



# 16<sup>th</sup> INTERNATIONAL CONFERENCE ON VECTOR BORNE DISEASES

21<sup>st</sup>-23<sup>rd</sup> November, 2025  
Cuttack, India

*Abstracts*



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## NAVBD PRESIDENT'S CORNER

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Vector-borne diseases are transmitted by insects such as mosquitoes, ticks, and flies that carry harmful pathogens. Common examples include malaria, visceral leishmaniasis, lymphatic filariasis, dengue, chikungunya, Zika, and Japanese encephalitis. These diseases are particularly prevalent in tropical and subtropical regions. These infections are spread from one host to another through vectors. Transmission occurs when vectors either support the growth of pathogens within their bodies or simply transport them between hosts.

Vector-borne diseases account for more than 17% of all infectious diseases worldwide. Their prevalence is strongly influenced by climate, urbanization, and human migration. As the world becomes increasingly interconnected, vectors—and the diseases they carry—are spreading to new geographic areas. Illnesses such as dengue, West Nile virus (WNV), and Eastern equine encephalitis (EEE) have recently gained global attention, underscoring the importance of public awareness and preventive action. Some of these infections are targeted for elimination by 2030, while others are prioritized for control and prevention. Effective prevention strategies include reducing vector populations, reducing man vector contact, improving sanitation and strengthening disease surveillance systems.

The Food and Agriculture Organization's (FAO) pest management initiatives emphasize promoting Integrated Pest Management (IPM) and minimizing pesticide-related risks through a comprehensive, ecosystem-based approach. Similarly, the World Health Organization (WHO) recommends Integrated Vector Management (IVM) as an effective strategy for controlling disease vectors. IVM aims to optimize the use of available resources to make vector control more efficient, affordable, environmentally sound, and sustainable.

Historically, agricultural scientists focused on pest control, while vector scientists emphasized vector control. Over time, the concepts evolved—from integrated pest management to integrated vector management. Recently, the Global Vector Control Response 2017–2030 has provided a strategic framework for enhancing global vector control through strengthened capacity building, improved surveillance systems, better intersectoral coordination, and integrated action across diseases and sectors.

In an opinion for *The Times of India* on May 26, 2023, I highlighted the acute shortage of vector scientists in India—a country that was once a global leader in this field. The lack of trained professionals has hindered effective monitoring of disease vectors and weakened outbreak prevention efforts. Many aspiring researchers also face a shortage of training and resources. To confront the growing challenges of vector-borne disease control, India must invest in training new experts, funding research in vector science, and developing robust surveillance networks to monitor disease trends and insecticide resistance.

Despite evolving terminologies and frameworks, these changes have not produced significant outcomes. It is therefore time to adopt an Outcome-Based Vector Control approach—one that combines the principles of vector management with a results-driven framework focused on measurable, quantifiable impacts rather than on routine activities. Outcome-Based Vector Control offers a strategic pathway to effectively manage the “daughters of destruction” (the vectors) and reduce the global burden of vector-borne diseases.



**Prof. Aditya Prasad Dash**



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# PLENARY LECTURES





## From Resistance to Resilience: Advancing Malaria Treatments for Elimination Goals

**Hanu Ramachandrani\***

Vice-President, Pharmaceutical Development - Medicines for Malaria Venture, Geneva

Malaria remains a significant public health challenge in India, which bears the highest malaria burden in the WHO South-East Asia Region, accounting for 1.7% (3–4 million) of global malaria cases in 2021. Despite recent progress and India's exit from the WHO High Burden to High Impact (HBHI) group in 2024, the country continues to face persistent transmission and emerging resistance to standard treatments.

Current first-line therapies for *Plasmodium falciparum* in India include artesunate plus sulfadoxine-pyrimethamine (AS+SP) in most states and artemether-lumefantrine (AL) in the Northeast, with ACTs (artemisinin-based combination therapies) as the global standard. For *P. vivax*, chloroquine (if sensitive) plus primaquine is used for radical cure. However, resistance to both artemisinin and partner drugs is rising, threatening the efficacy of existing regimens. Notably, India still widely uses AS+SP, which is discouraged by WHO due to resistance concerns, and preventive strategies like intermittent preventive treatment in pregnancy (IPTp) are not yet implemented.

To address these challenges, new antimalarial medicines are in development, focusing on novel mechanisms of action, higher barriers to resistance, and simplified dosing. Innovations include single-dose cures and long-acting chemoprevention, which aim to improve patient adherence, reduce drug pressure, and facilitate mass treatment campaigns. The pipeline includes long-acting injectables and pediatric formulations. The Zintrodiazine + Ferroquine combination is an example of a new therapy under development, showing promising safety and efficacy.

Special attention is being given to more vulnerable groups. New therapies are being designed for use in infants, young children, and pregnant women, with efforts to optimize dosing, safety, and accessibility, aiming to close treatment gaps and support malaria elimination goals by 2030.

## Plenary Lecture

**Optimising Vector Control for Elimination of Malaria and Dengue****Rajpal S. Yadav\***

Academy of Public Health Entomology, Udaipur, Rajasthan Society for Vector Ecology (Indian Region), Goa Saveetha Institute of Medical & Technological Sciences, Chennai (former Sr. Scientist, WHO, Geneva, Switzerland)

Vector control is essential for malaria elimination and dengue control, but its effectiveness is threatened by rapid urbanisation, expanding transmission, and growing insecticide resistance. To overcome these challenges, strategies must evolve from uniform interventions to micro-stratified, data-driven approaches. Integrated Vector Management (IVM) offers the framework for such optimisation. A key component of IVM is dynamic epidemiological micro-stratification, replacing static risk maps with time-series models that classify areas based on changing receptivity and vulnerability. These models integrate historical disease data, entomological indicators (vector density, insecticide susceptibility), environmental factors (water storage, rainfall, urban ecology), and human mobility. This enables early hotspot identification and precise intervention timing. Vector control should be guided by surveillance-based triggers rather than fixed schedules. For malaria, interventions include insecticide-treated nets (ITNs), dual-insecticide nets, indoor residual spraying (IRS), and targeted larviciding based on local vector ecology and resistance profiles. For dengue, strategies should focus on pupal surveys, community-based source reduction, and limiting fogging to outbreak control rather than routine use. Operational optimisation includes improving access to ITNs, using advanced spraying tools like control-flow sprayers and soluble insecticide packs, and adopting selective or focal IRS to reduce costs and insecticide load. Selective IRS must consider vector resting behaviour, house type, and indoor conditions. A critical operational question remains whether achieving 80–85% surface coverage in targeted areas can match the impact of full IRS, especially for *Aedes aegypti* in dense urban zones. Exit criteria for IRS should be defined for malaria elimination phases. With rising insecticide resistance and the discontinuation of DDT, new vector control tools are urgently needed—such as Wolbachia, sterile insect technique, spatial repellents, lethal ovitraps, indigenous traps, and mechanical aspirators. India must update evaluation and registration policies to accelerate product approval. Digital and genomic innovations—including mobile-based surveillance, drone-assisted AI habitat mapping, genomic sequencing for resistance tracking, and AI-driven outbreak prediction—enhance early warning and community engagement. However, limited skilled manpower, weak surveillance, and poor pesticide management constrain progress. Achieving malaria elimination and sustained dengue control requires increased investment in workforce training, digital surveillance, and technology-driven decision-making.

## Advancing Subnational Malaria Elimination in India

**Roop Kumari\***

Founder and Managing Director, CARAH Women Foundation

India has made significant progress toward malaria elimination, achieving a 78.1% reduction in cases and a 77.6% decline in deaths between 2015 and 2024. In line with the National Framework for Malaria Elimination (2016–2030) and the National Strategic Plan (NSP) 2023–2027, the Government of India has developed a structured approach to verify malaria elimination at subnational levels. The *Guidelines on Subnational Malaria Elimination Verification in India align with WHO guidelines*, provide standardized procedures for assessing, validating, and recognizing districts that sustain zero indigenous malaria cases for three consecutive years.

The verification process, led by the National Centre for Vector Borne Disease Control (NCVBDC), Ministry of Health and Family Welfare, includes district-level self-assessment, submission of a Subnational Elimination Report and dossier, independent review by the Subnational Malaria Elimination Advisory Committee (SMEAC), and field validation by an Evaluation Committee with technical support from WHO. The framework emphasizes evidence-based surveillance, sustainability of vector control measures, case investigation, and mechanisms to prevent re-establishment of transmission.

According to the NCVBDC 2023 report, 94 districts reported zero indigenous cases for three consecutive years (2021–2023), while 603 districts recorded an API below 1. In 2024, 160 districts reported zero cases, reflecting sustained progress toward elimination as per annual report of the program. Thus, several districts have maintained malaria-free status, while many others are transitioning from intensified control to elimination and prevention phases.

As per NCVBDC report of 2023, 94 districts have reported zero indigenous cases for three consecutive years (2021–2023), with 603 districts recording an API below 1. However, in 2024, 160 districts reported zero cases as per annual report of the program. Thus, several districts, have maintained malaria-free status, while many others are transitioning from intensified control to elimination or prevention phases.

Subnational malaria elimination verification represents a milestone-based approach fostering accountability, competition, and sustained vigilance across states and districts. The institutionalization of SMEAC and Evaluation Committee at National level for systematic verification marks a significant step toward India's national malaria elimination by 2030. Continued investments in surveillance through IHIP, enhanced entomological monitoring, early diagnosis and complete treatment and improved multi-sectoral coordination and prevention of re-establishment are critical to sustain these gains.

## Malaria Free India is Within our Reach

**Altaf A. Lal\***

Foundation for Disease Elimination and Control, India

The Malaria Elimination Demonstration Project (MEDP), implemented in Mandla district, Madhya Pradesh, through a Public–Private Partnership between the Indian Council of Medical Research (ICMR), the Government of Madhya Pradesh, and the Foundation for Disease Elimination and Control in India (FDEC India), demonstrated the feasibility and economic value of malaria elimination in high-burden, tribal-dominated areas. Covering a population of 1.2 million, MEDP adopted a comprehensive approach encompassing robust surveillance and case management, integrated vector management, community engagement, and capacity building, supplemented by innovations in monitoring, evaluation, and data reporting. With an additional investment of INR 180 million over four years, indigenous malaria was eliminated within the district, and the gains were sustained for three subsequent years.

The study employed a difference-in-difference regression model, comparing Mandla with four control districts, to estimate epidemiological outcomes. A cost-of-illness framework was also used to quantify economic benefits. In the absence of MEDP, Mandla was projected to record nearly 1,000 malaria cases annually (2017–2023). Instead, cases fell to near zero, at an additional cost of only INR 37 per capita per year. The intervention led to significant reductions in household treatment costs, productivity losses, and value of lives lost, while avoiding substantial expenditures on personal protection and enabling health system savings.

Annual economic savings attributable to MEDP were estimated at INR 73.6 million (approximately USD 0.89 million), compared to an annualised investment of INR 45 million. The resulting cost–benefit ratio was 1.63, indicating that every rupee invested yielded a return of INR 1.63 in economic benefits. The annual return on investment (ROI) was approximately 63%, corroborated by probabilistic sensitivity analysis. Beyond quantifiable benefits, MEDP contributed to improved tourism growth, reduced fear of disease, and strengthened local health systems.

These findings highlight MEDP as a scalable model for malaria elimination, demonstrating that targeted investments not only achieve public health gains but also deliver substantial long-term economic and societal returns.

## Quantifying the Investment Gap and Cost of Malaria Elimination in India: A Macroeconomic Analysis

**Mrigendra P Singh\***

Foundation for Disease Elimination and Control, India

Malaria remains a significant public health challenge in India, accounting for over half of cases and deaths in the WHO South-East Asia Region despite substantial progress in recent decades. The Government of India has committed to achieving zero indigenous malaria cases by 2027 and complete elimination by 2030, as outlined in the National Strategic Plan (NSP) 2023–2027. This study quantifies the economic burden of malaria, estimates the investment required for elimination, and assesses the potential returns on investment using a macroeconomic modelling approach.

A district-level SEIR model stratified by endemicity was applied to estimate intervention costs under three scenarios: (1) business-as-usual; (2) scale-up to NSP targets; and (3) intensified interventions modelled on the Mandla Malaria Elimination Demonstration Project (MEDP). The cost-of-illness method was employed to estimate the national economic burden, encompassing treatment expenses, productivity losses, premature deaths, and personal protection expenditures. Forecasting models, including Holt's multiplicative method, were used to project malaria incidence through 2030, while Disability-Adjusted Life Years (DALYs) and return on investment (ROI) were calculated to evaluate health and economic outcomes.

Results indicate that the current investment, averaging only INR 6.0 per capita annually, is insufficient to achieve elimination. The optimum annual investment required was estimated at INR 16,080 million (about INR 25 per capita), compared to the currently allocated INR 3,837 million. Under the business-as-usual scenario, malaria cases and deaths would persist beyond 2030, with an economic burden nearly four times higher than current investment. Scaling up to NSP targets (Scenario 2) could reduce cases to 20,637 and deaths to 23 by 2030, lowering DALYs from 4,881.8 in 2023 to 1,130.8 in 2030, with an ROI of 1.78. The intensified MEDP-based model (Scenario 3) projected near elimination, with only 1,720 cases and zero deaths by 2030, and a higher ROI of 3.84.

These findings underscore that bridging India's malaria investment gap is both a health imperative and an economic priority, yielding substantial returns through reduced disease burden, productivity gains, and sustainable development.

## Elimination of Malaria will Require Combination Vaccines Targeting Multiple Parasite Stages in Multiple *Plasmodium* Species

**Nirbhay Kumar\***

Department of Global Health, Milken Institute School of Public Health, George Washington University, Washington DC. (nkumar@gwu.edu)

Malaria remains a major public health problem with an estimated ¼ billion clinical cases and mortality of >600,000 in 2023. More than 90% of the global malaria burden is due to infection by *Plasmodium falciparum* (Pf) and *P. vivax* (Pv), often co-endemic in Asia, Pacific, Central and South America, with growing evidence in parts of Africa. The *P. falciparum* malaria vaccine RTS,S/AS01, approved by the WHO in 2021 and R21/Matrix-M approved in 2023 are significant steps toward malaria control. However, both vaccines exhibit partially protective efficacy of short duration, and emphasize the need for additional tools such as more effective and improved vaccines to interrupt malaria transmission. It is widely recognized that combination vaccines targeting antigenically distinct life cycle stages (sporozoites, erythrocytic asexual parasites and male and female gametocytes) in *P. falciparum* and *P. vivax* will provide effective immune protection against infection and reduce transmission. The primary focus of the research (in progress) in my lab is to develop combination vaccines targeting infection by sporozoites and mosquito transmission by gametocytes of *P. falciparum* and *P. vivax*. *P. falciparum* circumsporozoite protein (PfCSP) is the leading pre-erythrocytic vaccine (PEV) candidate. In addition to Pfs25, pre-clinical studies and limited phase 1 trials have identified Pfs230 as another strong candidate for the development of a transmission-blocking vaccine (TBV). Orthologous antigens (PvCSP and Pvs25) in *P. vivax* have also been identified as strong PEV and TBV candidates. We are employing recombinant protein adjuvant formulations, DNA plasmids and mRNA-LNP vaccine platforms for the development of such combination vaccines. The mRNA-LNP approach shown to be relatively safe and effective offers the flexibility to combine several antigens in a multivalent vaccine, and we have recently published studies on effective immunogenicity of mRNA-LNP vaccines encoding PfCSP and Pfs25. We are currently evaluating protective efficacy of Pfs25 and Pfs230 TBV antigens, individually or in combination with PfCSP. Similarly, studies are evaluating protective immunogenicity of Pvs25 and chPvCSP (a chimeric *P. vivax* CSP representing all three allelic types) combination. Our ultimate goal is to develop and evaluate a combination of TBVs and PEVs of both *Plasmodium* species in mice and nonhuman primates. These studies will establish that unique combinations of mRNA-LNP vaccines can be developed to target both infection and transmission of malaria. Vaccine combinations targeting two major species, *P. falciparum* and *P. vivax*, and validated in animal models (mice and NHPs), will serve as a guide for future clinical trials and play a significant role in reducing malaria burden and to achieve the long-term goal of global malaria elimination.

