predominantly residential area was divided into 2 parts; one part was for new interventional strategy and second without any interventional strategy by the investigators.

A coordination team of investigators in collaboration with local public health authority jointly evolved the comprehensive strategy. An integrated approach focusing on managerial, technical & community mobilization was implemented in interventional study area while the other area served as control without any comprehensive strategy but exposed to routine control activities implemented by local public health authorities.

The overall morbidity mortality rates indicated drastic decline of morbidity and zero mortality as compared to the previous years as well as in the control area. The decrease SPR, MBER in the interventional area further strengthen the effectiveness of the interventional strategy.
BIOTECHNOLOGICAL AND SOCIOLOGICAL “CHALLENGES IN VECTOR CONTROL”

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Biotechnological researches play a vital role in the advancement of technical tools, which have been utilized for promotion of public wealth and health. The application of biotechnological researches has been increased in this volume on various fields viz., agricultural, medicinal and mosquito borne disease control. In this paper, we have discussed about biomedical challenges in mosquito vector control in the sociological perspective viz., knowledge gap, and lack of people participation and community acceptance of new methods. Worldwide, half of the population is at risk by different kinds of vector borne diseases like malaria, dengue, filariasis, Japanese encephalitis and chikungunya. Biotechnological research has created some opportunity for the control of malaria and dengue through genetically engineered mosquitoes and the control JE vector mosquitoes through neem - coated urea in paddy fields. But the field level implication of these tools is still a big challenge because of the lack of understanding on socio-political situations, cultural and historical situations, lack of public knowledge and practices on particular disease spreading vectors and challenges in community to the disease control progammme. So the sociologist plays a crucial role for the successful implementation of biological control programmes. In this paper, we have discussed the unique responsibility of social science researches to exploring the people knowledge, cultural behavior, social and political role before implementation of biological product invented by biotechnical methods.
KAAP STUDY ON THE MOSQUITOES AND MOSQUITO-BORNE DISEASES AMONG SCHOOL CHILDREN IN THIRUMANGALAM

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An attempt was made to conduct a KAAP study among the school children in and around Thirumangalam town. One hundred boys and girls were given questionnaire to find out their knowledge, awareness, attitude and practice with reference to mosquito menace and mosquito-borne diseases. It was found out that they have a better knowledge about the feeding and breeding behaviour of the mosquitoes, the diseases caused by them and the management strategy. However, a small variation was noticed in between the boys and girls with respect to their expressions. The school children have learnt at various standards through the curriculum about the mosquitoes and mosquito-borne diseases and so they are able to express their opinions and understanding. It is suggested that school children could be used to create awareness among the general public and their participation in the management of both mosquito menace and mosquito-borne diseases. The other details are discussed in the paper.
NEW INSIGHTS TO THE EXPLOITATION OF MYCOPATHOGENS FOR MANAGEMENT OF MOSQUITOES: CHALLENGES AND OPPORTUNITIES

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Mosquito-borne diseases continue to have a significant impact on human health worldwide. Numerous information sources discuss mosquito biology, mosquito-borne diseases, methods of personal protection, and approaches to mosquito control. Approximately, one million death cases of malaria occur annually. Therefore, the management of mosquitoes is important for reducing their impacts on health and lifestyle. Currently available methods to mitigate mosquitoes include use of chemical pesticides and non-infective transgenic mosquitoes, but both methods have their own limitations. Furthermore, mosquitoes are becoming resistant to chemical pesticides while genetically engineered transgenic mosquitoes are not providing a practical or feasible solution to mitigate mosquito population. Resistance management strategies such as the use of chemical mixtures or rotations are suggested to improve the sustainability of these approaches, but with significant cross-resistance between the currently approved classes of insecticides, practical options are very few to contain the life-threatening vector-borne diseases. In recent years, interest in mosquito-killing fungi is reviving mainly due to increasing levels of insecticide resistance and increasing global risk of mosquito-borne diseases. Fungal mosquito-pathogens in the genera such as Lagenidium, Coelomomyces and Culcinomyces are known to decimate mosquito populations in spectacular epizootics. Many isolates of entomopathogenic fungi such as Beauveria and Metarhizium have also been investigated extensively as a new mosquito control agents because insecticide resistance is preventing successful mosquito control in many countries. Many of these fungi found to provide promising alternatives to chemical control through their unique mode of action. A handful of mycoinsecticide formulations have been commercialized. This paper attempts to addresses on the new insights to the exploitation of mosquito-pathogenic fungi, their potential and drawbacks to be used as biological control agents in mosquito abatement programme.

Key words: Myco-pathogens, mosquito management, public health.
MOSQUITO LARVICIDAL ACTIVITY OF ACTINOBACTERIAL ISOLATES AGAINST ANOPHELES AND CULEX LARVAE

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Mosquitoes, in almost all tropical and subtropical countries are responsible for the transmission of pathogens causing some of the most life-threatening and debilitating diseases of man, like malaria, yellow fever, dengue fever, chikungunya, filariasis, encephalitis etc. Repeated use of synthetic insecticides for mosquito control disrupts natural biological control systems. The use of biological control agents is therefore essential. Primary screening was carried out to evaluate the larvicidal activity of the actinobacterial isolates against the Culex and Anopheles mosquito larvae. Mosquito larvae were collected and identified as Culex and Anopheles mosquito larvae by their morphological characterization. Among 43 isolates, only five isolates were selected due to their significant larvicidal activity. The selected isolates were identified based on their morphological biochemical, physiological and molecular characterization. The isolates KA1-3, KA(4), KA6, KA5(10), KA5(A) were identified as Streptomyces sp, Streptomyces sp, Streptoverticillium sp, Streptomyces sp, Nocadiopsis sp respectively. Of the five isolates two potent isolates KA5(4) and KA5(10) were chosen for molecular taxonomic characterization. These two isolates were found to be Streptomyces sp and Streptomyces violaceorubidus by 16S rRNA sequence analysis. The Phylogenetic relationship with other potent actinobacterial isolates, 16S rRNA secondary structure prediction, restriction sites and GC content in 16S rRNA sequence were studied in detail. The larvicidal compound was extracted from KA1-3 and compound was evaluated for mosquito larvicidal activity. Secondary screening revealed that the crude extract of larvicidal compound have significant larvicidal activity at 1% concentration. The present findings reveals that the larvicidal activity of secondary metabolites of actinobacteria can be an alternative source for present chemical larvicides because they constitute a potential source of bio active chemicals and generally free from harmful effects. Further research is extended to find out the mass production, purification, physiochemical nature of larvicidal compounds and their invivo efficacy in laboratory and field trials.
DEVELOPMENT OF A RECOMBINANT BACILLUS SPHAERICUS WITH THE GENES ENCODING CYTOLYTIC AND CHAPERONE PROTEINS OF BACILLUS THURINGIENSIS SUBSP. ISRAELENSIS

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The Bacillus Sphaericus, which produces a binary toxin is a mosquitocidal bacterium developed as a commercial larvicide that is used currently in operational mosquito control programs in several countries. Wild type B. sphaericus is highly active against larvae of Culex and Anopheles mosquitoes; it is virtually nontoxic to Aedes aegypti, an important vector species and unfortunately, it is at high risk for selecting resistance in mosquito populations. Because its binary toxin only binds to a single receptor type on midgut microvilli. A potential key strategy for delaying resistance to insecticidal proteins and broad spectrum of activity is to use mixtures of toxins that act at different targets within the insect, especially mixtures that interact synergistically. In the present study, three cytolytic proteins and two chaperones encoding genes cyt1A, cyt2B, cyt1C, p19 and p21 have been amplified from Bacillus thuringiensis subsp. Israelensis (Bti) and cloned into B. sphaericus 1593 by electroporation with the shuttle vector pMK3. All these genes were expressed and confirmed by immunodetection. The level of expression of these genes in recombinant B. sphaericus was probably low when compared to wild type Bti but they are useful in overcoming the resistance to B. sphaericus binary toxin n resistant mosquito population and it showed lesser level of toxicity against A. aegypti. Present study revealed that the recombinant genes restore the susceptibility of the resistant mosquito larval population an through transforming all the crystal protein encoding genes of Bti into B. sphaericus, one can develop the B.sphaericus with higher level and broad spectrum of mosquito larvicidal activity without the development of resistance in mosquito larvae.
PREDATORY POTENTIAL OF LARVIVOROUS FISH, *GAMBUSIA AFFINIS* ON THE IMMATURE STAGES OF LYMPHATIC FILARIAISIS AND JAPANESE ENCEPHALITIS VECTORS