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## Cerebral Manifestations of Falciparum Malaria in Adults: More Than Meets the Eye

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Recent advances in quantitative neuroimaging have substantially advanced our understanding of the pathophysiological mechanisms underlying cerebral malaria (CM) in adults. Magnetic resonance imaging (MRI) has revealed characteristic patterns of brain alteration that closely correlate with disease severity. Importantly, similar—though subtler—changes have also been detected in non-cerebral and uncomplicated malaria, supporting the concept of a continuum of cerebral involvement across the clinical spectrum. These findings challenge traditional distinctions between “cerebral” and “non-cerebral” malaria and highlight shared mechanisms of brain injury, potentially influenced by dysfunction in distant organs such as the liver and kidneys. This talk will present an up-to-date synthesis of neuroimaging findings across malaria phenotypes and emphasise the urgent need for longitudinal studies integrating advanced imaging with targeted neurocognitive assessments to define the lasting neurological impact of malaria, identify those at greatest risk, and guide interventions to preserve brain health after infection.

## Tiny Vectors, Big Impact: Emerging Mite and Tick-Borne Diseases in India's Changing Landscape

**Tilak Rina\***, Scientist 'G' (Retd)

Department of Community Medicine, AFMC, Pune, India

The tiny yet mighty vectors – the mites and the ticks are decidedly impacting lives like never before! The diseases transmitted by these vectors are rapidly emerging as significant public health challenges in India, driven by the ecological transformation, climate variability, and increased human-animal interactions. Scrub typhus, transmitted by trombiculid mites, has re-emerged as a leading cause of acute febrile illness across diverse regions, often presenting with non-specific symptoms that challenge accurate diagnosis and prompt treatment. Tick-borne diseases like Kyasanur Forest Disease (KFD), Crimean-Congo Hemorrhagic Fever (CCHF), and Indian Tick Typhus are disconcertingly expanding their geographic and seasonal range courtesy of the adaptability of vector species and the changing environmental conditions. Lyme disease, yet another tick-borne disease, though traditionally underreported in India, is gaining prominence due to the various recent serosurveys indicating possible wider than anticipated endemicity.

The disquieting emergence of a plethora of mite and tick-borne diseases over the last decade has been greatly influenced by the complex eco-epidemiology of these diseases, coupled with factors such as vector competence, genetic diversity, and lack of awareness, which confound the control efforts. The lack of sensitive diagnostics, underreporting, and limited surveillance infrastructure further exacerbates the burden, often resulting in delayed detection and response. The disease dynamics are inopportunately challenging due to the tiny size of the vector mites and ticks.

It is thus important that a multidisciplinary approach involving integrated vector surveillance, improved access to diagnostics, research on vector biology and control, and initiatives to increase community awareness be adopted at the earliest. It is needless to emphasise that failure to address these issues may result in public health challenges, including increased rates of illness, death, and outbreaks that affect both populations and healthcare workers. Implementing the tenets of the One Health approach is even more crucial in mitigating the health impact of these rapidly emerging zoonotic diseases. Additionally, adoption of strategies such as predictive modelling and targeted interventions can aid in effective management of these rapidly emerging diseases, thereby facilitating India's preparedness for future outbreaks.

## Elimination of Lymphatic Filariasis in India: Goal & Challenges

**P. K. Srivastava\***

Director Absolute Human Care Foundation-A Non-profit Organization, New Delhi.

Former Joint Director, National Vector Borne Disease Control Programme, Government of India, Delhi – 110054

Lymphatic filariasis (LF) is a neglected tropical disease caused by *Wuchereria bancrofti*, *Brugia malayi*, and *B. timori*, transmitted by mosquitoes. It leads to chronic conditions such as lymphedema and hydrocele, causing social stigma, disability, and economic hardship. Following the WHO resolution WHA50.29 (1997), the Global Programme to Eliminate Lymphatic Filariasis (GPELF) was launched in 2000, with two main strategies: (1) interrupting transmission through mass drug administration (MDA) and (2) morbidity management and disability prevention (MMDP). In the WHO South-East Asia Region, nine of eleven countries were endemic, with India accounting for about 40% of the regional burden. LF is endemic in 21 Indian states/UTs, with 99.4% of infections due to *W. bancrofti* transmitted by *Culex quinquefasciatus*. *B. malayi* infections occur in limited areas of Kerala and sporadically in Odisha. India's LF control efforts date back to 1949, when a pilot project in Odisha showed that diethylcarbamazine (DEC) and indoor residual spraying could reduce transmission. The National Filaria Control Programme (NFCP) launched in 1955, initially emphasized selective treatment and anti-larval measures due to poor mass coverage. A major shift in the 1990s, guided by WHO, introduced annual MDA after successful ICMR and NCDC pilots. Nationwide MDA began in 2004 using DEC, later combined with Albendazole (DEC+ALB).

The programme expanded from 202 to 345 districts, and in 2018, the Accelerated Plan for Elimination of LF (APELF) introduced triple-drug therapy (Ivermectin + DEC + Albendazole, IDA) to hasten interruption, now implemented in 76 districts across 10 states. Covering over 600 million people, it is the world's largest LF elimination effort. Challenges include logistics, drug compliance, diagnostics, and workforce limitations. Innovations such as strip-packed DEC, flexible timelines, and independent coverage assessments improved performance, while Transmission Assessment Surveys (TAS) guide MDA cessation. However, some failures underscore the need for supervised intake and better tools. MMDP focuses on care for all lymphedema and hydrocele patients, though home-based care and surgeries remain limited. Medical colleges can strengthen diagnosis, care, and research. Sustaining financial and human resources is vital as India strives toward the 2021–2030 NTD Roadmap goals—sustainable MDA cessation, post-validation surveillance, and universal patient care.



# **SPECIAL SESSIONS**



## Dengue in High Risk Patient: Challenges and Insights

**Ashutosh Biswas\***

Executive Director & CEO, AIIMS, Bhubaneswar

Dengue is a rapidly spreading arboviral disease posing serious morbidity and mortality risks, particularly among vulnerable groups such as the elderly, pregnant women, children, and individuals with chronic illnesses like diabetes, hypertension, cardiovascular disease, or chronic kidney disease (CKD). While most infections are mild, these high-risk groups are prone to severe complications, making timely diagnosis and management challenging. In elderly patients, dengue often presents atypically, with absent or subtle symptoms like retro-orbital pain or arthralgia, leading to delayed diagnosis. Age-related comorbidities, weakened immunity, and the use of multiple medications increase risks of severe dengue, shock, multiorgan failure, and hospital-acquired infections, prolonging recovery. Patients with cardiovascular disease are at high risk due to anticoagulant or antiplatelet therapy. Dengue-associated thrombocytopenia increases bleeding risk, requiring careful balancing of antithrombotic treatment. Fluid therapy must be cautiously managed to prevent fluid overload or pulmonary edema.

Pregnant women face severe complications such as hemorrhage, shock, and preeclampsia-like syndromes, along with risks to fetal health like miscarriage or premature delivery. Physiological changes in pregnancy complicate both diagnosis and management. In hypertensive patients, inappropriate blood pressure correction can worsen shock, while diabetics are prone to uncontrolled hyperglycemia or ketoacidosis, requiring close glycemic monitoring and treatment adjustment. Children, particularly infants, are highly susceptible to shock and plasma leakage due to smaller blood volume and immature immunity. CKD patients pose major management challenges because of limited fluid tolerance. Overhydration can cause pulmonary edema, while under-resuscitation increases shock risk. Dialysis-dependent patients need individualized drug and electrolyte management. Co-infections with malaria, chikungunya, HIV, or tuberculosis further complicate diagnosis and treatment due to overlapping symptoms and drug interactions.

Managing dengue in high-risk groups requires a multidisciplinary approach involving infectious disease specialists, intensivists, obstetricians, nephrologists, cardiologists, and pediatricians. Early recognition, careful fluid resuscitation, individualized drug therapy, and close organ monitoring are essential. Developing dedicated clinical guidelines, enhancing diagnostic tools, and integrating comorbidity-specific protocols are critical to reducing dengue-related mortality and improving outcomes among these vulnerable populations.

## Dynamics of Immune response: Mild and severe dengue

**Neelika Malavige\***

Professor, Department of Immunology and Molecular Medicine  
University of Sri Jayewardenpura

Dengue is the fastest-emerging climate-sensitive infection, with escalating morbidity, mortality, and health-system strain. Despite its growing global burden, there are no specific treatments or reliable prognostic markers to identify patients at risk of severe disease. Most infections are mild, but some progress to vascular leakage, shock, and organ dysfunction. Endothelial permeability typically increases between days 3–6 of illness, driven by inflammatory mediators, and is transient yet potentially fatal if undetected. Early recognition of leakage and individualized fluid replacement can prevent shock, bleeding, and multiorgan failure, while timely cessation of fluids prevents overload. Severe liver dysfunction and bleeding may also occur without leakage, often related to immune dysregulation such as secondary hemophagocytic lymphohistiocytosis (HLH); in women, severe per vaginal bleeding is a common complication.

Rapid urbanization and climate change continue to fuel dengue epidemics, yet understanding of its complex pathogenesis remains limited, constraining the development of vaccines, antivirals, and biomarkers. Although antivirals are promising given dengue virus's direct pathogenic effects, clinical trials have not met primary endpoints due to challenges in assessing viral kinetics and intracellular infection. In addition to antivirals, drugs targeting inflammatory lipid mediators (e.g., rupatadine, montelukast, ketotifen) and immunomodulators show variable outcomes but warrant evaluation in combination therapy. Given rising incidence and complications, research priorities should focus on elucidating viral and host mechanisms, developing effective therapeutics, vaccines, and biomarkers, and moving beyond reliance on vector control and fluid management to reduce the global dengue burden.



## Dengue Alliance strategy for treatment development

**Shikha Malik\***

Senior Manager Scientific Affairs, Drugs for Neglected Diseases initiative (DNDi)

Dengue, a climate-sensitive neglected tropical disease, is one of the top ten threats to global public health worldwide. It is the fastest growing mosquito-borne viral disease with an estimated 390 million infections each year in more than 100 countries. More than 70% of the disease burden is estimated to be in Asia, with rapidly growing incidence in other parts of the world – particularly the Americas.

Despite its prevalence and severity, there is still no specific treatment available for dengue. Medicines that could prevent mild cases from becoming severe would therefore play an important role in reducing mortality and prevent hospitals from being overwhelmed during outbreaks.

In 2022, The Dengue Alliance –a global research and development (R&D) partnership was established to urgently address the major gap in dengue control: the lack of a specific treatment. The alliance is led by institutions from dengue-endemic countries—including Brazil, India, Malaysia, and Thailand and is designed as a collaborative, needs-driven effort. Its primary mission is to accelerate the discovery and delivery of affordable and accessible dengue therapeutics through partnerships with institutions in the most affected countries. Drugs for Neglected Diseases initiative (DNDi), a not-for-profit research and development (R&D) organization, serves as the secretariat of the Alliance, coordinating efforts across regions and partners. The Dengue Alliance is advancing urgently needed pre-clinical research, testing the efficacy of repurposed drug candidates, and implementing clinical trials of the most promising potential dengue treatments. Alliance members also coordinate efforts to help overcome knowledge gaps, expedite clinical research and regulatory review, address unmet needs in diagnostics, mobilize resources, and share research knowledge openly.

## Confronting the Public Health Challenge of Dengue: Integrated Approaches for Surveillance and Control in India

**Kalpana Baruah\***

Senior Consultant and Former Additional Director & Scientist 'V'

National Center for Vector Borne Diseases Control, Delhi

Dengue fever, caused by the dengue virus (DENV) and transmitted mainly by *Aedes aegypti* and *Aedes albopictus*, is a major vector-borne public health threat, particularly in tropical and subtropical regions. Its increasing geographic spread, outbreak frequency, and intensity are driven by rapid urbanisation, climate change, global travel, and weak vector control infrastructure. Addressing these challenges requires integrated surveillance and a holistic, evidence-based approach combining entomological, epidemiological, and community-driven interventions to reduce disease burden and strengthen health system resilience. In India, dengue prevention and control are coordinated by the National Centre for Vector Borne Diseases Control (NCVBDC) under the Ministry of Health and Family Welfare. The Centre provides technical and financial support to states for implementing national strategies. Laboratory-based surveillance offers early warning of transmission and free diagnosis. As no antiviral drug exists, preventing vector breeding through source reduction remains the key strategy. NCVBDC provides guidance to promote sustainable, locally adapted, community-based vector control. Vector control remains central to dengue prevention but faces diminishing returns due to operational and environmental challenges. To strengthen surveillance, entomological zones have expanded from 72 to 114, improving geographic coverage. To address the shortage of trained entomologists, NCVBDC has conducted targeted training to enhance capacity in surveillance, data analysis, and decision-making. Community engagement is vital for promoting behavioural change and ensuring compliance with control measures. Alongside conventional methods like larval source management and fogging, research on innovative, sustainable vector control tools is needed. Real-time digital health platforms are being introduced for early outbreak detection through integrated human and vector data monitoring. Given dengue's multisectoral nature, collaboration among ministries, state governments, private sectors, and NGOs is crucial. The complexity and adaptability of dengue transmission demand a comprehensive response built on strengthened surveillance, diversified vector control, and active community participation for effective and sustainable dengue control.

## The OPTIC Study: Key Results and Insights

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Glucose-6-phosphate dehydrogenase (G6PD) testing plays a critical role in safeguarding patients who require radical cure treatment for *Plasmodium vivax* malaria. The 8-aminoquinoline class of antimalarial drugs, including primaquine and the newer single-dose tafenoquine, are essential to eliminate dormant liver-stage hypnozoites and prevent relapse. However, both drugs are contraindicated in individuals with G6PD deficiency, as they can trigger severe drug-induced hemolysis. Until recently, the lack of an affordable, field-ready quantitative G6PD test limited safe implementation of radical cure therapy in routine malaria programmes. With the introduction of reliable point-of-care G6PD diagnostic devices, it has become necessary to determine whether these tests can be effectively integrated into India's national health system. The OPTIC study (Operational Feasibility of G6PD Testing in India for Radical Cure) was therefore designed to assess the real-world feasibility, acceptability, and operational readiness for large-scale G6PD testing in malaria-endemic regions of the country.

The study was conducted across four diverse geographic and epidemiological settings, covering both urban locations (Surat in Gujarat and Kolkata in West Bengal) and rural tribal-dominant areas (Kandhamal in Odisha and Jagdalpur in Chhattisgarh). A systematic random sampling approach was used to ensure representation of different health system levels. A total of 64 healthcare facilities were selected per study site, resulting in 256 facilities overall. At each facility, approximately 20 febrile patients were consecutively tested using the Standard G6PD Biosensor (SD Biosensor), a quantitative point-of-care device capable of measuring G6PD enzyme activity within minutes. Across all sites, more than 5000 patients presenting with fever were tested, generating one of the largest operational datasets on G6PD screening in India to date. The study provides crucial evidence on whether routine G6PD testing can be scaled up nationwide to support safe deployment of radical cure treatment and strengthen India's malaria elimination strategy.

- HCP proficiency in performing G6PD testing using a standardized assessment, ensuring accurate sample handling, device operation, result recording, and quality control.
- Appropriate interpretation of the G6PD test result, followed by the ability to link enzyme activity levels with correct treatment decisions, ensuring safe and effective radical cure therapy.

The results of the study will be presented and clearly indicate that G6PD testing using the SD Biosensor is not only feasible but also operationally sustainable across a wide range of Indian healthcare settings, including both urban and rural facilities. These findings highlight the strong potential for nationwide adoption of point-of-care G6PD testing to safely enable shorter radical cure regimens. Such scale-up would enhance patient access to correct treatment, improve adherence, reduce *Plasmodium vivax* relapse rates, and significantly accelerate India's progress toward malaria elimination.

## Notional Treatment Selection

**Dutta A<sup>1\*</sup>, Naser SM<sup>2\*</sup>**, Bharti PK<sup>3</sup>, Aditya Arya<sup>3</sup>, Singh K<sup>3</sup>, Ranjha R<sup>3</sup>, Singh PK<sup>3</sup>, Baharia RK<sup>4</sup>, Singh SP<sup>5</sup>, Sinha DP<sup>5</sup>, Nida<sup>3</sup>, Manjhi N<sup>5</sup>, Vadhel A<sup>4</sup>, Roy M<sup>2</sup>, Chakraborty A<sup>6</sup>, Choudhury SR<sup>6</sup>, Nag N<sup>7</sup>, Mandavi PL<sup>7</sup>, Tekam SS<sup>8</sup>, Maitry RC<sup>8</sup>, Basak S<sup>7</sup>, Patel D<sup>9</sup>, Umrigar PH<sup>9</sup>, Sailor P<sup>10</sup>, Patel A<sup>11</sup>, Mohanty S<sup>12</sup>, Sahu P<sup>12</sup>, Ghosal J<sup>1</sup>, Rath J<sup>1</sup>, Grewal Daumerie P<sup>13</sup>, Valecha N<sup>13</sup> and Anup Anvikar<sup>1</sup>

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We aimed to assess the feasibility of applying a G6PD-based treatment algorithm in different tiers of public health system of India by examining the appropriateness of notional prescription for radical treatment of *P.vivax* based on G6PD test results.

Sixty-four health facilities selected from each of the four sites, two rural (Jagdalpur and Kandhamal) and two urban (Kolkata and Surat) were categorized into higher and lower-level facilities based on presence / absence of a functional lab technician. Fever cases attending those facilities (n=256) underwent G6PD testing by trained designated staff to be followed by notional prescription of appropriate radical treatment for *P. vivax*: *single dose tafenoquine/7 days primaquine* for G6PD normal, *14 days primaquine* for G6PD intermediate and *no radical treatment* for G6PD deficient subjects.

Appropriateness of notional treatment was high in both urban and rural settings. Detailed results will be presented.

## G6PD Training : Approach, Experience and Lessons Learnt

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Inadequate compliance with radical cure for *Plasmodium vivax* malaria highlights the urgent need for improved treatment strategies. The use of shorter-course regimens can enhance adherence; however, these are contraindicated in individuals with glucose-6-phosphate dehydrogenase (G6PD) deficiency—an X-linked enzymatic disorder that increases the risk of hemolysis upon exposure to specific antimalarial drugs such as primaquine or tafenoquine. Therefore, routine G6PD testing is essential before administering radical cure to ensure patient safety and effective malaria elimination. A key component of the OPTIC study involved comprehensive training of healthcare workers (HCWs) across diverse healthcare settings to facilitate safe implementation of G6PD testing and treatment. The study included both urban and rural sites representing multiple levels of India's healthcare system. The structured curriculum combined theory and practice of G6PD testing, G6PD biology, its clinical implications, and its relevance to antimalarial policy. Ethical guidelines, safety precautions, and documentation procedures for informed consent and case records were also included. Pre-training theoretical assessments and post-training practical competency evaluations were

conducted, with the latter serving as the study's operational baseline. Comparative analysis after approximately one month demonstrated marked improvement in participants' knowledge and proficiency in performing POC G6PD testing. Supplementary visual job aids, such as infographics and pictorial guides, further reinforced learning outcomes.

The training program significantly improved healthcare workers' (HCWs) technical competence, confidence, and preparedness for implementing glucose-6-phosphate dehydrogenase (G6PD) testing across diverse healthcare settings. Through hands-on demonstrations, interactive sessions, and practical exercises, participants gained a clear understanding of test procedures, result interpretation, and appropriate patient management. The initiative also enhanced awareness of the importance of G6PD testing in guiding radical cure treatment for *Plasmodium vivax* malaria, thereby reducing the risk of hemolytic complications and improving patient safety.

Importantly, the training approach was dynamic and responsive—materials and methods were refined based on feedback from initial sessions to better address local needs, language preferences, and varying levels of technical expertise. This adaptability ensured greater engagement and effectiveness, particularly in lower-level health facilities with limited laboratory infrastructure. The success of the program demonstrates the feasibility, sustainability, and scalability of incorporating G6PD testing into routine malaria case management within India's public health system.

Overall, these findings provide valuable insights for national malaria elimination efforts. By institutionalizing standardized training and supportive supervision, G6PD testing can be seamlessly integrated into existing diagnostic and treatment protocols, ensuring safer, evidence-based radical cure practices that accelerate progress toward malaria elimination in India.

## Acceptability, and Experience with G6PD Testing: Insights from Qualitative Research

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Point-of-care (POC) Test of glucose-6-phosphate dehydrogenase (G6PD) holds significant promise for improving the safety of malaria treatment. It is especially crucial for individuals with G6PD deficiency, who are at risk of haemolysis when treated with certain antimalarial drugs. However, a major challenge lies in training healthcare workers (HCWs) across varied healthcare settings in India to use these appropriately. The study aimed to explore HCWs' perceptions and challenges regarding G6PD training and experience with G6PD testing. The study was conducted in rural (Jagdalpur, Kandhamal) and urban (Surat, Kolkata) regions across various levels of healthcare, including sub-centres, PHCs, and CHCs.

Data was collected through In-Depth Interviews (IDIs), Focused Group-discussions (FGDs), and Key-informant interview (KIIs) with doctors, lab technicians, and community health officers. Thematic analysis was performed using NVivo software, employing a deductive approach to identify key themes. Insights were captured through reflective memos, enriching the understanding of HCWs' experience and challenges in implementing G6PD testing.

G6PD testing was positively received by HCWs, with improvements in knowledge and competence post-training. HCW experience, challenges face and recommendations will be presented. The study highlights healthcare workers' ability to perform G6PD testing in routine practice after overcoming minor challenges.



## G6PD Prevalence: Variations Across the Study Sites

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Glucose-6-phosphate dehydrogenase (G6PD) deficiency is an X-linked recessive enzymatic disorder affecting red blood cell metabolism. Globally, an estimated 400 million individuals (1–8% of the population) are affected, with varying prevalence across regions. As 8-aminoquinoline-based drugs used for the radical cure of *P. vivax* malaria are contraindicated in G6PD-deficient individuals due to risk of haemolysis, reliable point-of-care testing becomes essential for providing safe treatment. The OPTIC study evaluated the feasibility and prevalence of G6PD deficiency across 64 health facilities spanning four geographically diverse sites—Jagdalpur, Kandhamal, Surat, and Kolkata. More than 5000 individuals were tested for G6PD enzyme activity using standardized quantitative methods.

Comprehensive prevalence data will be presented. The results highlight the variable prevalence of G6PD deficiency in India which is of importance beyond malaria.

## Malaria Prevalence During the Low Transmission Season

**Ranjha R<sup>1\*</sup>, Tekam SS<sup>2\*</sup>, Bharti PK<sup>1</sup>, Aditya Arya<sup>1</sup>, Singh K<sup>1</sup>, Singh PK<sup>1</sup>, Baharia RK<sup>3</sup>, Singh SP<sup>4</sup>, Sinha DP<sup>4</sup>, Nida<sup>1</sup>, Manjhi N<sup>4</sup>, Vadhel A<sup>3</sup>, Roy M<sup>3</sup>, Chakraborty A<sup>6</sup>, Naser SM<sup>4</sup>, Choudhury SR<sup>6</sup>, Nag N<sup>7</sup>, Mandavi PL<sup>7</sup>, Tekam SS<sup>2</sup>, Maitry RC<sup>2</sup>, Basak S<sup>6</sup>, Patel D<sup>8</sup>, Umrigar PH<sup>8</sup>, Sailor P<sup>9</sup>, Patel A<sup>10</sup>, Mohanty S<sup>11</sup>, Sahu P<sup>11</sup>, Ghosal J<sup>12</sup>, Rath J<sup>12</sup>, Dutta A<sup>12</sup>, Grewal Daumerie P<sup>13</sup>, Valecha N<sup>13</sup> and Anup Anvikar<sup>1</sup>**

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Malaria transmission in India is highly heterogeneous and seasonal. There are fewer malaria cases in low-transmission season, with a substantial increase after the monsoon. Understanding infection patterns during the low-transmission season is critical for identifying persistent transmission pockets and reservoirs that can sustain malaria year-round. As part of a feasibility study assessing point-of-care G6PD testing for safe radical cure of *Plasmodium vivax*, results of malaria tests were collected and analysed, across four geographically distinct study sites in India representing varying transmission intensities.

More than 5000 febrile individuals were tested for malaria infection using Rapid Diagnosis test (RDT)/Microscopy. Overall positivity for *P. vivax*, *P. falciparum* varied among the study sites but remained consistent with historical trends of endemicity in the region.

Detailed results will be presented. The persistence of both *P. falciparum* and *P. vivax* infections during the low transmission season highlights ongoing residual malaria transmission, even in periods of minimal vector activity. These findings underscore the need for continuous surveillance throughout the year and integrated interventions that combine case management with vector control. Moreover, the co-assessment of G6PD status alongside malaria screening provide additional epidemiological insights.

## Lessons Learnt for Scaling Up

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The OPTIC study generated critical evidence on the operational feasibility and readiness of India's health system to scale up glucose-6-phosphate dehydrogenase (G6PD) testing, a prerequisite for the safe administration of radical cure regimens such as primaquine. The study demonstrated that G6PD testing can be successfully implemented across different levels of healthcare facilities. Healthcare providers (HCPs) achieved the required proficiency score ( $\geq 75\%$ ), with 99% of HCPs at higher-level facilities and 91.1% at lower-level facilities reaching this benchmark. Most errors in rural sites occurred during routine procedural steps—such as checking test kit expiry dates and proper glove use—indicating the need to strengthen adherence to general clinical practices rather than test-specific skills. While the results affirm the technical feasibility of nationwide G6PD testing, a few challenges which require a nationwide training, logistics such as supply of kits are anticipated, yet easily manageable. Hence, introduction of reliable point-of-care G6PD testing holds transformative potential for India's malaria elimination program. It would allow safer deployment of shorter-course radical cure regimens, overcoming compliance issues associated with 14-day primaquine therapy and ensuring protection for G6PD-deficient individuals vulnerable to haemolysis. The OPTIC study provides a strong foundation for developing G6PD activity-based treatment algorithms and informed policy frameworks essential for the elimination of *Plasmodium vivax* malaria in India.

## Clinical manifestations and treatment outcome of severe vivax malaria in a tertiary care centre in Kolkata, West Bengal

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Malaria is a vector borne protozoan infection caused commonly by *Plasmodium falciparum* and *Plasmodium vivax* (*P. vivax*). In southeast Asia, most malaria cases were concentrated in India (66%) and about 94% of all deaths were reported from India and Indonesia. In India, 46% cases were due to *P. vivax*. Although previously believed to be less aggressive, *P. vivax* is now being recognised as causing severe and possibly fatal malaria.

From 2023, we conducted a prospective study to observe the clinico-laboratory profile and treatment outcome of severe vivax malaria as defined by the World Health Organization guidelines. We included all individuals with confirmed vivax malaria by Rapid Diagnostic Test (RDT) kit and peripheral blood smear who presented with severe symptoms to our centre. *P. falciparum* coinfecting cases were excluded. Out of twenty-eight patients with severe *P. vivax* mono-infection, the following severe manifestations were seen in the decreasing order of frequency-jaundice, hypotension & anaemia, followed by ARDS, acute renal failure, haematuria and impaired consciousness, prostration, melaena & haematemesis. Majority had thrombocytopenia with platelet counts of 20,000-80,000 cells/mm<sup>3</sup>. All patients were treated with weight based intravenous artesunate therapy followed by three days of oral artemether-lumefantrine and fourteen days of primaquine for radical cure. All patients were cured except one patient who left against medical advice.

In our study, common complications were hepatic dysfunction, shock, anaemia, ARDS and renal failure. Thrombocytopenia was a universal finding. Though vivax malaria can present with severe manifestations but clustering of severe vivax cases from a single centre and in a single season is not much common. Severe vivax infection is mostly underdiagnosed, we need to be more vigilant and provide timely appropriate treatment to prevent morbidity and mortality.

## A multi-centre randomised controlled trial for comparing the efficacy, safety, and tolerability of 0.5 mg/kg primaquine given over 7 days versus 0.25 mg/Kg primaquine given over 14 days

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The current recommended regimen of 0.25 mg/kg primaquine given over 14 days for *P. vivax* (for preventing relapses) has low-compliance. Therefore, this study was planned to assess the 6-month efficacy and safety of a 7-day course of primaquine treatment regimen (0.50 mg/kg/day) compared to the current 14-day primaquine regimen (0.25 mg/kg/day). An open-label randomized controlled trial conducted in two sites in India (Ahmedabad and Kolkata). The sample size was 400. Patients with uncomplicated vivax malaria, aged more than 16 years, G6PD normal activity, and Hb more than 8 g/dl were enrolled after obtaining informed consent and assessing other eligibility criteria. The patients were randomized in two arms: intervention and control, and followed up for 6-months for assessing efficacy and safety. At two sites, 433 patients were screened, and 400 patients were randomized to intervention (201) and control arm (199), and 368 patients completed 6-month follow-up. The intervention arm showed equivalent efficacy in comparison to the control arm (cumulative Incidence: 1.1% in both arms). In terms of safety outcomes related to haemolysis: one patient in control arm has hemoglobin drop to 5 g/dl in 14 days' follow-up. Intervention arm has equivalent efficacy with no major hemolytic safety concern. Additionally, intervention arm showed better compliance and less loss-to follow up, and thus can be considered for wider adoption in National programme for *P. vivax* management for preventing relapses.

**Journey With *Vivax* Malaria - Benign To Severe****Dr. Sanjay Kumar Kochar<sup>1</sup> , Dr. Dhanpat Kumar Kochar<sup>2</sup>**<sup>1</sup>Senior Professor, Department of Medicine, I/c Malaria Research Centre, Head: Respiratory Division, S.P. Medical College, Bikaner<sup>2</sup>Former Senior Professor, Department of Medicine, S.P. Medical College, Bikaner

Globally, *P. vivax* is the most widely distributed malaria parasite, responsible for millions of infections each year—particularly in Asia and Latin America. In India, it accounts for almost half of the malaria burden. It was long believed to cause only mild illness and therefore called “Benign Tertian Malaria” however, research over the past two and half decades, initially from Bikaner, Rajasthan, and later from different parts of India as well as all over the globe has shown that *P. vivax* can also lead to severe, life-threatening disease similar to *P. falciparum* malaria. The change in understanding began in 2003, when our team noticed unusually severe cases among patients infected only with *P. vivax* and the 2005 landmark series of 11 patients (PCR proved pure *vivax*) reported all sort of complication like jaundice, renal failure, severe anemia, cerebral malaria, acute respiratory distress syndrome, shock, and hypoglycemia. Subsequent studies (2009, 2014) involving over a thousand patients proved that severe *vivax* malaria occurs almost as often and can be just as dangerous as *falciparum* malaria, although risk of developing severe malaria, multi-organ dysfunction and mortality was more in patients of mixed infection. Severe thrombocytopenia was another important finding, and multi-organ dysfunction was detected in almost 50% patients. Later on global reports from Indonesia, Brazil, and Papua New Guinea and various other countries have corroborated these findings, establishing the worldwide existence of severe *P. vivax*. The exact reasons for this severity are still being studied. Possible mechanisms include strong inflammatory reactions, microcirculatory damage, and hidden parasite reservoirs in the spleen or bone marrow. Treatment principles for severe *vivax* malaria are the same as for severe *falciparum* malaria i.e. prompt hospitalization and intravenous artesunate, plus supportive care (oxygen, fluids, transfusions, dialysis if needed). After recovery, radical cure with primaquine is required to eliminate dormant liver stages. Globally in many countries tafenoquine is approved as single dose treatment of radical cure with prior G6PD testing. The world now recognizes that *P. vivax* is not a “benign” infection. It can cause severe disease and death, demanding early recognition, urgent treatment, and global attention to achieve true malaria elimination.