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Development of chemical-associated resistance in mosquitoes by the use of various chemicals to control them and its effects on non-target species evoke a reason to find alternative methods to control mosquitoes. Use of natural predators of mosquito larvae to control larval population has always been an interesting area of research. Fishes are well known biological control agents used for the control of mosquitoes especially malaria vectors. In this study, we have examined the predatory potential of the mosquito fish, *Gambusia affinis* (Baird & Girard) (Cyprinodontiformes: Poeciliidae) on larval stages of *Culex quinquefasciatus*, the principal vector of lymphatic filariasis and *Cx. tritaeniorhynchus*, the major vector of Japanese encephalitis. The 4th instar larvae of these mosquitoes were used as prey in laboratory conditions for 24 hrs. The predation experiment was conducted with three replicates. It was revealed that, the per day average consumption rate of *Gambusia* was 108.33 (n=120) for 4th instar larvae of *Cx. quinquefasciatus* and 81.94 (n=120) for *Cx. tritaeniorhynchus* respectively. On the whole, 91.27% of *Cx. quinquefasciatus* (n=120) and 82.94% of *Cx. tritaeniorhynchus* (n=120) 4th instar larvae had been consumed by the larvivorous fish for per day. These findings pointed out that this larvivorous fish, have slightly better preference for the filariasis vector. The experiments further indicate that *Gambusia affinis* can act as a potential bio-control agent to control immature stages of these important mosquito vectors. The study suggests that this fish may be used, after a careful area specific field trial, as a promising and sustainable biological control agent in controlling filariasis, Japanese Encephalitis vectors.
Notes
MOSQUITOCIDAL ACTIVITY OF MORINDA CITRIFOLIA LEAF EXTRACT AGAINST ANOPHELES STEPHENSI AND Aedes aegypti (DIPTERA: CULICIDAE)

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Vector-borne diseases are still major public health problems in the Southeast Asian countries because of their tropical or subtropical climate, due to the frequently poor drainage system especially during rainy seasons and the presence of many fish ponds, irrigation ditches and rice fields which provide abundant mosquito breeding places. Malaria is commonly associated with poverty, but is also a cause of poverty and a major hindrance to economic development. Aedes aegypti is known to carry pathogens responsible for dengue, yellow fever and Chikungunya. The present investigation an attempt has been made to study on the larvicidal and pupicidal effect of Morinda citrifolia on malarial vector, Anopheles stephensi and dengue vector, Aedes aegypti. Lethal Concentrations (LC₅₀ and LC₉₀) were calculated using the observed mortality. Observations were made on the efficacy of plant extract. The mortality of larval populations was noted at different period of exposures. Therefore, M. citrifolia used to control mosquito vector effectively by emitting active substances in the breeding habitats of vector species, A. stephensi and Aedes aegypti. In the review of current state of knowledge on larvicidal and pupicidal activity, extraction processes, botanical pesticides effects on non-target organisms for the control mosquito vectors.

Keywords: Morinda citrifolia, Anopheles stephensi, Aedes aegypti, Larvicidal and pupicidal activity
COMBINED EFFECT OF *ACALYPHA ALNIFOLIA* AND MICROBIAL INSECTICIDE, SPINOSAD AGAINST MALARIAL VECTOR, *ANOPHELES STEPHENSI*.

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Malaria is a parasitic disease from which more than 300 million people suffer yearly throughout the world. It is one of the main causes of infant and young child mortality. In India, malaria is transmitted by six vector species, in which *Anopheles stephensi* is responsible in urban areas. Synthetic insecticides are toxic and adversely affect the environment by contaminating soil, water and air. The chemicals derived from plants have been projected as weapons in future mosquito control program as they are shown to function as general toxicant, growth and reproductive inhibitors, repellents, and oviposition-deterrent *Acalypha alnifolia* are the medicinal plants also possess mosquitocidal properties. In the present study, *A. alnifolia* leaf extract and microbial insecticide, Spinosad was treated against *A. stephensi* larvae and pupae. Lethal concentrations (LC$_{50}$ and LC$_{90}$) were worked out for the after treatment. The results reveals that the leaves extract of *A. alnifolia* and microbial insecticide, Spinosad is promising as good larvicidal and pupicidal properties of against *A. stephensi*. This is an ideal eco-friendly approach for the control of malarial vector, *A. stephensi* as a target species of vector control programs.

*Key words: Acalypha alnifolia, Spinosad, A. stephensi, mosquito control.*
PREVENTION OF MOSQUITO BORNE DISEASES THROUGH SELF HYGIENE AND ENVIRONMENTAL PRACTICES

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Drain the water from garbage cans, house gutters, pool covers, coolers, toys, flower pots or any other containers where sprinkler or rain water has collected. Discard the old tires, drums, bottles, cans, pots and pans, broken appliances and other items that aren’t being used. Clean the birdbaths and pet’s water bowls at least once or twice a week and protect the boats and vehicles from rain with tarps that don’t accumulate water. Maintain the water balance (pool chemistry) of swimming pools and empty plastic swimming pools when not in use. Cover your skin with clothing and use mosquito repellent. If you must be outside when mosquitoes are active, cover up wear shoes, socks, long pants, and long sleeves. Apply the mosquito repellent to bare skin and clothing. Always use repellents according to the label. Repellents with DEET, picaridin, oil of lemon eucalyptus, and IR3535 are effective. Use mosquito netting to protect children younger than 2 months. And cover the doors and windows with screens to keep mosquitoes out. If you keep mosquitoes out of your house, repair the broken screens on windows, doors, porches, and patios. Limit the outdoor activity at when mosquitos are most active also avoid areas where there are a lot of mosquitoes. Contact your local mosquito control agency if there is a significant mosquito problem where you live or work. Fill in holes or dips in the ground that collect water. Level the ground around your home so water can run off. Stock your ornamental water garden with mosquito-eating fish (minnows, gambusia, goldfish, or guppies).

Key words: Repellent, DEET, Picaridin, Mosquito – eating fish.
MOSQUITO CONTROL IS A VITAL PUBLIC HEALTH PRACTICES

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Mosquito control manages the population of mosquitoes to reduce their damage to human health, economies, and enjoyment. Mosquito control is a vital public-health practice throughout the world and especially in the tropics because mosquitoes spread many diseases, such as malaria. Mosquito-control operations are targeted against three different problems: 1. Nuisance mosquitoes bother people around homes or in parks and recreational areas; 2. Economically important mosquitoes reduce real estate values, adversely affect tourism and related business interests, or negatively impact livestock or poultry production; 3. Public health is the focus when mosquitoes are vectors, or transmitters, of infectious disease. Disease organisms transmitted by mosquitoes include West Nile virus, Saint Louis encephalitis virus, Eastern equine encephalomyelitis virus, Everglades virus, Highlands J virus, La Crosse Encephalitis virus in the United States; dengue fever, yellow fever, Ilheus virus, malaria and filariasis in the American tropics; Rift Valley fever, Wuchereria bancrofti, Japanese Encephalitis, chikungunya, malaria and filariasis in Africa and Asia; and Murray Valley encephalitis in Australia. Generally, some techniques are used depending on the situation, source reduction, biocontrol, larviciding (control of larvae), or adulticiding (control of adults) and DDT may be used to manage mosquito populations. These techniques are accomplished using habitat modification, pesticide, biological-control agents, and trapping.

Key words: Repellent, Biocontrol, larviciding, adulticiding, DDT.
USE OF INSECT REPELLENTS FOR BETTER HEALTH
AND HYGIENE

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The prevention of mosquitoes to take the steps to reduce the mosquito population around your home. Using insect repellents and protective clothing also can help. Mosquitoes need stagnant or standing water to breed. Eliminate standing water, especially after rains, and you can reduce the mosquito population around your home and yard. When used properly, repellents are safe for kids and adults alike. Keep in mind that repellents don’t kill mosquitoes; they just make it harder for them to find you. That means even when a repellent is working, you may still see these annoying insects buzzing about. The commonly used repellents include DEET. The pesticide DEET blocks a mosquito’s ability to find people who’ve applied it. Apply the repellent with up to a 35 percent concentration of DEET to your skin and clothing. Choose the concentration based on the hours of protection you need — generally, the higher the concentration of DEET, the longer you are protected. A 10 percent concentration protects you for about two hours. Keep in mind that chemical repellents can be toxic, and use only the amount needed for the time you’ll be outdoors. Don’t use DEET on the hands of young children or on infants younger than age 2 months. The Picaridin repellent, also called KBR 3023, offers protection that’s comparable to DEET at similar concentrations. It also blocks a mosquito’s ability to find people who’ve applied it. Picaridin is nearly odorless, which may make it a good alternative if you’re sensitive to the smells of insect repellents. The Oil of lemon eucalyptus is a plant-based chemical may offer protection that’s comparable to low concentrations of DEET. Don’t use this product on children younger than 3 years. Others like shorter acting repellents — such as citronella — may offer limited protection. Check the labels of insect repellent products to see which chemicals or other ingredients they contain. And be sure to follow the product’s application guidelines. When you come indoors, wash your skin and your children’s skin with soap and water to remove any remaining repellent.