## A Glimpse into the RNA Methylome of Patient-derived *Plasmodium falciparum* and *Plasmodium vivax* isolates

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Epitranscriptomic modifications play pivotal roles in posttranscriptional regulation in *Plasmodium*, yet comprehensive mapping of both N<sup>6</sup>methyladenosine (m<sup>6</sup>A) and 5methylcytosine (m<sup>5</sup>C) modifications in parasites has not been attempted. One clinical isolate each for both the parasite species were subjected to direct RNA sequencing on the Oxford Nanopore platform. Approximately 50% from a total of ~5,000 annotated genes were expressed in each isolate, including novel isoforms and intergenic transcripts. m<sup>6</sup>A and m<sup>5</sup>C signatures were observed across sense transcripts, natural antisense transcripts (NATs), and intergenic regions. Dual modifications were present in 73% of PFC transcripts and 85% of PVC transcripts. Methylated transcripts included those from nuclear, mitochondrial, and apicoplast genomes, with a predominant distribution of modifications within the CDS, implicating roles in mRNA stability and translation. Diverse alternative splicing mechanisms were reported, with predominant alternative 3' and 5' end spliced isoforms, intrachromosomal fusion events, and novel transcripts. Similar methylation patterns within orthologous genes in both the parasite species were also discovered. This first single-base-resolved ex vivo profiling of m<sup>6</sup>A and m<sup>5</sup>C in the two parasite species with severe malaria reveals multiple post-transcriptional regulatory layers that may influence the parasite's inherent proteome plasticity, paving way for informed therapeutic interventions.



## Studying the severe vivax malaria trends in a tertiary hospital in Delhi

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Historically Vivax malaria was considered benign species but recently it is being recognised to be causing severe malaria in the patients similarly like *Plasmodium falciparum*. In the present study the authors report the trends in malaria severity due to *Plasmodium vivax* in the last two years from a Delhi government hospital to understand the disease severity due to severe vivax malaria.

In the reported study, after clinical diagnosis of the patients and tested positive on smear examination and/or rapid malaria antigen test [RMAT] for malaria, the medical case records of malaria positive patients admitted during the years 2017-2018 were analysed.

Of the 205 patients, 177 [86.3%] had *P. vivax* infection, 22 [10.7%] had *P. falciparum* infection and six [2.9%] had mixed infection as diagnosed by PBS and RMAT. A total of 177 *P. vivax* cases were included in this study where 32.7% cases presented one or more manifestations of severe malaria among which, 41 cases had only a single complication, while 17 cases presented

>1 or multiple complications. Severe anaemia (56.9%), jaundice (15%) and significant bleeding (15%) were the most common complications reported. Also, thrombocytopenia was found in a large proportion of patients.

From the data analysed in the malaria patients enrolled in hospital, it is evident that vivax malaria is emerging as the new severe disease in malaria patients, a significant shift in the paradigm of *P. vivax*. The spectrum of complications and alterations in the laboratory parameters in *P. vivax* clinical cases also indicate the recent shift in the disease pathogenesis.



## Integrated Entomo-Epidemiological Investigation of *Plasmodium vivax* Outbreak in a tribal belt along the Bhutan Border: A Matched Case-Control Study from West Bengal, India

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Globally, malaria cases remain a major public-health concern, with an estimated 249 million cases and 608,000 deaths reported in 2023. India has made notable progress toward the National Framework for Malaria Elimination (2030), yet focal outbreaks persist in hard-to-reach tribal and forest- fringe regions. Cross-border transmission poses an added challenge, particularly along porous boundaries adjoining Bhutan, which has entered the sustenance phase of malaria elimination.

To investigate a confirmed *Plasmodium vivax* (PV) malaria outbreak in a tribal tea-garden community at Jayanti TG HSC, Kumargram Block, Alipurduar District, West Bengal (India-Bhutan border area), through an integrated entomological and epidemiological outbreak investigation (OBI), the first of its kind in India, combining descriptive and analytical approaches. A retrospective matched case-control design was adopted (78 case-control pairs) following mass surveys among 5,865 residents. Laboratory-confirmed PV cases (May-November 2024) were compared with age-matched neighbourhood controls. Environmental, behavioural and vector data were collected using structured tools. Odds ratios (OR) and 95% confidence intervals (CI) were computed using R. A total of 252 malaria cases (PV = 250, PF = 2) were detected with an attack rate of 42.9/1,000 population. *Anopheles culicifacies*, *An. fluviatilis*, *An. annularis*, and *An. barbirostris* were the predominant vectors. Tea-garden (OR = 2.18; 95% CI: 1.15-4.13) and forest exposure (OR = 2.55; 95% CI: 1.31-4.94) were significantly associated with infection. The population-attributable fraction (PAF) was 36-38%, indicating strong occupational and environmental determinants. Occupational exposure among tea-garden labourers and forest dwellers, combined with the absence of IRS/LLIN coverage in recent years, fuelled the PV outbreak. Strengthened cross-border surveillance, proactive vector control, and biannual active case detection are crucial to sustain malaria- free status along the Bhutan-India border.



# INVITED LECTURES



## Genetic characterization of *Plasmodium falciparum* histidine-rich protein 2 (*pfhrp2*) deletions in Odisha, India

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The persistence of *Plasmodium falciparum* (*P. falciparum*) malaria in India remains a major public health concern ahead of the country's national framework for malaria elimination (NFME) goal-2030, despite a global decline in the malaria cases and several successful in-country malaria elimination programs in place. The rapid diagnostic test kits (RDTs) based on PfHRP2 (*P. falciparum* Histidine Rich Protein2) are the mainstay of malaria diagnosis, however, recent reports confirm the proportion of false-negative PfHRP2-RDTs are on the rise, owing to the deletion in the parasite *pfhrp2* gene. We investigated the prevalence of *pfhrp2* gene deletions in 15 malaria endemic villages in the state of Odisha, India. The exon 2, exon 1-2 and two flanking genes in the upstream and downstream regions of the *pfhrp2* gene were characterized for the presence of deletions. We found that 61.6% of subpatent *P. falciparum* infections (*i.e.* RDT-negative and positive by PCR) had *pfhrp2* deletions, predominantly located in the exon 2 region (96.2%) and largely identified in samples from febrile individuals (82.6%). DNA sequencing and protein diversity features were characterized in a subset of samples from individuals with subpatent infections carrying intact *pfhrp2* exon 2 loci. Our analyses revealed novel amino acid repeat motifs (231-293 amino acids), and these variant repeat sequences differed from those of RDT+/PCR+ samples. Our findings highlight the need for alternative diagnostic targets and tools as India moves toward its goal of malaria elimination by 2030.

## Synergistic effect of methanol extract derived bioactive fractions of *Cuscuta reflexa* with artemisinin and *in silico* prediction of potential antimalarial compounds

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Ethnopharmacological relevance: Drug resistance to currently used antimalarial drugs provoke development of new drug with broad-spectrum antimalarial activity. Having inspired with previous work, the two most potent column fractions (F3 and F4) derived from ethnomedicinally used *Cuscuta reflexa* were explored further in search of potential antimalarial compounds. Methods: Effect of F3 and F4, and their interactions with artemisinin on *in vitro* growth inhibition of *P. falciparum* (3D7), and *in vivo* suppression of parasitemia including parasite multiplication rate in *P. berghei* ANKA infected mice were determined. Besides, alteration in expression of four different inflammatory cytokines (such as TNF- $\alpha$ , IL-10, IL-1 $\beta$  and IFN- $\gamma$ ) and dye permeability of brain tissues following *P. berghei* infection in mice was assessed by qRT-PCR and Evan's blue staining, respectively. Major phytoconstituents from these fractions were virtually screened for molecular docking against validated drug targets. Results: Both F3 and F4 exhibited synergistic drug interactions with artemisinin and had parasite growth inhibition comparable to artemisinin at equipotent ratio of their corresponding IC<sub>50</sub>. Although significant *in vivo* suppression of parasitemia was demonstrated in all treatment groups, improvement in survival was more pronounced in F4 treated mice, alone and in combination with artemisinin. Moreover, up-regulated expression of anti-inflammatory IL-10, and no dye impregnation in the brains was observed in F4+artemisinin treated group. *In silico* analysis revealed ligands 5283469 and 3277971 derived from both F3 and F4 as promising drug candidates. Conclusion: Evaluation through multiple approaches confirms antimalarial activity of *Cuscuta reflexa*. It harbors important broad-spectrum anti-malarial compounds which could be lead drug and needs to be explored further.

## Advances in Malaria Diagnostics: Bridging the Gap from Conventional Methods to Innovative Solutions

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Malaria, a life-threatening vector-borne disease, continues to pose a significant global health burden, particularly in endemic regions. Accurate and timely diagnosis is essential for effective disease management and control. This presentation explores the evolution of malaria diagnostic methods, tracing the journey from conventional techniques to cutting-edge innovations. Traditional methods, such as microscopy and rapid diagnostic tests (RDTs), have long been the cornerstone of malaria diagnosis, providing reliable results in resource-limited settings. However, these techniques face limitations, including the need for skilled personnel and reduced sensitivity in low-parasitemia cases.

Recent technological breakthroughs, including molecular diagnostics like PCR, LAMP, and CRISPR-based assays, offer unparalleled sensitivity and specificity, enabling the detection of asymptomatic carriers and mixed-species infections. Additionally, the integration of digital tools such as automated image analysis and machine learning enhances diagnostic accuracy and reduces dependency on highly skilled personnel. Emerging biosensor and nanotechnology-based platforms promise rapid, cost-effective, and point-of-care solutions, addressing gaps in remote and resource-limited settings. Recent advancements in artificial intelligence (AI) and machine learning (ML) are transforming the field of malaria diagnostics, bridging gaps in accuracy, speed, and scalability. Additionally, ML models integrated with molecular diagnostic platforms, like CRISPR and LAMP, facilitate rapid and highly sensitive detection of malaria parasites, including drug-resistant strains and mixed-species infections. By bridging the gap between traditional and modern approaches, we can accelerate progress toward a malaria-free world.

## Antibodies to PfGBP-130 inhibit RBC invasion *in vitro* and protect humans against falciparum malaria.

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Even after decades of effort, *Plasmodium falciparum* malaria continues to be a leading cause of death in children. Effective public health interventions are constrained by the absence of a very effective vaccine and the emergence of parasites resistant to both detection and frontline therapeutic medication. Current malaria vaccines, RTS, S, and R21, are only moderately effective pre-erythrocytic vaccines and do not confer sterile protection. This situation necessitates the development of innovative novel blood-stage vaccine candidates that will attenuate clinical disease and incorporate effective blood-stage components into the current pre-erythrocytic vaccines. We employed our whole parasite proteome screening method to discover new vaccine candidates. We identified PfGBP130 as a parasite protein recognized specifically by antibodies from children resistant to *P. falciparum* infection but not recognized by antibodies from susceptible children. We formulated PfGBP130 as lipid-encapsulated mRNA, DNA plasmid, and recombinant protein-based immunogens and evaluated the efficacy of murine polyclonal anti-PfGBP130 antisera to inhibit parasite growth *in vitro*. Immunization of mice with PfGBP130-A (aa 111–374), the region identified in our differential screen, formulated as a DNA plasmid or lipid encapsulated mRNA, but not as a recombinant protein, induced antibodies that inhibited RBC invasion *in vitro*. We also constructed a VHH antibody phage display library containing phages that specifically recognized PfGBP130. We grafted PfGBP130-binding VHH onto a human IgG1 constant region and expressed and purified these camelid/human chimeric mAbs. Three out of 22 of these chimeric VHH-hIgG1 inhibited parasite growth, while 19 had no activity *in vitro*. We are currently evaluating the efficacy of these VHH-hIgG1 *in vivo* in the NSG/*P. falciparum* challenge model. Based on our data demonstrating that anti-PfGBP130-A antibodies generated by lipid-encapsulated mRNA-based vaccination and humanized monoclonal antibodies attenuate parasite growth, we are advancing this antigen/delivery platform combination toward vaccine trials in non-human primates.



## **Cerebral and Severe Malaria – An Unmet Need of an Adjunct Drug**

**Arun Nagaraj\***

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Malaria remains a major health concern claiming ~0.6 million lives every year mainly affecting the children below five years of age. The mortality due to cerebral and severe malaria occurs despite the antimalarial treatment and parasite clearance. Hence, there is a need for an adjunct drug to prevent malaria mortality. Efforts made in the past for a suitable adjunct drug by various groups have not come to fruition. In my talk, I will highlight our research work on the role of parasite heme in disease pathogenesis and the potential of targeting it for adjunct therapy to prevent malaria mortality. Parasite heme is essential for the mosquito and liver stages, and dispensable for the blood stages. Interestingly, the blood-stage parasites synthesize heme despite having the ability to acquire host heme through hemoglobin degradation. The new insights gained on the role of parasite heme in the blood-stage infections and the attempts being made for clinical trials in humans using griseofulvin as an adjunct drug with the existing artemisinin-based combination therapies for cerebral and severe malaria will be discussed.

## Malaria Elimination in High Transmission Hard-to-Reach Areas of Odisha (India): State's Initiatives and Challenges

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Malaria poses immense challenge in Odisha, an east coast state of India since decades. Odisha contributed 40% of country's annual malaria cases and 91.5% falciparum malaria in 2011. Asymptomatic malaria poses immense problem causing malaria persistence in state's remote villages with vast forest tracks and limited transportation facility. Odisha adopted "Durgama Anchalare Malaria Nirakaran" (DAMaN) strategy meaning malaria elimination in hard-to-reach areas in 2017 along with implementation of large scale Long Lasting Insecticidal Net (LLIN) and community awareness campaign. DAMaN's key components for malaria elimination are mass screening and treatment (MSaT), vector control and community mobilisation.

We conducted the evaluation study to examine the beneficial outcomes of the DAMaN programme, explore the DAMaN's implementation challenges and potential strength for its enhancement for malaria elimination efforts. The study was conducted in six sampled districts representing different physiographical regions of the state. Data collected from two biennial surveys, state's routine malaria programme and community-based interviews were analysed using appropriate tools and methods for each category. Within a few months of DAMaN implementation in 2017, malaria cases started declining with 80.9% reduction within a year. The DAMaN villages showed remarkable case reduction, and the overall decline continued till 2022 despite COVID 19 pandemic during 2019-2021. Malaria positivity rate was significantly higher during the first biennial survey in 2019, compared to the second biennial survey in 2021. An alarming proportion of asymptomatic malaria (75% to 100%) was found during second biennial survey. Perspective of the community level health care givers and community opinion leaders on DAMaN was largely encouraging.

Malaria reduction was notably high in areas where DAMaN's programme was effective. The overall positive findings indicate that DAMaN being a community-based malaria elimination initiative in far-flung villages of Odisha state, serves as a suitable supplement to the routine malaria control and elimination programme and is replicable.

## Genomic evidence for ongoing zoonotic infection of *Plasmodium falciparum* malaria in India

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Human malaria is a vector borne disease and is aimed to be eliminated in India by the year 2030. There are several factors that might hinder complete malaria elimination in specified time; one of such aspects is possible zoonotic transmission of the human malaria parasite(s). While the zoonotic malaria parasite *Plasmodium knowlesi* is known to be present in the Andaman and Nicobar Islands, similar transmission patterns might be found for other malaria parasites in other parts of the country. In order to test this hypothesis, we sequenced several whole mitochondria genomes of the deadliest form of human malaria parasite, *P. falciparum* collected in different malaria endemic locations in India. DNA sequences were also generated for *P. falciparum*-like malaria parasites (*P. coatneyi* and *P. fragile*) infecting Indian Rhesus macaque. Comparative and phylogenomic analyses provided evidence for a new, hitherto unreported form of *P. falciparum* (named as *PfIndia\**) that is circulating in both Indians and Indian Rhesus macaque. Further evidence for infection of *P. falciparum* in Indian Rhesus and Bonnet macaque for the first time indicate ongoing zoonotic transmission of *P. falciparum* in India. These research-based evidence suggest that malaria elimination will face an uphill task if zoonotic transmission potentials of Indian malaria parasites is not considered in the program.