Key words: Repellent, DEET, Picaridin, Oil of lemon.
ANTIPLASMODIAL PROPERTIES AND BIOASSAY-GUIDED FRACTIONATION OF THE METHANOLIC EXTRACTS FROM SOME CAMEROONIAN MEDICINAL PLANTS.

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Developing countries, where malaria is endemic, depend strongly on traditional medicine as a source for inexpensive treatment of this disease. However, scientific data to validate the antimalarial properties of these herbal remedies are scarce. Consequently, it is important that antimalarial medicinal plants are investigated, in order to establish their efficacy and to determine their potential as sources of new antimalarial drugs.

In this study, we evaluated the claimed antimalarial properties of ten fractions from two plants used in traditional medicine against malaria and fever, mainly in some African and Asian regions. The air dried powdered plant leaves were extracted with methanol as solvent. Further, the fractionation of the methanolic extract was done using solvents from the less to the most polar. The last water fraction in case was also tested. The blood schizontocidal activity was measured using a standard in vivo assay, with Plasmodium berghei K173 strain and in vitro assay using Plasmodium falciparum. Fractions \( F_2 \) and \( F_3 \) of the first and second plant species were the most potent and have shown a significant activity in vivo as well as in vitro \((P<0.05)\). The bioassay-guided purification and characterisation of the most active fractions is ongoing.

Key words: Traditional medicine; Medicinal plants; Antimalarial; Plasmodium berghei K173.
EFFECT OF LEAF EXTRACT OF JATROPHA CURCAS ON THE SURVIVAL AND DEVELOPMENT OF THIRD INSTAR LARVAE OF CULEX QUINQUEFASCIATUS AND Aedes Aegypti

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Bioefficacy of petroleum ether leaf extract of Jatropha curcas was studied on the survival and development of the two vector mosquitoes Culex quinquefasciatus and Aedes aegypti at 10 different concentrations. The extract showed 100 percent larval mortality at 20ppm concentration for Culex quinquefasciatus and Aedes aegypti. LC50 values were 6.46ppm and 7.94ppm respectively to Culex quinquefasciatus and Aedes aegypti. Adult emergence was reduced in both species and larval – pupal durations were also prolonged with increasing concentrations.

Key note: Jatropha curcas, Culex quinquefasciatus and Aedes aegypti
EVALUATION INSECTICIDAL PROPERTIES OF AN INDIGENOUS PLANT, *CITRULLUS COLOCYNTHIS* (SCHRAD) AGAINST MOSQUITO LARVAE UNDER LABORATORY CONDITIONS

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Bioefficacy of acetone seed extract of *Citrullus colocynthis* was studied on the survival and development of the two vector mosquitoes *Culex quinquefasciatus* and *Aedes aegypti* at different concentrations. The extracts showed 100 percent larval mortality at 50ppm concentration for *Culex quinquefasciatus* and 40ppm concentration for *Aedes aegypti*. LC$_{50}$ values were 21.4ppm and 16.7ppm respectively of *Culex quinquefasciatus* and *Aedes aegypti*.

Adult emergence was decreased with increasing concentration of the test solution. Larval and pupal duration were prolonged with increasing concentrations. Third instar of *Culex quinquefasciatus* was found to be more susceptible to the seed extract of *Citrullus colocynthis* than *Aedes aegypti*.

Key note: *Citrullus colocynthis, Culex quinquefasciatus* and *Aedes aegypti*. 
Notes
INFLUENCE OF MANAGEMENT PRACTICES ON THE CONGLOMERATION POTENTIAL OF VECTOR BORNE MOSQUITO SPECIES AND THEIR PREDATORY ARTHROPODS IN AN IRRIGATED RICE ECOSYSTEM

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Irrigation to rice field depends upon the mode and source of irrigation practices of the semi-arid zones of south India. Present study was focused on the assemblage and application patterns of aquatic arthropods and pesticides and fertilizers over the constellation of vector borne mosquito species and their predators. Predatory efficiency of controphic arthropods such as the nymphs of dragon fly, Cyclopoid copepods and field collected tadpoles over Culex tritaeniorhynchus Giles were chosen for this investigation in rice fields with different management practices such as rice field with canal irrigation, rice field with pond irrigation and rice field with well irrigation. Ex-situ studies on cages on site revealed that efficiency of controphic predatory species over Culex tritaeniorhynchus lies with the size of the larvae. In all the three types of rice fields the copepod prefer only first instar larvae of Culex and the number of larvae consumed an average of 30 per day. In rice with pond irrigation and canal irrigation predatory dragon fly nymph consumed an average of 20 third instar larvae and 25 fourth instar larvae per day. The tadpole consumed maximum number of larvae of all the instars in all the three types of rice field. But applications of both fertilizer and pesticides over the rice field showed different results on the predatory potentials of the controphic species. There was no reduction in the consumption of Culex sp by tadpoles whereas dragonfly nymphs and cyclopoid copepods showed significant reduction in the predatory potentials. The results clearly revealed that the basic principle of community ecology of mosquitoes in general and C. tritaeniorhynchus in particular and their interaction with controphic species is important to understand the prey-predator relationships in the ricefield environment.
The effect of *Acorus calamus* (Aeraceae) on larvae of *Culex quinquefasciatus* Say. was studied under laboratory conditions. Preliminary screening of Calamus oil at 500ppm showed 100 percent larval mortality were recorded after 24hours. At various concentrations, viz. 12.5, 25, 50, 100 and 200ppm, the oil caused concentration dependent larval mortality. The LC₅₀ and LC₉₀ of Calamus oil were 127.4 and 283.3ppm respectively for larvicidal activities. The results suggest that the Calamus oil is an effective larvicidal agent against *Cx. quinquefasciatus*.

**Key words:** Larvicidal, *Acorus calamus*, *Culex quinquefasciatus* Say.
RESPONSE OF THE FILARIAL VECTOR MOSQUITO, 
*CULEX QUINQUEFASCIATUS* TO SELECTED SYNTHETIC 
AND NATURAL REPELLENTS

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Mosquitoes have become a menace to human beings and other animals not only by their biting nuisance but also by transmitting pathogens causing diseases like malaria, filariasis, chikungunya, Japanese encephalitis, Dengue fever in human beings and a variety of diseases in other animals. As these vector mosquitoes are increasing in number it becomes necessary to control their population effectively by using appropriate methods. Hence in the present work an attempt has been made to assess the response of *Culex quinquefasciatus* to nine repellents in study site at Jaihindpuram, Madurai. Three synthetic repellents (Odomos cream, Goodknight lotion and gel), three plant oils (Citronella oil, Eucalyptus oil and Lemon grass oil) and three leaf extracts (*Mentha piperita, Coleus amboinicus* and *Ocimum basilicum*) were tested for their protection time and suppression effect on the biting population. The crude and water extract of the plants exhibited low protection time and suppression effect but plant oils showed a reasonably high degree of suppression, which were on par with the synthetic repellent. Therefore, it is suggested that the plant oils could be mixed with suitable base and advocated as repellents against the mosquitoes. This would become the eco-friendly products.

Key Words: *Culex quinquefasciatus*; Synthetic repellents; Plant oils; Leaf extracts.
EFFECT OF BASAL MEDIA ON MICROPROPAGATION TECHNOLOGY IN *AZADIRACHTA INDICA* (NEEM) FOR MOSQUITO LARVAL CONTROL

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During recent years many advances have been made in the development of mosquito larval control. Neem callus is one of the biological control to reduce mosquito larval populations. Micropropagation is the application of tissue culture technology for mass propagation of any economically important plant species, micropropagation is a practice of rapidly multiplying stock plant material to produce a large number of progeny plants from very small plant parts such as buds, nodes, leaves and root segment etc. Plant tissue culture micropropagation technology needs the following steps - explants selection, surface sterilization, media preparation, and inoculation of explants material. The time required for callus induction is 2-4 weeks. Several types of media were used for plant tissue culture technology. Experimentally were used three types of media such as Woody Plant Medium (WPM), Gamborg B5 Medium (B5) and Murashige & Shouog Medium (MS), effect of WPM medium was productive in callus induction with frequency 71%, effect of B5 medium was productive in callus induction with frequency 73%, MS medium was more effective compared another two media and productive in callus induction with high frequency 80%. Recent year the bio-control agent neem callus extract is a well known environment friendly method for mosquito larval population control.
EICHHORNIA CRASSIPES PROVIDE CONDUCIVE ENVIRONMENT FOR BREEDING MOSQUITOES

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The present investigation was carried out to determine the role of aquatic vegetation in mosquito breeding and habitat selection of adult mosquitoes in Vaigai river in Madurai city. We found that Eichhornia crassipes supports mosquito breeding particularly the immature. The identified larvae were of Culex tritaeniorhynchus and Anopheles subpictus. Further the nutrient rich sewage water supporting both the growth of high mass of floating vegetation and growth of mosquitoes. The association between aquatic vegetation density and the mosquito larval communities present in Vaigai River was determined by visual estimate of emergent vegetation made on each site and each visit. The total and marginal emergent vegetation was estimated by visually ranking the percentage of vegetation cover. Sites were ranked as “low” when estimates of each marginal and total emergent vegetation cover fell between 1 - 25%, “moderately low” if each of the vegetation cover was between 25 - 50%, “moderately high” if the vegetation cover was between 50 - 75% and “very high” if estimates fell between 75 - 100%.
PREFERENTIAL HABITAT SELECTION BEHAVIOR OF MOSQUITOES IN VARIOUS ECOSYSTEMS

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The successful proliferation of mosquitoes in an area depends upon the availability of many habitats and also different types of habitats. Four ecosystems were selected in Sirumalai Hills area, which is located in Dindigul District, Tamil Nadu to study the breeding behavior of mosquitoes with reference the preference the preferential habitat selection. Systematic larval collections were made in the available habitats for a period of two years, 2010 – 2012. Fourteen habitats (both on the ground and above ground habitats) were chosen by more than 30 species of mosquitoes belong to Aedes, Anopheles, Armigeres, Culex, Lutzia, Uranotaenia and Toxorhynchites. The other details are discussed in the paper.
SPATIAL VARIATION IN THE BIODIVERSITY OF MOSQUITO FAUNA IN SIRUMALAI HILLS, TAMILNADU

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Faunistic survey of the mosquitoes in various land scapes, major ecosystems and even in small ecozones is essential and this will help to recognize the presence of vectors, which are known for the transmission of Vector borne diseases. In this context, Sirumalai Hills, an area that is located in Dindigul District, Tamil Nadu was chosen. Systematic larval and adult collections were made for a period of two years (2010 – 2012) in four selected zones, which include forest, village, paddy field and coffee estate.

During this survey, forty two species were recorded. The occurrence of mosquitoes among the selected sites showed a variation. The spatial variation in the occurrence of mosquitoes among various habitats is correlated to the diversity of habitats and vegetation. Species richness of mosquitoes was confirmed.
ON THE BIOLOGY OF SIMULIUM GRAVELYI
(DIPTERA: SIMULIIDAE) IN INDIA: TAXONOMY, ECOLOGY
AND MOLECULAR DATA

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Simulium gravelyi (Diptera: Simuliidae) is a species of blackfly widely
distributed in Palni hills of southern India. In order to understand the current
status and importance of blackfly in India, the biology of S. gravelyi of Palni
hills of southern India was studied. The larva of S. gravelyi is described for
the first time. The taxonomical study indicated that S. gravelyi is close to S.
gurneyae. The lower relationship was obtained ($\alpha^2 = 0.2$) between larval
abundances and six different elevational sites. The highest larval abundance
was occurred during November. Assemblage of larvae was higher on leaf litter
substratum followed by boulders/cobbles, woody debris, rocky and gravels/
pebbles and waste discharges. Statistical analyses predicting two major
ecological factors (conductivity and current velocity) were responsible for the
distribution of larvae. The molecular characterization was analyzed based on
ITS1 and ITS4 sequences. The analyzed sequences were closely related to
the vector species of Simulium vittatum in their ITS sequences. These results
also facilitate an understanding of the species groups of Indian black flies,
and agree with the current understanding of relationships.
MEDICALLY IMPORTANT ARTHROPODS IN A HOUSING
COMPLEX AND ITS SURROUNDINGS

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About 75% of all animal species described so far belong to the Phylum
Arthropoda. No other group of animals approaches this magnitude of species
richness. They occupy various ecological niches in environment. History of
co-existence of certain arthropods and humans in the made- made environment
has been known for a long time. Many arthropod species are of medical
importance since they can mediate transmission of a variety of diseases. For
example, mosquitoes, sand flies, house flies, cockroaches, fleas, ticks, mites,
lice etc. Certain arthropods may inflict venom or poisonous fluids by
biting (eg., spider) and stinging (eg., scorpion, bees ). Keeping in view the above
perspectives, an attempt has been made to know the prevalence of medically
important arthropods (MIAs) in a housing complex having parks and gardens
and its surroundings in Calcutta. The MIAs that have been observed and
collected during a preliminary survey in the month of July, 2012 in the studied
area are mosquitoes (Cx. quinquefasciatus, Ae. aegypti, Ae. albopictus, Armegères
subalbatus), house flies (Musca domestica), cockroaches (Periplaneta
americana), scorpions, spiders, bees (Apis sp.). Role of certain MIAs in the
context of human health has been reviewed.
FACTORS AFFECTING THE SEASONAL PREVALENCE OF MALARIA INCIDENCE IN KEONJHAR DISTRICT OF ORISSA, INDIA

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The transmission of malaria is governed by local and focal factors leading to vectors abundance under favorable conditions. Odisha state with 4% of the total population of India, accounts for 47% of Plasmodium falciparum (Pf) cases and 24% of all reported deaths due to malaria in the country. Keonjhar is a tribal district (44% tribal population), highly endemic to malaria in Odisha. District surveillance data showed Plasmodium falciparum (Pf) > 93%, Annual Parasite Incidence (API) >17, and deaths 10% of the state (Keonjhar 23 and Orissa 239) in 2008. This district is having 34% of hilly forested areas and extreme climatic conditions.

A study on the seasonal malaria incidence and metrological factors as well as role of Anopheles culicifacies in the district was carried out between 2007 January to 2008 December. A total of 1304 specimens of An. culicifacies were collected. The findings showed density of An. culicifacies started increasing in winter season (6.64 PMHD) and peaked in rainy season (7.1 PMHD) and was least in summer season (4.19 PMHD). It was due to more availability of breeding sites in rainy season. However, malaria incidence was high in summer season (2922) due to higher anthropophilic index (34.7) and sporozoite rate (2.13) in the vector An. culicifacies as compared to rainy (anthropophilic index :26.9; sporozoite rate:1.3) and winter (anthropophilic index : 21.3 ; sporozoite rate: 0.89) seasons. This may be because of the favour of higher temperature for faster malaria parasite development (sporogony) inside the mosquito (i.e. extrinsic incubation period or EIP shortens at higher temperature and vectors become infectious sooner). In summer though breeding sites were less still the population of the vector was maintained, as temperature
also has effect on larval development and pupation rate. The study revealed that malaria incidence in the district Keonjhar is prejudiced by seasonal climatic conditions such as rainfall pattern, temperature and ultimately influencing the bionomics of the vectors.

The improved understanding of the pattern of malaria transmission and information on factors affecting it may pave the way for designing appropriate vector control strategies.
SEASONAL PREVALENCE OF THE VECTOR MOSQUITOES OF JAPANESE ENCEPHALITIS VIRUS IN ALAPPUZHA DISTRICT, KERALA STATE

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Abstract: The disease burden caused by mosquito vectors has increased annually, and agents such as Japanese encephalitis virus have emerged and caused epidemics in Alappuzha. So, a two year entomological study was carried out in Kerala, south India, to identify the mosquito vectors of Japanese encephalitis (JE) virus and to determine their seasonal abundance and their distribution. A total of 46,954 mosquitoes belonging to five genera and 22 species were collected. *Culex tritaeniorhynchus* (71.66%) was the most abundant species in 2009, followed by *Culex gelidus* (13.76%), *Mansonia indiana* (2.61%), and *Mansonia uniformis* (1.54%) and 2010-2011 *Culex tritaeniorhynchus* (68.26%) was the most abundant mosquito, followed by *Cx. gelidus* (14.25%), *Mansonia uniformis* (7.28%), and *Anopheles peditaeniatus* (5.43%) were found to be the most common in these areas. General mosquito population showed bimodal pattern, peak occurred during February and October. Other vector species perhaps have a limited role in the transmission of JE virus due to their extremely poor density.