## Apoptosis and Survival: A Balancing Act in the Life Cycle of *Plasmodium*

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Apoptosis, a form of “cellular suicide” has long been believed to be confined to metazoans. Indeed, it was considered counter-intuitive and evolutionarily unfavorable in unicellular organisms until recently few reports indicated typical mammalian programmed cell death features from several unicellular organisms including protozoan parasites. Apoptosis plays an important role in density regulation of parasite in both the host and the vector, thereby preventing the premature mortality of the host before a small part of the parasite population is transmitted to another host. In addition, it helps to remove damaged, aged or genetically disadvantageous parasites from the population and saves resources for survival of the fittest. Despite knowing *Plasmodium* for more than a century, apoptotic pathways remain uncharacterized in the parasite. This is due to the fact that the major regulators of apoptosis in metazoans; the caspases and the members of the Bcl family are reported to be absent in *Plasmodium*. This indicates to either an absence of apoptotic pathways in the parasite or suggest different underlying apoptotic mechanisms.

Here, to delve into the pathways of programmed cell death in the apicomplexan *Plasmodium falciparum*, we examined the parasite growth under the influence of apoptotic inducer Etoposide, along with the present chemotherapeutic drugs used for malaria treatment. *Plasmodium falciparum* culture when treated with various drugs, such as Chloroquine, Artemisinin and etoposide presented a reduced cell size. To identify the major players of cell death in *Plasmodium*, we initiated our studies with *in silico* homology based search of the *Plasmodium* genome, where we identified several putative cell death-associated genes encoding proteases, nucleases and several apoptosis regulators. This included previously identified Meta-caspases from *Plasmodium*, which share homology with caspases, but have different roles assigned to them in the parasite. Drug-treated parasites exhibited differential regulation of several of the above shortlisted genes in a time-dependent fashion suggesting that *P. falciparum* possesses ancient apoptosis-like cell death machinery which can be triggered by chemotherapeutic agents.

## **Parasitic Co-infections in Endemic Regions: Interactions of Visceral Leishmaniasis and Lymphatic Filariasis with Leprosy and Tuberculosis, and Their Implications for Diagnosis, Treatment, and Control**

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In co-infections, pathological interactions, diagnostic hurdles, and control challenges link visceral leishmaniasis (VL) and lymphatic filariasis (LF) with leprosy and tuberculosis (TB) in endemic settings. The immunomodulation caused by VL and chronic filarial infections skews host responses toward regulatory and anti-inflammatory phenotypes, down-regulating Th1/Th17 immunity required for containment of intracellular organisms; this predisposes to reactivation or dissemination of TB and complicates the clinical picture. Leprosy and VL share spectral immune presentations, and rare co-infections produce overlapping skin and systemic findings that can delay correct classification and treatment. Diagnostic assays are affected in multiple ways: serological tests for VL lose sensitivity in immunosuppressed or co-infected patients, PCR on peripheral blood shows variable sensitivity and is often needed to confirm cases, and antigen/antibody assays for LF have cross-reactivity and variable windows of detection. Smear microscopy and histopathology for leprosy and TB can be confounded by secondary infections and altered granulomatous responses in patients with helminth or leishmanial co-infections, leading to false negatives or misinterpretation. Treatment challenges include drug–disease and drug–drug interactions, overlapping toxicities, and paradoxical immune reconstitution reactions when antiparasitic therapy restores cell-mediated immunity, unmasking mycobacterial or mycobacterial-like disease. Control programs face operational problems: integrated case finding is hampered by diagnostic gaps, and mass drug administration for LF may not reach individuals with occult VL or leprosy who require case management. Addressing these problems requires combined strategies: strengthen molecular diagnostics (multiplex PCR), implement routine screening for common co-infections in endemic areas, adapt treatment algorithms to account for immunosuppression and interaction risks, and integrate NTD and TB/leprosy services for surveillance and morbidity management. Research priorities include validated multiplex diagnostics, operational studies of integrated care pathways, and immunological studies to guide adjunctive therapies. Policy support and sustained funding for integrated approaches are essential to prevent diagnostic delays, reduce mortality, and achieve control targets across these overlapping diseases.

## Molecular Xenomonitoring of filarial infections using non-vectorized mosquitoes

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At least five parasite-vector combinations are evident in the transmission of different races of filarial parasites. *Wuchereria bancrofti* is the most prevalent infection, responsible for about 90% of the global burden of filariasis and is transmitted primarily by *Culex quinquefasciatus*. This vector species is amenable to collection using gravid traps in sufficient numbers for monitoring infection. Molecular xenomonitoring (MX) can be one of the four WHO-recommended surveillance strategies. Endemic countries are encouraged to use any two strategies for post-validation surveillance of the filariasis elimination programme. MX is an indication of an early signal of transmission risk and the need to respond when it exceeds the threshold of infection. Either two rounds of targeted MDA (preventive chemotherapy) can be undertaken in the MX-positive clusters, or confirm the risk by conducting a blood survey in humans following the XenoFil approach (MX and serology). However, the entire process relies on the collection of at least the estimated sample size for a critical cutoff value of 0.5% vector infection, which is derived from 1% threshold of human infection. A number of mosquito sampling techniques are used to meet the sample size. MX is considered a proxy to human sampling, and expects every sampled mosquito to have at least one contact with a human. MX is a potential surveillance tool, but it samples vectors other than *Cx. quinquefasciatus* which is challenging due to varied feeding behaviour. Alternatively, the use of predominantly anthropophilic non-vector mosquito species as a proxy for the vector mosquitoes can be a better option for monitoring. Parasite DNA is reported to persist for a longer period of time, and past infection could also be detected. *Cx. quinquefasciatus*, a ubiquitous mosquito, can be used for any situation other than diurnally periodic parasite species.

Balasore, Odisha, is endemic for both *W. bancrofti* and *B. malayi*, and we sampled mosquitoes using various sampling methods. About 90% of 2903 mosquitoes were *Cx. quinquefasciatus*. *Mansonioides* constituted only about 7% of the collection. Pools of *Cx. quinquefasciatus* were detected with *W. bancrofti* DNA, and none of the *Mansonioides* were positive with either of the parasite species. This could probably be due to a very low sample size. *Cx. quinquefasciatus* is a non-vector of *B. malayi* infection. Studies have shown the presence of *B. malayi* parasite DNA both in vectors and non-vector mosquitoes in Indonesia, and a similar report of *W. bancrofti* in vector and non-vector species is from Mali. The situation where xenomonitoring using non-vector may be undertaken, and the need to use appropriate sampling methods are discussed.

## The Rickettsia Paradox: Simple treatment, complex diagnosis-Untangling the web of rickettsial diseases

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Rickettsial diseases, caused by obligate intracellular gram-negative bacteria of the genera *Rickettsia* and *Orientia*, are a significant yet under-recognised cause of acute febrile illness (AFI) in Northeast India. This region is hyper-endemic for scrub typhus, spotted fever group, and typhus group rickettsioses, with outbreaks increasingly reported over recent years. Despite the diseases' potentially severe complications, including multiorgan dysfunction and death, treatment remains straightforward with timely administration of doxycycline or related antibiotics. Yet, diagnosis remains a major challenge due to the heterogeneous, nonspecific clinical manifestations and limited access to specific confirmatory laboratory tests. Overlapping symptoms with other tropical febrile illnesses, co-infections, and a lack of widespread awareness among healthcare providers complicate the accuracy of diagnosis. Serological tests such as the Weil-Felix test, rapid immunochromatographic assays, and PCR-based techniques have variable sensitivity and limited availability in rural and resource-limited settings. This diagnostic paradox often leads to delayed or empiric treatment, which may mitigate mortality but complicates epidemiological tracking and precise case management. Epidemiological studies from this region reveal a high burden of rickettsial diseases in all eight NE states of India, with notable case fatality rates despite antibiotic sensitivity. Public health responses integrating enhanced screening, clinician awareness programs, and vector control are critical to reducing disease impact. Further research into local vector ecology, host reservoirs, and the socio-environmental determinants is vital for comprehensive control strategies.

This study highlights the urgent need to untangle the complex clinical and diagnostic web surrounding rickettsial diseases. Strengthening laboratory infrastructure, improving clinician education on endemic rickettsioses and ensuring prompt treatment can mitigate morbidity and mortality associated with these deceptively simple yet diagnostically challenging infections.

## Innovations in Dengue Virus detection

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Dengue is one of the fastest-growing diseases in the world, affecting mostly tropical and sub-tropical nations. Globally, this illness is regarded as a serious public health issue. The dengue virus, a significant arbovirus, is the cause of dengue infection and is transmitted by female *Aedes* mosquitoes. The dengue virus (DENV) is a single-stranded RNA virus with four serotypes (DENV 1–4). Any one of the four serotypes might result in dengue, from asymptomatic primary infections to deadly secondary infections like dengue hemorrhagic fever (DHF) and dengue shock syndrome (DSS). As of right now, dengue sickness cannot be prevented or treated with a licensed vaccine or antiviral medication. This makes recovery challenging and time-consuming. As a result, timely and precise laboratory diagnostic testing is desperately needed for patient triage and disease confirmation.

Reverse transcriptase PCR amplification, viral detection in cell culture, and serological testing are the conventional diagnostic methods for the dengue virus. The current methods that have been developed for the detection of various Dengue virus biomarkers and serotypes are optical, electrochemical, microfluidic, enzyme-linked immunosorbent assay (ELISA), finite-difference time-domain (FDTD) method, biosensor using DNA nanotechnology, CRISPR/Cas system integrated with biosensing platforms and smartphone-based biosensors. Furthermore, the role of bio recognition molecules such as lectins, nucleic acids, and antibodies in electrochemical sensing of DENV has been studied. The current study demonstrates a thorough analysis of the current fabrication techniques and designs used in the development of electrochemical biosensors for the early detection of dengue. Moreover, benefits and drawbacks have been thoroughly examined from a variety of angles, including the type of biomarker, sensitivity, accuracy, rate of detection, commercialisation potential, availability, detection limit, linear range, simplicity, detection mechanism, and clinical application suitability. In addition, the present work demonstrates the significance of the electrochemical approach in contrast to conventional diagnostic techniques.



## Effect of Urban Heat Island and vector-borne diseases in India

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Rapid unplanned urbanization in India is resulting in major vector-borne diseases in India. Most importantly, urban malaria transmitted by *Anopheles stephensi* and *Aedes* borne diseases like dengue and chikungunya. Urban areas usually experience the urban heat island (UHI) effect, significantly warmer than surrounding rural areas. Urban areas occupy about 0.5% of the Earth's land surface that hosts more than half of the world's population. The UHI phenomenon affects our daily life by exacerbating heat discomfort, increased energy consumption, and urban air pollution. It has been observed that UHI intensity varies across the country between 2 and 10 °C, with the northwest seeing a more pronounced temperature gradient. In recent years, most Indian cities are experiencing more cases of dengue and malaria, for example, Delhi, Mumbai, Kolkata, Hyderabad and Bangalore. In 2024, Karnataka reported highest number of dengue cases, and Bangalore contributed almost half of the cases. In Bangalore city, malaria cases are reported mainly from migratory cases, but the ongoing spread on *Anopheles stephensi* needs a proper study.

## Surveillance of bats and pathogenic ecology for overcoming pandemic earth

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Bats, belonging to the order Chiroptera, are the only mammals capable of true flight—an evolutionary adaptation that has enabled them to thrive in diverse ecological niches. Primarily nocturnal, they are classified into two suborders: Megachiroptera (fruit-eating bats) and Microchiroptera (insect-eating bats). Their roosting habits vary widely, with colonies found in caves, buildings, temples, trees, hill slopes, and even urban structures. One of their most remarkable traits is echolocation, an acoustic navigation system that helps them detect obstacles and locate prey in complete darkness.

Ecologically, bats play vital roles. Megachiropterans serve as key pollinators and seed dispersers—a process called chiropterochory—contributing to forest regeneration. Insectivorous bats act as natural pest controllers, consuming vast numbers of insects and reducing the need for chemical pesticides.

However, bats are also known reservoirs of numerous pathogens. They carry and transmit a wide range of viruses, bacteria, and fungi capable of infecting humans and animals. Notable zoonotic viruses linked to bats include SARS-CoV, SARS-CoV-2, Nipah, and Hendra, responsible for outbreaks with high mortality. While bats have been suggested as the original source of COVID-19, the intermediate host remains uncertain, with pangolins also implicated. Transmission can occur directly or indirectly through animals such as pigs, horses, or cats.

Environmental and socio-economic changes—such as deforestation, climate change, urbanisation, and wildlife trade—have intensified spillover events from bats to humans. Research shows that ecological disruptions increase pathogen loads in bat populations, heightening the risk of emerging infectious diseases. Studies have identified nearly 200 viruses, 290 fungi, and many bacteria associated with bats, underscoring their importance as zoonotic reservoirs.

Preventing future pandemics requires enhanced surveillance of bat ecology, including behaviour, migration, and population dynamics. Using tools like GIS, telemetry, and AI-driven monitoring can improve early warning systems and strengthen global preparedness for emerging infections.

## Multi-epitope protein based medical countermeasures against tick-borne viruses

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Tick-borne viruses are emerging and reemerging as major One Health challenges in many parts of the world, including India. Crimean-congo hemorrhagic fever (CCHF) and Kyasanur forest disease (KFD) constitute two most important tick-borne viral infections affecting many parts of India. CCHF infection remains sporadic; however, KFD is expanding its geographical territories across the Western Ghats. Both these zoonoses are caused by high-risk viruses. No specific medical countermeasures, including therapeutics or vaccines, are currently available against CCHF and KFD. This emphasises the importance of early detection for patient care and disease containment. The vast diversity of RNA viruses and the existence of multiple genotypes make the development of an antigenic candidate highly challenging using a conventional approach. The recent advancement in immuno-informatics enables the design of conserved B and T cell epitopes. The epitopes were selected from immunogenic structural and non-structural proteins and linked together to construct a multi-epitope protein (MEP). The MEP was expressed and purified to homogeneity using a prokaryotic expression system. The evaluation of MEP antigens against native viral antigens based immunodiagnostic system reveals high concordance, indicating their relevance for immunodiagnosis. The MEP candidate has the potential to serve as an effective antigen or immunogen for the development of medical countermeasures, including diagnostics and prophylaxis for emerging tick-borne viral infections.

## Decoding the involvement of ABC transporter genes in *Aedes aegypti* midgut immune modulation

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The midgut of *Aedes aegypti*, a primary arboviral vector, is the main site for digestion and possible site for major immune interactions. Adults mosquitoes can be exposed to different microbes through nectar feeding and physical injuries. In addition, their haematophagous nature exposes them to microbes of the host skin and different blood-borne pathogens like dengue virus (DENV), Zika virus (ZIKV), and malaria parasite *Plasmodium* through blood-feeding. These interactions result in production of several immune molecules that are responsible to protect the midgut from invading pathogens and limits their propagation. However, the details of ABC transporters in insect midgut immunity remains unexplored. In our previous report we analyzed the mosquito midgut transcriptome following pathogen exposure which indicated to the involvement of ABC transporter genes in mosquito immunity. To explore the midgut-specific ABC transporters, we performed gene expression profiling of ABC transporter genes by challenging the mosquitoes with a mixture of gram-positive and gram-negative bacteria by systemic and local bacterial challenge. Among the genes showing upregulation, one gene was shortlisted for RNAi-mediated gene silencing to assess its role in midgut immune responses.

We observed expression of 48 microbial-induced midgut-expressing *Ae. aegypti* ABC transporter genes upon systemic or local bacterial challenges. We used immune genes *defensin* and *cercopin* as standard to comparatively analyse expression profile of *AaeABCG3* using RNAi-mediated gene silencing and characterization. The *AaeABCG3* silencing affected the mosquito midgut immune response and disturbed the midgut microbiota homeostasis. The involvement of conventional immune pathways JAK-STAT, Toll and IMD immune pathways was also explored.