*Keyword Index:* *Culex gelidus; Culex tritaeniorhynchus; Japanese encephalitis.*
EFFECT OF WATER PHYSICO-CHEMICAL CHARACTERISTICS ON SIMULIIDAE (DIPTERA) PREVALENCE IN SOME STREAMS OF MEGHALAYA, INDIA

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The density and distribution of simuliids in 16 fresh water streams and rivers was examined in the present study. The aquatic stages of simuliids were collected and speciated. Water samples were analysed for various water variables such as, water temperature, pH, dissolved oxygen concentration, dissolved oxygen saturation, conductivity, total dissolved solute, turbidity, resistivity and salinity. Regression was used to determine relationship between simuliids density and water variables, whereas Principal Component Analysis was used to determine water parameters association with Simulium distribution. *Simulium* (*S*) *barraudi*, Puri, *S. (S) striatum*, Brunetti and *S. (S) himalayense* were recorded in the present study. *S. barraudi* was the most abundant (56.8%) and its density was high ($\bar{x}^2 = 289.3; \ df = 2; p<0.0001$) as compared to the other. The average population size of each species was 188.3 whereas, Simpson and Shannon-Wiener diversity indices were 0.4466 and 1.306 respectively. Linear regression shows that simuliids density was associated with the water flow rate. Principal Component Analysis indicated that the water parameters accounted for 42.25% variation along D1 axis, while 24.1% variation along D2 axis. Atleast two principal components have eigenvalue > 1 and accounted for 32.6% of variation. Turbidity, water flow and pH are important water parameters affecting the simuliid species prevalence and density in rivers. The study indicates that each simuliid species prefer different sets of physico-chemical parameters in breeding habitat, which are specific to that particular species. Therefore simuliid species community as a whole cannot be considered as a suitable indicator of the streams water quality.

Key words: Simuliids, immature stages, density, water parameters, Meghalaya.
PROPAGATION OF STEGOMYIA ALBOPICTA (SKUSE) IN TO URBAN AREA THROUGH CEMETRY VASES

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During the course of dengue vector surveillance around the town of Coonoor, Nilgiri during post monsoon season in between 2011-2012. So far there is no record of dengue vectors in residential area in the Nilgiris, however Stegomyia albopicta (skuse) is the dominant species in the forest area. It has been observed that the sylvatic species Stegomyia albopicta is propagating through cemetery vases in the yards located in forest fringe and semi urban areas, to waste tyres on the road sides and then to residential areas of the town. Even though this kind of propagation has been recorded elsewhere in the world, this is the first report form India. The survey was conducted in three occasions in following locations-one cemetery yard each in forest fringe and semi-urban area, four road side locations having unused tyres, four residential areas (2 semi-urban and 2 urban areas). Most of these breeding habitats were holding rain fed water except a few portable water storing containers around human habitats. It has been found that the most preferred breeding habitat of Stegomyia albopicta being cemetery cases with 74.5% (38/51), followed by waste tyres with 61.5% (144/234) and the other containers in nearer to human habitations with 20.7% (45/217) positivity rates respectively. Interestingly 71.4% of ovi position rate and 29.6% of mean egg density were recorded in a semi-urban area in an occasion during June 2012 which indicates the spread of this species towards the residential area. The mode in spread of Stegomyia albopicta, a sylvatic species and its establishment nearer to human habitation occupying a breeding habitat of the domesticated mosquito Stegomyia aegypti (Linnaeus). A single specimen of St. aegypti was obtained through ovi trap. The propagation mode of the dengue vectors are discussed in detail.
INTERSPECIFIC MATING BETWEEN *Aedes aegypti* (L.) AND *Aedes albopictus* (Skuse) UNDER LABORATORY CONDITIONS IN KANYAKUMARI DISTRICT

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Cross and homologous mating experiments between *Aedes aegypti* (Linn.) and *Aedes albopictus* (Skuse) were conducted in the laboratory. The experiment consisted of mating between *Ae. aegypti* female x *Ae. albopictus* male and a reciprocal cross of *Ae. albopictus* female x *Ae. aegypti* male and mating of homologous *Ae. aegypti* female x *Ae. aegypti* male, *Ae. albopictus* female x *Ae. albopictus* male. This study demonstrates that there is reproductive isolation only between *Ae. albopictus* female x *Ae. aegypti* male. Insemination occurred in cross- mating experiments between *Ae. aegypti* females and *Ae. albopictus* males and produced viable eggs. The morphology of the resultant hybrid is that of female parent, *Ae. aegypti*. From F4 to F6 generation, hybrid takes about 28 -40 days to complete life stages from one generation first adult to next generation first adult. But F1 - F3 generation takes average of 17 - 18 days. Further, F7 generation takes 18 days only. The cross mating between *Ae. albopictus* female x *Ae. aegypti* male resulted in some eggs that did not hatch. Mating of homologous *Ae. aegypti* female x *Ae. aegypti* male and *Ae. albopictus* female x *Ae. albopictus* male produced viable eggs.
INFLUENCE OF ENVIRONMENTAL FACTORS ON THE POPULATION DYNAMICS OF Aedes albopictus IN A SELECTED RUBBER PLANTATION OF KANYAKUMARI DISTRICT

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Rubber plantations are one of the important grounds for the proliferation of mosquitoes especially Aedes albopictus, the main vector for Dengue and Chickungunya in the southern peninsular region. They provide favourable environmental conditions such as sufficient rainfall, enough shade, relative humidity, temperature, moderate light and canopy dense vegetation and serve as an ideal breeding ground for mosquitoes. Aedes albopictus breeds in the coconut shells used to collect latex while tapping. The population of larval instars of Aedes albopictus was studied in a selected rubber plantation in Kanyakumari district, Tamil Nadu for a period of twelve months. The abiotic factors like rainfall, temperature, humidity and light intensity were studied and positive correlation between the abiotic factors and the larval instars were observed. The larval density was found to be directly proportional to rainfall during north east monsoon during which there is total suspension of latex tapping and high breeding of mosquitoes.

Key words:- Aedes albopictus, Rubber plantations, abiotic factors
EFFECT OF TEMPERATURE ON GONOTROPHIC CYCLE OF WILD CAUGHT *ANOPHELES.CULICIFACIES*

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Weather affects the malaria incidence mostly through its effects on both the mosquito vector (species, population dynamics, gonotrophic cycle and survivorship and the development of the malaria parasite inside the mosquito vector. The transmission of malaria would largely be regulated by the survivorship and fecundity of the vector. The present study has been undertaken on the gonotrophic cycle of widely distributed *An. culicifacies*, which contributes to about 60-65% of all malaria cases of India mainly in rural and peri-urban areas throughout the country.

The laboratory study of gonotrophic cycle of wild caught *An.culicifacies* in different seasons showed, in summer season the species took even less then two days (41 hours) for completion of its gonotrophic cycle, when the average temperature was (30°±0.76). The average time taken for egg laying increased with the decrease in average temperature during rainy (54 hours) and winter season (66 hours). Linear regression test of temperature and gonotrophic cycle of *An.culicifacies* showed positive correlation (Correlation coefficient value $r^2 =0.99$, p value= 0.016). In terms of malaria transmission a reduction in the duration of the gonotrophic cycle could increase transmission of the disease, by increasing the frequency of human-vector contact. However, in the dynamics of malaria transmission other variables, besides gonotrophic cycle are sensitive to temperature changes, such as the duration of sporogonic period.

Since, now days due to the effect of global warming the average environmental temperature increases, the duration of the gonotrophic cycle and the sporogonic period may be shortened as well as vector longevity, which has great implications in disease transmission. So, actual disease estimation must be predicted. Simultaneously the biological control agents need to be linked to monitoring not only the “kill factor’ of the application but also fecundity of survivors which will help to assess the overall impact of the control agent and malaria transmission.
AN OBSERVATION ON BEHAVIOURAL CHANGES OF BREEDING HABITATS OF GENUS TOXORHYNCHITES SPECIES IN INDIA

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The tropics of both the Old and New Worlds of Toxorhynchites of some species are being found at elevations from sea level to 7000 feet or more. About 88 species have been described. Species of Toxorhynchites are largely confined to tropical regions. Only eight species were recorded in India, and one new species named Tx. tyagii yet to be published.

Toxorhynchites includes the largest mosquitoes. Adults are easily recognized by their large size and strongly bent proboscis. The body is covered with brightly coloured iridescent scales and the posterior abdominal segments have lateral scale-tufts. The larval habitats are mainly tree-holes and bamboo, but a few species are found in leaf axils, pitcher plants, and artificial containers. The larvae of all species are predacious. They feed mainly on the larvae of other mosquito species, but exhibit cannibalism in the absence of suitable prey. Males and females both feed exclusively on nectar and other sugary substances.

We conducted a survey to determine behavioral changes of breeding habitats at different locations like Gudaloor, (Nilgiri hills), Andaman and Nicobar and North West Bengal. And recorded the habitats like tin, coconut shell, mud pot, plantation leaf axil, coco shell and discarded vehicle battery. The behavioral changes due to deforestation for construction of dams, industrialization etc., the mosquitoes are disturbed and evading to the fringes of the forest. The fringe forests are associated with peripheral villages or towns where the preferential or suitable habitats. The above habitats are typical for Aedes species also breeding which are providing food for cannibalistic Toxorhynchites. Changes in breeding behavior of Toxorhynchites species might be an added benefit to the reduction of dengue fever vector density (Ae. albopictus and Ae. aegypti).
BITING ACTIVITY OF TWENTY-FOUR HOUR MAN LANDING MOSQUITO DURING DIFFERENT SEASONS AT MADURAI.

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The innate biting behaviour of mosquitoes to feed on vertebrate blood is inevitable. This physiological feature is either rhythmic or arrhythmic i.e. diurnal or nocturnal or crepuscular in its behavior species of Aedes, Anopheles, Armigeres, Culex and Mansonia exhibited temporal variation of biting behavior in the partially sub urban, rural and cultivated areas of study. The recorded biting activity of twenty six species did not overlap and inter / intra specifically. The unimodal or biomodal peak biting period exhibited by the recorded species depicted varying biting rhythms. The biting rhythmic for species of Aedes ranged from 11 to 21 hr; Anopheles from 20 to 22 hr 30 min; Armigeres subalbatus from 18 hr 43 min to 19 hr 07 min; culex from 18 hr 30 min to 24 hr 30 min and Mansonia uniformis from 19 hr 40 min to 20 hr. On the other hand species of culex quinquefasciatus and culex tritaeniorhynchus exhibited 24 hr biting behavior and their peak biting hours ranged between 23 hr 25 min – 23 hr 52 min respectively. However during different seasons the abundance of biting mosquitoes increased but the peak biting activity was more or less the same or showed negligible variation. Vector borne diseases such as Malaria, Dengue, Japanese encephalitis, filariasis and chikungunya are primarily transmitted only through the pathogen carrying mosquito bites. Henceforth the present study focuses on the in nature of biting mosquitoes which will help to control potential vector transmitting mosquito.
MOSQUITO BIODIVERSITY IN WESTERN GHATS AFTER A GAP OF 25 YEARS

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The Western Ghats hill ranges in Indian peninsula are one of the 25 hot spots in the world. Western Ghats hill ranges are rich with fauna and flora. Earlier in the past studies have shows that in India about 330 species of mosquitoes are present, which is close to 10% of the global mosquito fauna. The CRME has made much successful mosquito survey during the end of 20th century in some of the most important eco systems included Western Ghats, Eastern Ghats, mangrove forest, Assam and Andaman Nicobar Islands, and recorded a total of 234 species belonging to 33 genera and 30 sub genera which is close to 70% of the Indian mosquito fauna.

After a gap of 25 years CRME again surveyed in mosquito fauna in seven hill ranges of Western Ghats viz; Srivilliputhur, Palani, Anamalai, Sathiyamangalam, Nilgiri, foot hills of Coimbatore, and Wynad hill ranges. Altitude of the hill ranges ranged between 300m -2300m which receive rains from both southwest and northeast monsoons. Major emphasis was given to immature and adult collection of the mosquitoes. A total of 11,704 immature were collected from different habitats light tree holes bamboo stumps stream slow flowing, rocky pools, paddy fields, leaf axils etc., and a reared in the lab up to adult stages from these 7,313 specimen were examined and a total of 98 species classified under 19 genera and 20 sub genera were recorded. Among these 24 Anopheles and 74 culicines species were recorded. Three malaria vectors (An. culicifacies, An. fluviatilis, and An. stephensi) were collected in low density. In culicine, St. albopicta was dominant followed by Fr. vittatus and Oc. pseudotaeniatus. From the total collection more than 53% of mosquito species were recorded in rocky pools and tree holes.
Lutzia vorax, Lt. raptor, Orthopodomyia flavicosta, St. flavopictus, and Tx. alibipes are additional to CRME museum. Tx. alibipes was recorded first time in southern India, which was earlier recorded in western Himalayas. Tx. tyagii new species also collected and yet to be published.
MOSQUITO FAUNA SURVEY IN ANAMALAI TIGER RESERVE HILL RANGES IN WESTERN GHATS, TAMIL NADU, SOUTH INDIA.

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Anamalai Tiger Reserve which is under the Indira Gandhi National Park (IGNP). It is located in Pollachi Taluk (35 Km. distance) of Coimbatore district of Tamil Nadu at southern part of Nilgiri Biosphere which is contiguous to the Parambikulam Wild Life Sanctuary (Kerala) to its west and Parambikulam National Park to the south. The whole terrain with altitude ranges between 340m and 2510m which receive rains from both southwest and northeast monsoons with annual rain fall range between 800mm and 4500mm.

Mainly 8 localities were surveyed for mosquito fauna collection viz., (i) Karianshola (ii) Kozhikamuthi, (iii) Erumaparai, (iv) Kadamparai, (v) Tiger valley (vi) Iyyarpadi (vii) Navamalai, & (viii) Attakatti. The survey comprises mainly immature collection which was made inside the forest major breeding habitats were tree holes bamboo stumps, stream slow flowing, rocky pools etc. A total of 815 immature specimens were collected from different habitats and reared in the lab up to adult stages. Altogether 17 species were recorded belongs to 10 genera and 10 subgenera. Tree hole and bamboo stump habitats have contributed maximum number (12 species) of specimens. As far as the disease concern dengue and chikungunya vector (Stegomyia albopicta) was recorded in both larval and adult collections. It was found prevalent in all the four areas surveyed and breeding mainly in tree holes, bamboo stumps, and rocky pools.
MOSQUITO FAUNA SURVEY IN SATHYAMANGALAM HILL RANGES IN WESTERN GHATS, TAMIL NADU, SOUTH INDIA.

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Sathyamangalam hills located in southern part of Western Ghats. It is connecting place of western Ghats and Eastern Ghats and also connecting place of Tamil Nadu and Karnataka State. This forest comprises mainly tropical evergreen, semi evergreen, dry & wet deciduous forest with patches of rolling grass lands. Many parts of evergreen forest found mixed with teak plantation. The whole terrain with altitude ranges between 350 m and 1100 m which receive rains from both southwest and northeast monsoons. Sathyamangalam hills coming under Sathyamangalam taluk Erode district. It is divided in to five forest ranges viz. Sathyamangalam range, Bhavanisagar range, Thukkanayakkan palayam range, Asanoor range, and Thalavadi range.

Mainly 20 localities were surveyed minimum 5 areas each ranges. The survey comprises mainly immature collection which was made inside the forest major breeding habitats were tree holes bamboo stumps, stream slow flowing, rocky pools etc. A total of 2910 immature were collected from different habitats and reared in the lab up to adult stages. Altogether 2086 specimens belongs to 34 species were recorded to 13 genera and 14 subgenera. Lutzia (Metalutzia) raptor, Lutzia (Metalutzia) vorax and Stegomyia flavopictus are new addition of CRME Museum. Good number of Mucidus scatophagoides (24) were collected in the hill ranges.
ENTOMOLOGICAL SURVEILLANCE TO MONITOR THE JAPANESE ENCEPHALITIS VECTORS FROM THE GORAKHPUR DIVISION OF UTTAR PRADESH, INDIA.