## Changing scenario of Vector Borne Diseases in view of Climatic and environmental changes

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Vector-borne diseases (VBDs) continue to pose a major global public health challenge, even as some diseases such as malaria, visceral leishmaniasis, and lymphatic filariasis progress toward elimination. At the same time, several other VBDs are expanding geographically due to environmental, ecological, and climatic changes. Diseases like scrub typhus, Kyasanur Forest Disease (KFD), Chandipura virus, Crimean-Congo Hemorrhagic Fever (CCHF), and cutaneous leishmaniasis have spread into new territories, creating fresh obstacles for disease control and elimination programmes. Understanding the ecological and climatic factors responsible for these shifts is essential for developing preparedness strategies and preventing the reintroduction of diseases after elimination.

Ecological transformations have played a significant role in modifying malaria transmission patterns in India. The Malnad region of Karnataka, once heavily malarious due to the presence of forest streams that supported breeding of *Anopheles fluviatilis*, experienced a decline in malaria after deforestation and land conversion to plantations eliminated the natural breeding habitats of the vector. In contrast, irrigation projects like the Visvesvaraya and Almatti dams in northern Karnataka unintentionally increased malaria risk. Although cement-lined canals prevented mosquito breeding, seepage water created new drainage channels that became ideal habitats for *Anopheles culicifacies*, leading to persistent malaria in previously water-scarce districts such as Raichur, Gulbarga, Bijapur, and Bagalkot. Similar ecological effects were observed in Assam, where deforestation replaced *Anopheles minimus* with *Anopheles culicifacies*, altering vector dominance and improving conditions for malaria transmission.

Climate change is now emerging as a major driver of VBD expansion. Rising temperatures, unpredictable rainfall, and sea-level rise are projected to alter both the spatial and seasonal distribution of vectors such as mosquitoes and ticks. Warmer conditions will make previously unsuitable regions conducive to transmission. This trend is already visible in the Himalayan states, where malaria and dengue have been reported in higher-altitude areas that were once free from these diseases. Outbreaks in Uttarakhand's Nainital district and Himachal Pradesh's Solan district indicate a clear upward shift in vector distribution.

Other VBDs are also expanding rapidly. Scrub typhus, once restricted to limited regions, is now prevalent in Rajasthan and the northeastern states. KFD has moved beyond Karnataka into Kerala, Goa, Tamil Nadu, and Maharashtra. Chandipura virus and CCHF have also spread beyond their earlier locations, while cutaneous leishmaniasis has appeared in new states including Himachal Pradesh, Jammu & Kashmir, and Kerala. These changing patterns highlight the urgent need for continuous ecological monitoring, climate-linked surveillance, and timely intervention to protect elimination gains and prevent future outbreaks.

## Investigation of *V1016G kdr* Mutation during Pyrethroid Insecticide Resistance in *Aedes aegypti* from Nagapattinam district of Tamil Nadu, India

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*Aedes aegypti* is a primary vector of Dengue, Chikungunya, and Zika viruses, and its control relies heavily on pyrethroid insecticides. However, the development of resistance in mosquito populations poses a major challenge to effective vector control. This study aimed to investigate the presence of the V1016G knockdown resistance (*kdr*) mutation in *Aedes aegypti* collected from Nagapattinam district, Tamil Nadu, India. Where insecticide uses is common.

*Aedes* immature were collected from selected field sites and subjected to insecticide susceptibility testing using World Health Organization (WHO) guidelines. Genomic DNA was extracted, and allele-specific PCR was performed to detect the V1016G mutation in the voltage-gated sodium channel gene (*vgsc*). The analysis revealed the presence of the V1016G mutation in heterozygous form (VG) among the tested samples. No homozygous mutants (GG) were detected. The presence of heterozygous individuals indicates an early stage of resistance development, likely due to ongoing insecticide selection pressure. Bioassay results showed reduced susceptibility to pyrethroids in these populations, supporting the molecular findings. This study provides baseline data on the genetic mechanism of pyrethroid resistance in *Aedes aegypti* from Nagapattinam district. Regular monitoring of *kdr* mutations is essential to detect resistance early and guide insecticide use policies. Integrating molecular surveillance with conventional control measures will help delay the spread of resistance and improve the long-term success of vector management programs. The findings highlight the need for alternative control strategies and responsible insecticide use to maintain the effectiveness of current interventions.

## Kala-azar Situation in West Bengal: An Elimination Success Story

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Kala-azar (Visceral Leishmaniasis) is a neglected tropical disease that disproportionately affects the poorest populations. The first modern medical description was reported from Bengal in 1827, with major outbreaks documented in Jessore during 1824-1825. The term “*Kala-azar*”, meaning “black disease,” was coined in the 19th century from the characteristic skin pigmentation of affected patients. Globally, the disease remains endemic in 83 countries, closely linked to poverty and malnutrition. Each year, 50,000-90,000 new cases occur, mainly in Brazil, East Africa, and India.

In India, 633 blocks across 54 districts in four states: Bihar, Jharkhand, Uttar Pradesh, and West Bengal, are endemic. In 2024, only 449 cases were reported nationally, reflecting a 95% decline since 2014, with all endemic blocks achieving the elimination target of less than one case per 10,000 population. In West Bengal, 120 blocks across 12 districts remain endemic; however, the state has maintained elimination since 2017. Reported cases declined to 33 in 2024, marking a 95% reduction compared with 2014.

State-level innovative initiatives have sustained this progress. These include Indoor Residual Spraying (IRS) with Alphacypermethrin and pre- and post-spray entomological assessments, enhanced wage-loss compensation for PKDL cases, integration with the National Leprosy Eradication Programme (NLEP), intensified Active Case Search (ACS) rounds, strengthened pharmacovigilance, nutritional supplementation, operational research on novel diagnostics, and capacity building of health workers.

Sustaining elimination remains challenging due to rising PKDL and HIV-VL co-infections, inter-state movement and tracking of cases, delayed treatment-seeking, poor private-sector referrals, immature sandfly control, and the long duration of PKDL treatment, causing poor adherence. To address these, emphasis is placed on intensified IEC campaigns, community engagement, complete IRS coverage, research on immature control, and improved supervision and monitoring. Further research on shorter PKDL treatment regimens and host-parasite immune interactions could help optimize management strategies and reduce transmission risk, ensuring long-term elimination.

## Mosquito-directed PROTACs to block malaria transmission

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The mosquito stage of the *Plasmodium falciparum* life cycle is an attractive target for intervention since it is crucial for the sexual reproduction and transmission of parasites to human host. Mosquito determinants crucial for parasite infection and growth pose as lucrative targets for transmission blockers. Owing to the fact that p38 MAPK has role in immune response and vector competence, we have evaluated the potential of PROTAC molecule (NR-7h) to degrade *Anopheles stephensi* p38 MAPK (Asp38 MAPK), a conserved serine/threonine kinase involved in stress reactions, midgut homeostasis, and parasite survival. PROTAC-mediated degradation of Asp38 MAPK led to the disrupted development of the parasite, suggesting its crucial function in vector competence. Furthermore, NR-7h-treated mosquitoes showed higher expression of immune genes such Rel-2, TEPI, APL1, and NOS, suggesting that p38 MAPK regulates host immunity in a way that promotes parasite persistence. PROTAC-mediated degradation of target proteins, provides a more persistent and resistance-proof therapeutic effect than traditional kinase inhibitors. Our findings establish PROTACs as a novel vector-targeted strategy for the development of endectocides to limit malaria transmission.



## From Outbreaks to Ownership: Community-Driven and Digital Innovations in Dengue Control from Urban Odisha

### From Outbreaks to Ownership: Evaluating Community-Driven and Digital Innovations Strategies for Sustainable Dengue Control in Urban Odisha”

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The dengue scenario in Odisha has transitioned from a seasonal to a perennial pattern, creating a sustained public health challenge with a major concentration of cases in urban areas. Bhubaneswar Municipal Corporation (BMC) alone contributed over half of the state's dengue burden in 2021. Recognizing the limitations of reactive outbreak responses, the state adopted a proactive, year-long five-pronged strategic intervention emphasizing (i) community engagement, (ii) digital surveillance, (iii) IEC-Behaviour Change Campaigns (BCC) campaigns, (iv) Health system preparedness, and High Ownership and People-Centred Interventions (HOPCI) strategy focusing on sustained ownership, interdepartmental accountability, and community-led source reduction. A descriptive analysis was conducted using secondary surveillance data, entomological surveys, and programmatic reports from 2021–2024. Trends in test positivity rate (TPR), district contribution, and surveillance coverage were analysed to measure impact.

The results highlight the significant innovations and best practices. Community engagement mobilized 850 frontline volunteers (ASHAs, Anganwadi Workers, Swachha Sathis) who mapped 13,000 lanes for source reduction, reinforced by household “No Dengue Breeding Source” pledges fostering responsibility. Digital Surveillance through the “Safa Mobile App” facilitated real-time mapping of fever cases and larval habitats, triggering rapid hotspot response. IEC and BCC expanded public outreach through School-based campaigns, “NIDHI RATHA” IEC vans, hoardings, and dengue awareness through garbage vehicles. Health System Preparedness was strengthened with dedicated dengue wards, entomological risk assessments, and inter-departmental microplanning, ensuring readiness. “HOPCI” approach: Institutionalized ward-level community ownership mechanism with fortnightly reviews linking health, sanitation, and community groups, leading to sustained source reduction and improved accountability.

The intervention produced a measurable epidemiological impact. TPR declined from 13.03% (2022) to 7.86% (2023) and 5.7% (2024). Khurda district's share in state cases dropped from 51% (2021) to 16.5% (2024), while surveillance coverage rose by 5.7% in 2024. Odisha's “*From Outbreaks to Ownership*” model, reinforced by the HOPCI strategy, demonstrates that community-led accountability, people-centred interventions, and digital surveillance can substantially reduce dengue transmission in urban ecosystems. The approach offers a scalable framework for integration under the National Urban Health Mission and other vector-borne disease programmes.

## An Update on Impact of Ecological Changes on Culicinae Diversity of Punjab

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Biodiversity loss is a major factor in the spread of vector-borne diseases which in turn have negative impact on the economy and human health. Further change in land use can alter the diversity, distribution, abundance and feeding patterns of mosquito populations due to alternations in landscapes they occupy. The change in mosquito populations can influence the transmission dynamics of emerging and re-emerging diseases that are transmitted by them.

During the period of last 5-6 decades, the ecology of agricultural state of Punjab has tremendously changed owing to the change in the cropping patterns. In addition to this, the cultivation of paddy crop during summer and monsoon months has made the availability of water throughout the state in abundance. This has increased the mosquito transmitted diseases in different parts of the state. The knowledge on vector species is scanty. The present study attempts to compile the information on diversity and abundance of mosquito populations in different districts of Punjab.

Our study concluded that many species have completely disappeared whereas many have made their way in this prosperous state of North India during the last 2-3 decades. *Culex quinquefasciatus* is the dominant species throughout the state. There has been a change in distribution trend of vector species especially *Aedes aegypti* and *Aedes albopictus* as a result of which diseases like Dengue and Chikungunya are spreading on a large scale within the state.

## Purification, identification and characterization of larvicidal proteins from *Calotropis procera* leaves

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The health of people worldwide is greatly impacted by vector-borne diseases, which account for 17% of all infectious diseases. Mosquitoes are vectors of diseases such as Zika, West Nile fever, Dengue, Chikungunya, Malaria, Lymphatic filariasis, Leishmaniasis, etc. Malaria is one of the most life-threatening infectious diseases caused by vector *Anopheles* mosquitoes. The outbreak of malaria due to climate change in some countries emphasizes the critical need for improved strategies to treat, prevent, and control malaria. The urban malaria vector *Anopheles stephensi* has developed resistance to multiple pesticides. With around 40% of people in sub-Saharan African cities, where *An. stephensi* has invaded, this is worrying. Mosquito control is the most efficient way to decrease malaria transmission at the community level. To control mosquito transmission, there is a need to search the natural agents from botanical sources with selective biopesticides. *Calotropis procera* has been widely used as a traditional medicine in recent times to treat various diseases. The present work is carried out to evaluate the larvicidal potential of *C. procera* against *Anopheles stephensi*. In the present study we purified, identified and characterized the larvicidal protein from *C. procera*. The crude protein from leaves showed 100% mortality of 3rd instar larvae of *An. stephensi* at the concentration of 5.5 mg/ml after 24 h of exposure. The leaves crude protein was further purified by ion exchange chromatography and eluted fractions were tested for larvicidal potential. The purified single protein fractions L2 and L3 from *C. procera* leaves showed 100% mortality at concentration of 0.06 mg/ml. The homogeneity of purified protein was confirmed by SDS-PAGE and two bands of 26 kDa and 15 kDa proteins were observed. The peptide sequence "R.SQMLENSFLIENVMKR.L" was identified in the trypsin-digested homogenous protein fraction L2 and "R.DRGSQKR.N" peptide sequence in L3 fraction by LC-MS/ESI-MS. The CprL2 peptide showed sequence similarity with the protein maturase K and CprL3 peptide showed the highest sequence similarity with ribosomal protein L20 of *C. procera*. The identified proteins showed strong larvicidal efficacy at very low concentrations. The identified proteins are novel and natural larvicidal agents against *An. stephensi* and hence can be used to control malaria.

## **Bio-surveillance of Hard ticks from Madhya Pradesh for presence of Rickettsial pathogens.**

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Ticks are arthropod vectors that transmit pathogens during biting and bloodsucking, and carry or transmit variety of bacterial, viral, rickettsial agents. Most of these agents are natural route of zoonotic diseases, globally. Over past few decades, occurrence and distribution of TBDs have been steadily increasing in many continents leading to morbidity and mortality. The constant rise in incidence and geographical spread of these diseases necessitates implementation of surveillance systems to monitor their occurrence, spread, and effect on human health. Understanding the rise of these diseases, their impact on public health, and the crucial role of surveillance is essential in designing effective prevention and management strategy.

A comprehensive surveillance of ticks from Gwalior and adjoining districts was conducted from in last two years. Around 1600 ticks were collected from various host animals and flag drag. Collected ticks were morphologically identified with taxonomic keys. It was found that *Hyalomma*, *Haemaphysalis* genus ticks were predominant on host collection whereas *Haemaphysalis* was predominant in flag drag. Tick pools were screened using RT-PCR for presence of *Rickettsia*, *Borellia*, *Coxiella*, *Bartonella*. RT-PCR and nested PCR revealed presence of Spotted fever rickettsia (SFGR) in ticks. SFGR positivity was around 15%. *Hyalomma*, *Rhipicephalus* and *Haemaphysalis* genus ticks were found to acquire SFGR. Presence of SFGR in ticks pose risk of spotted fever in these regions. The detailed study is therefore require for Bio-surveillance of ticks and monitoring presence of bacterial/ rickettsial pathogen in ticks.

## Lethal ovitrap (EcoBioTraps<sup>®</sup>): an innovative vector control device for Integrated Vector Management

**Das N – Research Scholar and Programme Manager\***

Evaluation of EcoBioTraps<sup>®</sup> Project, Bruhat Bengaluru Mahanagara Palike (BBMP), Karnataka

Integrated Vector Management (IVM) which combines physical, chemical, and biological methods, is indeed essential for controlling and eliminating vector-borne diseases. The major challenges in the IVM are insecticide resistance, climate change and poor community engagement. Therefore, current vector control strategies require innovative solutions. The EcoBioTraps<sup>®</sup> is a device that can be effectively integrated into IVM.

Two-month longitudinal study was conducted in urban and rural area of Bengaluru during September – October 2024, which was endemic for dengue in the area. The study covered 1,863 households. Total 480 number of EcoBioTraps<sup>®</sup> containing mosquito attractants and Pyriproxyfen anti-larval insect growth regulator (IGR) as test traps (EBT) and another 480 number of EcoBioTraps<sup>®</sup> without mosquito attractants and IGR as control traps were deployed in two urban and two rural clusters (240 traps each cluster). Two teams of trained health staff (ASHA workers) weekly monitored the traps over a 28-day period, with follow-ups on 7, 14, 21, and 28 days. Data were collected digitally and analysed.