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Japanese encephalitis is currently the most important global cause of encephalitis and so far confirmed to have caused major epidemics in India. Japanese encephalitis is a non contagious, mosquito-borne, arboviral, dreadful zoonotic disease of major public health importance in India. JE epidemics are one of the greatest threats especially to paediatric populations resulting in very high mortality in children and the survivors get permanent neurological deformities which are caused by JE virus belonging to family flaviviridae and transmitted by the principal mosquito vector Culex tritaeniorhynchus. The estimated global burden of JE was 709,000 disability-adjusted life years (DALY) lost in 2003, which is the highest among the encephalitis reported. Japanese encephalitis outbreaks occur frequently in 20 Asian countries with millions people at risk of infection. Around 35,000– 50,000 JE cases are reported every year of which 10,000–15,000 died. Approximately 50% of JE patients face severe neurological problems like brain dysfunction, mental retardness and language impairment. In the past 50 years the geographical area affected by Japanese encephalitis virus has expanded. Epidemics of JE have become regular features in the northern states of India. Especially in Eastern Uttar Pradesh where virus infected area is increasing every year. More JE cases were reported especially from the Gorakhpur division of Uttar Pradesh and the entomological investigations were conducted in the affected places of Gorakhpur division during 2011. Mosquitoes resting on vegetation and bushes around cattle sheds and pigsties were collected after dusk hours using an oral aspirator. A total of 5,055 mosquitoes collected in 60 hours comprised of eleven species of mosquitoes and four genera with the PMH density of 84.25 were collected during dusk hours. Culex vishnui subgroup mosquitoes are the
primary vectors of Japanese encephalitis, which were collected in meager numbers compared to the total mosquitoes collected. Indoor resting collection showed 7346 mosquitoes belonging to 5 species in 62 hours with the 118.48 PMH density. *Culex quinquefasciatus* was the predominant species. Few more species of mosquitoes like *An. subpictus*, *An. vagus*, *Cx. vishnui* and *Ar. subalbatus* were also collected. In the outdoor resting collection, 10 different species of mosquitoes were collected with the PMH density of 33.96. A lot of bio-ecological changes had taken place in the Gorakhpur division for more than four decades in water courses and construction of irrigation canals and small dams were modified to suit the agricultural pattern in this area brought a vast expansion of water catchment areas which supported the mosquito breeding. Longitudinal study is required to bring out the actual situation from that area to plan the effective vector control strategy.
BIODIVERSITY OF MOSQUITOES FROM MUZAFFARPUR DISTRICT, BIHAR INDIA

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Vector borne diseases are the major causes of human suffering by means of ever increasing morbidity and mortality on one hand and also stunting the intellectual and economic growth of the country as a whole. There has been a worldwide resurgence of vector borne diseases and thus the control of these diseases is very important which are generally focussed mainly on the arthropod vectors. Among the arthropod borne vectors, mosquitoes are the most remarkable group of insects of public health importance which continue to successfully transmit many diseases. Moreover there are several instances of mosquito borne viral diseases epidemic reported annually in several parts of the country. The environmental and ecological factors are held responsible which needs further investigations, with a view to control the spread of this disease. Thus it was felt necessary to record the mosquito vectors present in that area. For the proper understanding of the mosquito vector species composition from the different places where the disease had been recorded, an entomological investigation was undertaken to know the species diversity and density of the vectors prevalent in this area. Small numbers of adults were also collected from resting sites in dwellings places of cattle with the use of an aspirator and identified to species based on the available identification keys. In the Musahari, Kanti, Bochahan, Motipur and Sakra PHCs mosquitoes were sampled. Thus a total of 11 species of mosquitoes such as Cx. tritaeniorhynchus, Cx. gelidus, Cx. pseudovishnui,Cx. fuscocephala, Cx. quinquefasciatus, Mn. uniformis, An. subpictus, An. vagus, Ae. lineatopenne, Lut. fuscanus and Ar. subalbatus were collected. The principal JE vectors namely Cx. tritaeniorhynchus, and Cx. gelidus were also recorded aplenty. In addition to this the mosquitoes like An. subpictus, Ae. vittatus Cx. infula Ae. lineatopenne were also collected from the larval survey conducted in wild paddy fields, cement cisterns etc. This study revealed the existing rich mosquito biodiversity from this district of Bihar.
MOSQUITO SPECIES SUCCESSION IN SITHERI HILLS, DHARMAPURI DISTRICT

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Sitheri hill is situated at Pappireddyapatti Taluk, Dharmapuri district (Tamil Nadu) around 28 kilometer of distance from Harur town. It is located at altitude range of 3600ft. (MSL) and covered 400sq.Km in wooded area. It includes 59 hamlets one PHC (Sitheri) and 4 HSC and having 9045 Tribal peoples. The project was carried out to develop tribal health management through Remote sensing and Geographical information system (RS/GIS). An entomological investigation was carried out during the period of Dec 2009 to Feb 2012 (survey were carried out to irregular months). Species succession was observed from dusk collection, indoor resting collection, outdoor resting collection, immature survey etc. In the whole survey, 53 species and 11 genera were captured in the Sitheri hills. The major emphasis was given to find out the prevalence of vectors of JE, chikungunya, dengue, filariasis and malaria etc.

In the whole survey, the Cx. tritaeniorhynchus was dominant species (25.34%) and Aedes albopictus was the second dominant species (20.51%) followed by An. subpictus 12.72%, Cx. quinquefasciatus 10.35%, An. vagus 5.24% Cx. fuscocephala 5.23%, Ar. subalbatus 5.12%, Ae. vittatus 4.31%, An. peditaeniatus 2.45%, Cx. vishnui 2.26 other species were 6.47%.

The following species were carried out during the dusk collection, Cx. tritaeniorhynchus was 46.47% followed by An. subpictus was 16.25%, Cx. fuscocephala 9.81%, Cx. vishnui 4.31%, An. peditaeniatus 4.70%, Ar. subalbatus 3.95%, Cx. quinquefasciatus was 2.74% other species were 7.44%.
ASSESSMENT OF THE HOST SELECTION BEHAVIOR OF URBAN MOSQUITOES

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Using Immunodiffusion as a tool, studies were done on the host selection behavior of *Culex quinquefasciatus* and *Armigerous subalbatus*, which were predominant in Madurai, Tamil Nadu, India. Mosquitoes were collected from natural environment and exposed to different hosts (Rabbit, Hen and Man). The hosts were exposed to the mosquitoes during the peak time of biting activity. Blood fed mosquitoes were counted and blood smear were prepared. Then the identified host were confirmed by the Immunodiffusion method. The result showed that both *Culex quinquefasciatus* and *Armigeres subalbatus* mainly exhibited zoophilic nature during the study period. This host selection behavior helps to locate the vectors of many arthropod borne diseases. Periodic screening of host selection behavior of mosquitoes available in an area will help locate vectors which are responsible for spreading diseases.
JAPANESE ENCEPHALITIS OUTBREAK IN WEST BENGAL HAS UNUSUAL VECTORS

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During September 2011, Japanese Encephalitis (JE) cases were reported from four different districts (Jalpaiguri, Darjeeling, Dakshin Dinajpur and Cooch Behar) of West Bengal (North) from first quarter onwards. Mosquito samples collected from four districts namely Jalpaiguri, Darjeeling, Dakshin Dinajpur and Coochbehar of West Bengal were screened for Japanese encephalitis virus (JEV) by JE antigen - capture ELISA. Of 280 mosquito pools tested, one pool of Cx. pseudovishnui collected from Khudtrampalli village in Jalpaiguri district and three pools of Cx. quinquefasciatus collected one pool each from Sitarayan colony, Sushorta Nagar & Matigara Bazar were found positive for JEV. The positive pool is further confirmed as JEV by Toxo-IFA using JE specific monoclonal antibodies. In West Bengal, Cx. quinquefasciatus stood first most abundant species in indoor and outdoor collections in the present study. So far two isolates of JEV has been obtained from Cx. quinquefasciatus in India - one isolation in Bankura district, West Bengal in 1975 and another one in Kolar district, Karnataka in 1986.

It is the most common domestic species in urban, semi urban and rural areas. It is strongly anthropophilic (53.2 – 62.7%). 7-14% cattle feeding and 1.5% feeding on pigs were also observed in Cuddalore district. Two isolates of WN virus from this species have been obtained from Manjri, near Pune. Cx. quinquefasciatus has been shown to be capable of transmitting JEV and WN viruses in the laboratory.