The analysis of breeding incidence was likely to be 3 times higher (OR = 2.87; 95% CI: 2.50, 3.29) in EBT than control traps ( $P < 0.0001$ ). The difference in breeding instances between EBT and control traps was higher in urban (OR = 4.77; 95% CI: 3.91, 5.82) than rural (OR = 1.78; 95% CI: 1.46, 2.16) area.

The EcoBioTraps<sup>®</sup> is cost-effective, biodegradable, and easy to deploy in both rural and urban areas, proving to be an effective mosquito breeding control tool. The EcoBioTraps<sup>®</sup> primarily attracted gravid *Aedes aegypti* mosquitoes, significantly contributing to breeding reduction as part of source reduction/larval source management. This device also serves as a tool for larval surveillance and can be further studied for its longitudinal impact on vector control and disease burden.

## Cold fogging an effective tool for adult mosquito management

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Over the past 50 years, most vector control programs primarily relied on thermal fogging for adult mosquito management, which was appropriate given the available technologies at the time. However, as engineering and chemistry have advanced, it is crucial to move with the time and break old paradigms. Cold fogging (Water based Fogging) is an effective mosquito control method that utilizes air pressure to convert liquid insecticide into fine, ultra-low-volume droplets. These droplets are then sprayed into the air. Unlike thermal fogging, which heats insecticides to produce vapours, cold fogging does not create vapours and does not affect visibility. It is suitable for both outdoor and indoor environments, effectively killing adult mosquitoes on contact. Additionally, cold fogging machines are user-friendly and provide enhanced effectiveness from chemical pesticides while offering energy savings. This method can reduce fogging operation costs by up to 95% and also offers administrative benefits regarding the management of diesel oil used for fogging. A variety of chemicals have now been approved by the Government of India and the National Center for Vector Borne Diseases Control (NCVBDC). This technology has also been included by the World Health Organization (WHO) in its vector management applications. The NCVBDC program can use this tool as an alternative to thermal fogging for adult mosquito control in both indoor and outdoor settings, thereby supporting ongoing efforts to eliminate vector-borne diseases in India.

## **Studies on the unique features of Risk Factors in the Tribal Malaria-Endemic Regions of NE India: Targeted and tailor-made intervention cocktail for accelerated Malaria control among Jhum Cultivators of Tripura**

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India's goal of malaria elimination by 2030 requires identifying region-specific gaps and locally tailored interventions, particularly in endemic areas like the North-Eastern (NE) states. Although these states account for less than 4% of India's population, they contribute disproportionately to malaria morbidity and mortality, especially among tribal communities. Several districts in this region, where over half the population belongs to Scheduled Tribes (ST), continue to experience high transmission despite an overall decline in malaria incidence (API <1). Micro-level analyses showed that persistent high-case pockets are concentrated in remote, forested tribal areas. Field investigations across Tripura, Mizoram, Meghalaya, and Arunachal Pradesh examined epidemiological, entomological, and drug-resistance factors to identify local drivers of malaria persistence. In Tripura's Dhalai district, the most malaria-endemic area, infections were mainly confined to tribal pockets. High-resolution satellite mapping linked malaria incidence with ecological features such as jhum (slash-and-burn) cultivation, plantations, and forest cover. Integration of land-use, meteorological, and demographic data confirmed a positive correlation between malaria incidence and the extent of jhum cultivation, implicating shifting agriculture practices in disease persistence. Based on these findings, a targeted intervention package was developed and implemented, enhancing vector control, surveillance, and community engagement. Time-series analysis demonstrated reduced slide positivity rates and good community acceptance, validating the feasibility of localized strategies. Entomological studies revealed complex vector ecology, with both primary and secondary *Anopheles* species contributing to transmission, highlighting the need for integrated vector management adapted to local conditions. Drug-resistance monitoring revealed early and widespread resistance across the region. Sequencing of over 100 *Plasmodium* isolates (2014–2024) showed evolving mutation profiles requiring continuous monitoring. Mass surveillance in Dhalai (2014, 2021) detected high levels of asymptomatic, RDT-negative, submicroscopic infections—mostly *P. vivax* and mixed *P. vivax*–*P. falciparum*—indicating hidden reservoirs sustaining transmission. Detection of *P. malariae* and rising *P. vivax* proportions suggest changing species dynamics that must be addressed in elimination efforts. Overall, malaria persistence in NE India is shaped by ecological, vectorial, and parasitological complexities. Evidence-based, site-specific, and community-accepted interventions are essential to accelerate control and help achieve India's 2030 malaria elimination target.

## The Duffy blood groups of Jarawas - the primitive and vanishing tribe of Andaman and Nicobar Islands of India

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Andaman and Nicobar Islands, union territory of India were inhabited by 14 aboriginal tribes. Some of these primitive tribal populations have already become extinct, and the numbers of the existing ones are also dwindling. This group of islands being highly endemic for malaria, it was considered worthwhile to study the hunter-gatherer primitive tribe, Jarawas, for their Duffy blood group phenotype.

The Jarawas, like other tribes of the Negrito race, have short stature, black skin and frizzy hair. They mainly inhabit an area of about 200km<sup>2</sup>, called the Jarawa Reserve Forest, that lies on the Western coasts of South and Middle Andaman. Blood samples of 116 Jarawas were collected and tested for Duffy blood group and malarial parasite infectivity. The Duffy blood grouping was performed as per standard serological techniques, and peripheral smears were screened for malarial parasite and if present parasite density count was performed and the species identified. The results showed a total absence of both Fy(a) and Fy(b) antigens in two areas (Kadamtala and R.K. Nallah) and low prevalence of Fy(a) antigen in another two areas (Jirkatang and Tirur). There was absence of malarial parasite *Plasmodium vivax* infection though *Plasmodium falciparum* infection was present in 27.59% of cases. A very high frequency of Fy (a-b-) in the Jarawa tribe from all the four jungle areas of Andaman Islands along with total absence of *P. vivax* infections suggests the selective advantage offered to Fy (a-b-) individuals against *P. vivax* infection.



## Multistakeholder Partnership for fighting Dengue: An Experience of Equi-Health Alliance

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Vector-borne diseases (VBDs) account for over 17% of global infectious diseases, with dengue emerging as the fastest-growing VBD worldwide. Nearly 40–50% of the global population is at risk, and 50–100 million infections occur annually. India bears over one-third of the global dengue burden, with cases increasing sharply from 28,000 in 2010 to nearly 2,90,000 in 2023, and approximately 2.3 lakh cases recorded in 2024. This surge is driven by climatic changes—warmer temperatures and altered rainfall patterns that extend transmission seasons—as well as ecological pressures such as deforestation, water scarcity, and poor waste management. Unplanned urbanization, overcrowding, inadequate sanitation, and household water storage have created ideal breeding environments for *Aedes* mosquitoes, pushing dengue into rural regions, highlands, and making it a year-round threat. With no effective therapeutics and limited vaccine options, prevention and community engagement remain central to dengue control. The Equi-Health Alliance (EHA), established in 2025, convened regional workshops in Tamil Nadu and Karnataka and a major session at the World Health Summit in New Delhi. These engagements highlighted priorities in prevention and behaviour change, data and surveillance, and financing. Stakeholders emphasized the need for public awareness, integrated surveillance, scaling vaccine development, community-driven solutions, and cross-border data sharing. The World Environment Day Summit on 5 June 2025 further underscored that dengue is a systemic crisis shaped by urbanization, climate change, and migration. A shift is needed from awareness to ownership through localized, inclusive, and gender-responsive communication; school-based interventions; contextual treatment guidelines; and training for frontline workers such as ASHAs. A real-time, AI-enabled surveillance system integrating data from public hospitals, private laboratories, and local authorities is essential for decision-making. Sustainable financing, predictive modelling, and strong public–private partnerships will also be critical. Ultimately, defeating dengue requires coordinated, multisectoral, equity-driven, and climate-resilient action across India.



# ORAL PRESENTATIONS



## Oral Presentation

## Clinical Appearance of malaria and its Epidemiological Burden in Mangalore, India in 2002 to 2022

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Incomplete antimalarial medication, inadequate patient's follow-up on day 7 after first-line antimalarial treatment, especially among migrant labourers and destitute inpatients who escaped from the hospital during intermittent treatment, and patients who did not receive their antimalarial medicines after testing positive for malaria parasites (MP) are significant challenges. These issues contribute to antimalarial resistance, genetic mutations, recurrent malaria, early treatment failure (ETF), and late treatment failure (LTF), hindering efforts to eliminate malaria in India. Need to examine the current malaria burden in India, particular emphasis on *P. vivax* and asymptomatic low-density infections, suggests that existing control tools may be insufficient. Methods: A clinical case study of malaria was conducted at Wenlock District Government Hospital, Mangalore, India, from 2002 to 2022. Suspected malaria cases were confirmed through symptomatic clinical features and microscopic diagnosis of peripheral blood smears. Clinical parameters and a second peripheral blood smear were taken on day 7 to evaluate antimalarial drug response and efficacy. Artificial Intelligence (AI) tools were used to analyse 21 years of clinical malaria cases, including heatmap display correlations between *Plasmodium falciparum*, *Plasmodium vivax*, and mixed infections. Python, Plotly and R Studio were used for effective and interactive visualization of malaria infections status. Findings: Out of 317,210 suspected malaria cases, 77,995 (24.58%) were confirmed as microscopy-positive. Among these, 72.52% were *P. vivax*, 24.78% were *P. falciparum*, and 2.67% were mixed infections. Notably, only 12.48% of treated patients had follow-up on day 7 and 0.77% of patients did not receive medication after testing positive. Hypertension was found in 39.60% of patients, with highest rates in mixed infections (98.32%). Parasitaemia levels were highest in *P. falciparum* (16,68,824/ $\mu$ l). Interpretation: Incomplete treatment and inadequate follow-up are significant issues. Over the 21-year study periods, *P. vivax* infections increased significantly from 53.39% in 2005 to 86.95% in 2022, while *P. falciparum* cases declined from 45.15% in 2005 to 10.81% in 2016, due to substantial progress in malaria control measures. The incidence of mixed infections gradually increased whereas *P. falciparum* cases declined. There is an urgent need for breakthroughs in compulsory treatment for all malaria-positive patients and mandatory follow-up on day 7 to reduce treatment failures and prevent subsequent transmission.

## Oral Presentation

**Emergence of new malaria vectors in the western part of India****Mohanty AK\***

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Goa state has become endemic to malaria ever since the first outbreak occurred in 1986. The incidence of malaria has increased, particularly in the coastal areas, coinciding with the rapid urbanization and construction activities in the state. In the last two decades, Goa has undergone many ecological changes due to increases in population, rapid urbanization, deforestation, and migration of labourers from malaria-endemic states to other parts. With ongoing environmental and demographic changes, new vector-parasite interactions are likely, and new vectors may transmit malaria. Therefore, there is an urgent need to revisit the vector parasite interactions. Our study aimed to determine the susceptibility of non-vector *Anopheline* species to *Plasmodium* infection and their capacity to transmit the parasite. The development of *Plasmodium* infection in the midgut and salivary glands of non-vector *Anopheline* species was assessed using artificial membrane feeding assays. We analysed the expression of mosquito genes involved in parasite interactions and their role in *Plasmodium* development. This study has helped us identify newly evolved mosquito-parasite interactions in Goa, which will be important in understanding the role of non-vector *Anopheline* species in malaria transmission during elimination efforts. This information is essential for planning improved vector control strategies for malaria elimination programs.

## Hidden Malaria Reservoirs: Asymptomatic/Sub-Patent Infections Among Tribal Populations in Hard-to-Reach areas of Odisha– A Silent Barrier to Elimination

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Malaria continues to be a major public health challenge in India, despite significant strides in control and prevention over the years. Odisha, an eastern state of India, covering 4% of the country's land area and 3% population of the country has long been known for its high prevalence of malaria, particularly in rural and underserved areas. The state is also home to 62 tribal and 13 Particularly Vulnerable Tribal Groups (PVTG) who predominantly reside in the hard to reach, forest covered, underserved areas of the state. In 2023, Odisha reported a sharp rise in malaria cases representing about 18.7% of the national total. The present study was conducted in malaria endemic tribal dominated districts of Odisha to find the malaria burden during peak transmission season in 2024. A total of 3512 participants were enrolled in the study. Out of 3512, 50% belonged to Kondha tribe, 5 % were Gond tribe, 43% belonged to Kutia Kondha PVTG community.

Among the participants, 29% tested positive for malaria using Rapid Diagnostic Tests (RDTs)—with 20.1% *P. falciparum* (Pf) positive, 4.45% pan positive, and 13.88% Pf-pan mixed positive. Out of the RDT positives, 85% were asymptomatic. Polymerase Chain Reaction (PCR) results revealed an even higher positivity rate of 34%, comprising 19.3% *P. falciparum* (Pf), 11.7% *P. vivax* (Pv), 0.92% *P. ovale* (Po), and 14.45% mixed infections. Prevalence of sub-patent infection was 24.1%.

The study findings reveal a high prevalence of asymptomatic and sub-patent infection among tribal and PVTG communities. Persistence of sub-patent/asymptomatic infection in the community, remain undetected by conventional tests, may act as malaria reservoirs and left untreated can derail elimination campaigns. Further, coexistence of mixed infections indicates ongoing and complex transmission dynamics in these regions. These findings emphasise the urgent need for sustained malaria surveillance, strengthened vector control strategies, and community-based awareness programs tailored to the specific challenges faced by the tribal and PVTG communities.

## Prevalence of *pfhrp2/3* Gene Deletions in Jagdalpur, Chhattisgarh

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*Plasmodium falciparum* remains the most prevalent species, responsible for over 90% of malaria infections. Antigen-detecting rapid diagnostic tests (RDTs), particularly *hrp2*-based tests, are widely used for confirming *P. falciparum* infections, especially in resource-limited settings. Global reports indicate that deletions in the histidine-rich protein (*pfhrp2/3*) genes compromise the effectiveness of *hrp2*-based malaria RDTs. *P. falciparum* with a deleted *pfhrp2* gene can potentially escape detection using *hrp2*-based tests. This study investigated *pfhrp2/3* deletions from Jagdalpur, Chhattisgarh.

During the study period July 2021 to June 2022 total of 14,550 febrile patients screened and 824 patients were found malaria-positive by microscopy. After informed consent, a total of 306 blood samples were collected. *P. falciparum* infection was confirmed using species-specific nested PCR targeting the 18S ribosomal RNA. DNA quality was assessed with *pfmsp1* and *pfmsp2* primers. Exon 1, Exon 2, and Exon 1-2 regions of *pfhrp2* and *pfhrp3* genes were amplified using specific primers. PCR was also performed for flanking regions of *pfhrp2* (PF3D7\_0831900 upstream and PF3D7\_0831700 downstream) and *pfhrp3* (PF3D7\_1372100 upstream and PF3D7\_1372400 downstream). *P. falciparum* strains 3D7 and Dd2 were used as positive and negative controls, respectively, for *pfhrp2* analysis. The analysis revealed various deletion patterns: Complete *hrp2* and *hrp3* deletions were observed in 26/306 (8.40%) samples, complete *hrp2* deletion in 1/306 (0.33%) samples, complete *hrp3* deletion in 3/306 (0.98%) samples, partial *hrp2* deletion in 3/306 (0.98%) samples, partial *hrp3* deletion in 6/306 (1.96%) samples, and partial *hrp2* and *hrp3* deletions in 1/306 (0.33%) samples.

The findings indicate that 8.40% of samples exhibited complete *hrp2* and *hrp3* deletions, with smaller proportions showing partial deletions. These results underscore the need for systematic surveillance to monitor deletion trends and assess whether the prevalence of deletions has reached the threshold for switching to non-*hrp2* RDTs. These infections complicate transmission control and hinder malaria elimination efforts.