The isolation of JEV other than Cx. tritaeniorhynchus prompted the study of the transmission potentiality of other species. The possible role of Cx. quinquefasciatus mosquitoes as a transmitter of JEV in West Bengal has been discussed.
MOSQUITO FAUNA OF MALABAR SQUIRREL SANCTUARY AT SRIVILLIPUTUR HILL RANGES IN WESTERN GHATS, TAMIL NADU, SOUTH INDIA

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A rich mosquito fauna has been reported from Western Ghats in recent time, however, mosquito fauna survey was carried out in Malabar Squirrel Sanctuary Forest area at Srivilliputur hill ranges, a part of the Western Ghats. The region is located at Virudhunagar district about 70 kms. from Madurai. The survey was conducted in five localities of four forest ranges viz., Ayanar Koil area (Rajapalayam), Senbaga area (Srivilliputtur), Kovilar dam and Pilavakkal dam (Watrap), and Sathuragiri hills (Saptur), at an altitude ranging between 150-1200 mts. The major survey include immature collection in the interior of the forest and the major breeding habitats are tree holes, rocky pools, stream slow flowing, spring pool, fallen leaf and discarded containers. A total of 1171 immature stages were collected from different habitats and reared in the laboratory up to adult stage and 589 adults were emerged. Altogether 22 species were recorded which belongs to 10 genera and 7 subgenera.

Culex (Eumelanomyia) brevipalpis was dominant (30%) species followed by Stegomyia albopicta (26%), Fredwardsius vittatus (22%), Culex (Culex) bitaeniorynchus (6%), and the remaining very few in numbers. Among Anophelines only 4 species were recorded, including malaria vector of Anopheles minimus and Anopheles varuna. Culex vishnui the vector of Japanese encephalitis (JE) and Stegomyia albopicta the vector of chikungunya and dengue fever, were also recorded. The major contributing habitats are rocky pool and tree hole (18 species). The Stegomyia albopicta was found to breed in more than 6 habitats. Adult collection was restricted to human landing collection inside the forest during day time.
Biodiversity and abundance of mosquito species are central to arboviral surveillance and control programs. In the current study, we used an extensive surveillance dataset to describe the biodiversity patterns of mosquito species throughout the diverse ecological regions of the study area. This paper represents the results of the 24-h biting activity conducted in Viraganoor which is a peri-urban area 10 km from Madurai. The area of the collection place lies on the banks of the Vaigai River and on the National highway 49 that connects Madurai with Rameshwaram lies between the latitude 9° 532 51.943 North and the longitude 78° 102 34.163 East. Stratified random sampling method was used to select the collection sites in the peri-urban areas. Therefore, three ecologically different hot spot regions (Teachers colony, Dam site, Viraganoor bus stop on NH 49) in the study area were identified based on the mosquitogenic conditions. The mosquitoes landing on human volunteers for biting were collected for every hour. Collections were made between April 2009 and April 2010. Mosquitoes landing for biting on human bait were collected for a period of 24 hours continuously in each site by using the method adopted by Pandian & Chandra Shekaran (1980) and the collected mosquitoes were identified up to species level by using the (Christophers 1933; Barraud 1934; Reinert 1973; Sirivanakarn 1976; Reuben et al. 1994; Reinert 2000, 2001; Kaur 2003) systemic identification key. The species richness index i.e. the indicator of biodiversity was calculated for each site and the indices are remarkably very high in most of the sites indicating a high degree of diversity. This revealed the occurrence of nearly 10 species belonging to 5 genera, such as Culex, Aedes, Anopheles, Mansonia and Armigeres under 2 families and under the order diptera. Culex genus represented by 5 species, Aedes, Armigeres, Mansonia and Anopheles were represented by one species in each genus. Culex quinquefasciatus was the dominant species recorded throughout the study period and Aedes aegypti, Mansonia indiana, Anopheles subpictus,
The diversity of these species mainly depends upon the animal hosts. Vector mosquitoes for Filariasis, Dengue fever and Japanese encephalitis fever were recorded in the study areas. Hence, the biodiversity studies and the factors influence the density and diversity pattern of mosquitoes in the peri-urban areas are necessary for the implementation of appropriate control strategies.
PHYSICO-CHEMICAL DETERMINANTS FOR THE GROWTH OF DENGUE AND CHIKUNGUNYA LARVAE IN CEMENT TANK – A LONGITUDINAL STUDY IN THIRUVANANTHAPURAM DISTRICT, KERALA.


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Aedes aegypti and Aedes albopictus are the two major vectors for Dengue and Chikungunya worldwide. Aedes aegypti is known as a competent vector because of its close association with humans in tropical areas and also Ae. aegypti depends highly on human blood and tends to bite and rest indoors whereas Ae. albopictus feeds on a variety of vertebrates outdoors. Ae. albopictus seems to be restricted to forest areas next to humans. Conversely, Ae. aegypti can be found in a variety of urban habitats including the highly urbanized areas without thick vegetation. A longitudinal study was carried out to monitor the physico-chemical parameters present in the different breeding sources of Ae. aegypti and Ae. albopictus throughout four different seasons in different physiographic regions of Thiruvananthapuram district, Kerala. Thiruvananthapuram district is a well known endemic area for dengue as well as Chikungunya and it maintains a strong seasonality in both the cases. Breeding habitats were enumerated mainly from the indoor and outdoor habitats like cement tank, cement cistern, mudpot, plastic container, metal container, metal drum, rubber tyre, areca nuts, coconut shells, rubber cups, flower pots and discarded containers. Based on the results, cement tank is found to be the major potential breeding sources of aedes vectors found in both the indoor and outdoor habitats for water storage because of its consistent larval production. Cement tank contained many larvae in the rainy season than in the hot summer. Over 132 cement tanks surveyed, 82% showed positive for Aedes larvae. The relationship of physico-chemical factors such as colour,
temperature, pH, alkalinity, hardness, salinity, conductivity, turbidity, total dissolved solids, ammonia, iron, sulphate, nitrate, nitrite and phosphate with the abundance of aedes immatures were analyzed. Logistic regression was used for statistical analysis using SPSS software version 15. The results were shown that there is significant difference was observed. This study found that the abundance of aedes larvae in the cement tank is significantly associated with the pH, hardness, ammonia and nitrate.
REVIEW ON SYNTHESIS AND THEIR BIOMETRICALLY
APPLICATION OF HYDRAZIDES AND HYDRAZONES LIGAND
AND THEIR TRANSITION METAL COMPLEXES

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Abstract: Coordination chemistry is the most active research area in
inorganic chemistry. Several thousands of coordination complexes have been
synthesized and investigated during the past few decades. Ever since the
importance of coordination phenomenon in biological processes was realized,
lot of metal containing macromolecules have been synthesized and studied
to understand the mechanism of complexes in biological reactions. This has
resulted in the emergence of an important branch of inorganic chemistry viz.
bioinorganic chemistry. Similarly the importance of coordination of substrate
molecules on metal ions in catalysis was understood nd a lot of research
work is being carried out on this aspect. Since structure and reactivity are
interdependent, studies on molecular structure of transition metal complexes
are of paramount importance. Excellent modern theories on metal ligand bonding
are available. These theories aid in interpreting the experimental data obtained
using sophisticated instrumental techniques available to the research workers.
LARVICIDAL ACTIVITY OF BACILLUS CEREUS AGAINST ANOPHELES STEPHENSI

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In the present investigation, a total of 25 soils were collected from different regions of the Southern Tamil Nadu. BDB1 had higher concentration of protein and was selected for bioassay. The higher protein content isolate, BDB1 subjected to 16S rRNA sequencing and was identified as Bacillus cereus (Accession No: JX276537). Bioassay was performed with B. cereus culture pellet against An. stephensi. The mortality caused by B. cereus, 3.33 – 90% at the day of post treatment. The lowest LC\textsubscript{50} and LC\textsubscript{90} (355.17 and 1.494) value of the B. cereus was observed against An. stephensi. The chi-square value was highly significant against An. stephensi (0.587).

Key words: Bacillus cereus, Anopheles stephensi, Bioassay, Percent mortality, Chi-square
DEPA CREAM – A NEW MOSQUITO REPELLENT
FROM DRDO

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Defense Research and Development Organisation, GOVT OF INDIA, has developed a new mosquito repellent CREAM AND SPRAY based on Diethyl Phenyl Acetamide (DEPA) and transferred the technology to Tagros Chemicals India Ltd., for manufacturing in large scale and supply to Indian Army and for open market to combat the mosquito menace. Following are the highlights of this product:

1. DEPA contains NO pesticides – based on pharma compound – Di Ethyl Phenyl Acetamide.
2. Repels mosquitoes causing diseases like Malaria, Dengu, Chikungunya, etc.
3. Protection up to 6 hours
4. Effective against blood sucking organisms like bed bugs, leeches, etc.
5. Environmentally safe
6. Non-poisonous
7. No irritation to eyes, skin or nose
8. Listed in Indian Pharmacopea and Registered with the Drug Controller of India
9. Added Citronella oil for pleasant smell
10. Indian Army use this is big volumes and found highly effective
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