## Forest Malaria in West Bengal: Key Challenges and Feasible Ways for Mitigation

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Forest malaria remains a stubborn obstacle for malaria elimination by 2030. West Bengal Districts, namely - Alipurduar, Jalpaiguri, Purulia, Jhargram & Bankura have large forest covers, sharing also common forest borders with endemic states like Jharkhand and Assam. The state, presently in Category I of Elimination, aims to achieve Category 0 by 2027. With all efforts, when overall malaria incidence is declining, residual foci remain in hard-to-reach areas adjacent to forests. In recent years, about 80% of the case load is reported from these remote villages mostly inhabited by tribes.

The disproportionate case distribution is driven by ecological suitability for vectors, man-mosquito contact in the forest (where people must have to enter for livelihood) and programmatic gaps at those remote places. The same factors also result in persistence of the transmission pockets.

The streams in the forest, slow in non-monsoon seasons, allows vector breeding and thus sustain the transmission of malaria even during the dry summer.

While LLIN or IRS were provided in high endemic villages, the moderate endemic areas were catered with ITBN. Water stagnation pockets at the flanks of the streams were sprayed with bio-larvicide in a supervised manner.

Despite all, malaria transmission couldn't be fully controlled due to vector bites in the outdoor environment. Fever surveys were then geared up to mass malaria survey. In forest villages, asymptomatic malaria is common. Mass surveys detected even the asymptomatic cases and acted as a game changer.

Entomological and epidemiological data, if cross-shared, can be very productive in such situations. They supplement each other with a dual system to identify an early signal – either as a vector alert or as an early surge of cases. However, xeno-monitoring of vectors can reinforce the entomological alerts and help prioritize the vector control measures.

## Deep Learning based Malaria Epidemiology Forecasting in Mizoram, India

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Mizoram state reports about 42% of malaria cases in the Northeast region of India, with unique epidemiological and climatic characteristics shaping its malaria dynamics. Despite substantial progress in malaria control efforts, the state continues to report persistent transmission, influenced by various climatic factors. Understanding the epidemiological profile of malaria in Mizoram through retrospective analysis is essential to unravel the spatiotemporal patterns and underlying determinants driving malaria dynamics in the region. Furthermore, with the increasing impact of climate change on disease transmission, there is an urgent need to develop an efficient climate-based malaria forecasting model. Therefore, in the present study, a detailed analysis of malaria epidemiological profile of Mizoram was performed for the 2010-2023 period and an efficient malaria forecasting model was developed using long short-term memory (LSTM), a deep learning technique. We have employed unidirectional LSTM (Uni-LSTM) and bidirectional LSTM (BiLSTM) neural networks with both univariate and multivariate frameworks to predict the malaria prevalence by leveraging historical malaria data (2010-2019) and climatic variables (temperature, rainfall, relative humidity, soil moisture and NINO3.4). The analysis revealed a fluctuating trend in malaria cases over the study period, with the majority occurring in individuals aged >15 years and a higher prevalence among males. A pronounced seasonal pattern was observed, with peaks during the monsoon months (June-September), highlighting the critical influence of rainfall and humidity on disease transmission. Spatial distribution studies revealed that the high-risk areas of malaria located along the international borders, highlighting the role of cross-border transmission. This study exemplifies that, the multivariate version of Uni-LSTM outperformed with respectable potential for the malaria prediction (RMSE: 0.20 and  $R^2$ :0.81). The proposed multivariate Uni-LSTM has great potential for applications in epidemiology and can be followed by other states of India as well. The outcome of this study can help the public health authorities and policymakers to draw appropriate control measures before the onset of malaria.

## Oral Presentation

**Exploring the susceptibility status of non-vector *Anopheline* species to *Plasmodium* infection under laboratory conditions****Mohanty AK\***

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A mosquito is a competent vector for malaria when it supports the complete sporogony phase of parasite development, from oocyst formation to release of sporozoites that migrate to salivary glands, allowing the transmission of viable sporozoites. Worldwide, 41 *Anopheles* species are competent vectors with varying capacities and transmit malaria. Even though we have understood vector competence and capacity of known vectors, information on new emerging vectors is vital to control/eliminate malaria in many parts of the world. We have to be vigilant of new vectors emerging, consequent to extensive anthropogenic changes in the ecosystem, and specific strategies required for their control. Our study aimed to determine the susceptibility status of non-vector *Anopheline* species to *Plasmodium* infection and their capacity to transmit the parasite. The development of *Plasmodium* infection in the midgut and salivary glands of different non-vector *Anopheline* species was assessed using artificial membrane feeding assays. Additionally, we analyzed the expression of mosquito genes involved in interactions with the parasite and their role in *Plasmodium* development. This study has helped us to identify non-vector *Anopheline* species ability to support complete *Plasmodium* sporogony. However, their potential to transmit malaria in natural settings requires urgent attention, particularly as India aims to eliminate malaria by 2030.

## Self-treatment and Clinical Outcomes of *Plasmodium falciparum* in central India

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Remote tribal areas in central India contribute substantially to the country's malaria burden. In these settings, self-treatment is widespread, often based on nonspecific symptoms and over-the-counter antimalarials. Such practices obscure mixed-species infections and undermine malaria elimination efforts. To assess the clinical and diagnostic impact, we conducted parallel hospital (n = 305 symptomatic patients) and community (n = 432 febrile participants) studies. Generalized linear models were applied to evaluate the relationship between symptoms and disease severity, and diagnostic performance of microscopy, bivalent rapid diagnostic tests (RDTs), and PCR was compared. Patient characteristics and species distribution were similar across cohorts; however, hospital patients presented with more severe disease and stronger, more frequent symptoms. Both microscopy and RDTs failed to reliably detect mixed-species infections, while PCR and RDTs captured a larger proportion of community infections, many of which were sub microscopic due to prior self-treatment. Less symptomatic cases were most often subject to inappropriate self-medication, leading to residual parasitaemia, risk of recrudescence or relapse, reduced sensitivity of microscopy, and underdiagnosis of mixed infections. Inadequate treatment, particularly lack of radical cure, further compounds the problem. To advance towards elimination, malaria control strategies in India must prioritize addressing self-treatment practices and strengthening diagnostics, especially with RDTs capable of detecting less common malaria species.

## Kelch-13 Mutations in *Plasmodium falciparum* from Mayurbhanj, Odisha: Molecular Surveillance and Artemisinin Interaction Analysis

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The global malaria control and elimination program faces significant challenges due to the emergence and spread of *Plasmodium falciparum* strains resistant to antimalarial drugs. Monitoring mutations in the Kelch-13 gene, a crucial marker for artemisinin (ART) resistance, is essential for addressing this growing threat. Recently, a synonymous mutation associated with artemisinin resistance was reported in the neighbouring state of West Bengal. As Mayurbhanj district in Odisha shares its border with West Bengal and is predominantly inhabited by tribal populations, it becomes critical to investigate this region.

This study aimed to establish baseline data on *Pfkelch13* gene mutations in particularly Odisha and eastern India in general. The *kelch-13* gene was amplified using nested PCR and sequenced via Sanger sequencing, with the findings compared to the reference 3D7 clone (*PF3D7\_1343700*). Our analysis revealed no mutations in the Kelch-13 propeller domain, and the Tajima test confirmed the absence of natural selection favoring drug resistance.

Additionally, the study incorporated in silico analyses to assess ART interactions with the Kelch-13 protein crystal structure (PDB ID: 4ZGC). Structural modeling, molecular docking, molecular dynamics (MD) simulations, and binding free energy calculations were performed to examine the interactions between ART and the Kelch-13 propeller domain. The results were compared to those of the apoprotein (KELCH-APO), which does not contain the bound drug. The analyses demonstrated that ART exhibits a high binding affinity for the Kelch-13 propeller domain, underscoring its effectiveness in interacting with this critical protein region associated with drug resistance.

## Oral Presentation

**Malaria control in Mizoram: Importance and Implications in India's malaria elimination efforts****Balabaskaran Nina P\***

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For decades, malaria, predominantly (>80%) *P. falciparum*, has been a major public health problem in Mizoram, especially in its western districts of Mamit, Lunglei, and Lawngtlai, which are hyper-endemic for malaria. In 2023, the annual parasite index (API) in these districts was 34, 24, and 48, respectively. To put this endemicity into context, India's National Vector Borne Disease Control Program categorizes districts with API  $\geq 2$  as level 3—needing intense control. Historically, India's first resistance to antimalarials has originated in the NE region. As the forested terrain makes vector control strategies difficult, effective malaria control in Mizoram is feasible only through parasite control. In addition to the autochthonous emergence of local drug resistance, there is a threat of drug-resistant malaria parasites from Myanmar and Bangladesh entering Mizoram through its porous international borders. Here, we detail Mizoram's malaria epidemiology in the last decade, especially in its western districts, and how malaria control in this region is critical to India's malaria elimination efforts. In addition, the current and future climate change trends and their effect on malaria transmission dynamics will be discussed. Furthermore, the shifting epidemiology towards *P. vivax* infections will also be discussed.

## Oral Presentation

## Influences of Rainfall and Temperature on Malaria Endemicity in Cameroon: Emphasis on Bonaberi District

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Relating the influence of climate on the occurrence of a vector-borne disease like malaria quantitatively is quite challenging. To better understand the disease endemicity, the effects of climate variables on the distribution of malaria in Cameroon are studied over space and time, with emphasis on the Bonaberi district. Meteorological monitoring can lead to proactive control. The government of Cameroon, through the National Control Malaria Program, has put in place strategies to control and stop the spread of the disease. This study is therefore geared towards assessing the yearly parasite ratio of malaria over the ten regions of Cameroon and to work out the influence of rainfall and temperature on disease endemicity with emphasis on a district of Douala. The model used is the VECTRI model, which shows the dynamic link between climatic variables and malaria transmission. The parasite ratio observed and simulated showed a maximum correlation of 0.75 in 2015. A positive relationship between temperature, rainfall and malaria is revealed in this study but Bonaberi has malaria all year round. The West region is the least affected by malaria. We recommend that For the VECTRI model to perform better, the population could be incorporated in the model.

## Implications of Social Factors in Controlling Malaria in Rayagada District of Odsiha

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Rayagada district of Odisha is one of the high malaria burden districts of Odisha for the last two decades. In 2016, the district was the 2<sup>nd</sup> highest malaria burden district in India in terms of Annual Parasite Incidence (API). Although, the API has come down from 64.85 in 2016 to 2.58 in 2022, malaria transmission continues to be persisted despite so many interventions like, Long Lasting Insecticidal Nets (LLIN), Indoor Residual Spray (IRS), Early Diagnosis & Complete Treatment, biological control etc. Social factors like community participation, health seeking behavior, socio-cultural practices, illiteracy, poverty plays a significant role for the outcome of the malaria control programme. Therefore, there is need for better understanding of these social aspects for developing community specific programme interventions.

The study used mixed method approach i.e. analysis and synthesis of malaria epidemiological surveillance data of Rayagada of last 10 years i.e. 2015-2024 along with qualitative surveys like focus group discussion, interview with ASHA, community members, tribal groups and KII with health service providers like Health Workers, Supervisors, Medical Officers. The study analyses the correlation and linkages between various socio-cultural-economic conditions, cultural beliefs and practices, health seeking behavior and acceptance of different malaria control interventions. The study revealed that impoverish housing conditions, dependence on forest livelihood, shifting cultivation, outside sleeping makes the people more vulnerable to mosquito biting. Illiteracy, traditional socio cultural practices, dependence on traditional healers makes the tribal people more risky for malaria infection. Indifferent attitude towards sleeping under LLIN particularly in summer, resistance to IRS and less faith towards formal health service providers have been found as major hindrances. However, it was also found that with strong community mobilization & participation and community having strong connection with front line workers demonstrated good health seeking behavior both in prevention and curative practices.

Malaria control programme in Rayagada requires more than technical factors and interventions. There is a need to think about socio-cultural factors, health seeking behavior, community mobilization, area specific communication strategy, strategy to integrate traditional health care personnel in the malaria control programme to eliminate malaria from high burden tribal districts.



## Oral Presentation

## Mesenchymal Stem Cells alleviate experimental cerebral malaria disease severity by inducing RoRyt<sup>+</sup> Foxp3<sup>+</sup> regulatory T cells and modulating the dysregulated Th17/Treg axis

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Cerebral malaria (CM) is associated with dysregulated immune response against the blood stage of malaria parasite that often leads to serious organ damage ultimately causing fatal pathological complications. Conventional treatments although effective in controlling the parasite often fail to address the severe immunopathology associated with the disease. Herein we investigated the therapeutic potential of Mesenchymal stem cell (MSC) in managing the excess proinflammatory response and maintaining immune homeostasis in PbA infected C57BL/6 mice, an Experimental Cerebral Malaria (ECM) disease model. Parasitemia and survival was monitored regularly along with the neurological complications associated with disease. Immunophenotyping along with programmed cell death of splenocytes was determined via flow cytometry and cytokine levels were analyzed at different time points in serum as well as spleen through bioplex assay and qRT-PCR. It was found that MSC effectively reduced parasitemia, increased survival and decreased hemozoin accumulation in spleens of PbA infected mice along with improving brain pathology by preventing vascular leakage and protecting the Blood Brain Barrier (BBB). MSCs not only rescued the lymphocytes from apoptosis by downregulating PD-1/PD-L1 and ROS levels but also effectively modulated the Th17/Treg imbalance and maintained immune homeostasis by downregulating Interleukin-6 (IL-6) and Interleukin-17 (IL-17) cytokines and upregulating Interleukin-10 (IL-10) cytokine in infected mice. For the first time we reported that MSCs were able to induce a dual phenotype effector Treg cell subset (Tr17) which are known to express both RoRyt and Foxp3 transcription factors and possess high suppressive activity against pathogenic Th17 cells. In conclusion, our findings offer insight into how the infusion of MSCs reduces the severity of experimental CM by modulating Th17/Treg balance and inducing Tr17 effector Treg response against Th17 cells. Thus, MSCs could potentially be used as an adjunct therapy for addressing the immunopathological complications of CM.

## Mapping Malaria: AI, Remote Sensing, and GIS Applications

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Malaria, a significant public health challenge, is caused by *Plasmodium* parasites and transmitted through the bites of *Anopheles* mosquitoes. The disease is targeted for elimination by 2030. Although various tools and techniques are currently employed for its surveillance, monitoring, control, and prevention, recent advancements in artificial intelligence (AI) have brought renewed optimism in malaria management. AI, along with Geographic Information Systems (GIS) and remote sensing, represents a powerful suite of geospatial technologies. These tools enable the prediction, mapping, and management of malaria risk by analyzing environmental factors—such as temperature, vegetation, water bodies, and humidity—that influence mosquito breeding and disease transmission. AI models use satellite and remote sensing data to detect malaria-prone areas and breeding sites, providing real-time alerts to health officials. *GIS-based spatial analyses—such as Getis-Ord Gi hot spot analysis and Moran's I spatial autocorrelation—have been used to map malaria clusters at the block level in high-burden states like Odisha. This targeted approach has led to reduction in malaria cases.*

### Mapping mosquito vector habitats using AI and remote sensing involves:

High-resolution imagery from satellites like Sentinel-2 and Landsat 8, along with drones, to detect land features linked to breeding—such as water bodies, vegetation, and land use.

CNN-based water body detection, which uses deep learning to accurately identify ponds, marshes, and canals, achieving over 90% accuracy.

Habitat suitability models that combine indices like NDVI and NDWI with AI classifiers (e.g., Random Forest, XGBoost) to map breeding risk, even in temporary or hard-to-detect water pools.

## Molecular Insights into ACTs Drug Resistance in *Plasmodium falciparum* Populations of Eastern India

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Odisha, a major contributor to India's malaria case burden, is implementing the NFME 2016-2030 strategy to eradicate malaria by 2030. However, wide coverage of data on antimalarial drug resistance is needed for the proper implementation of antimalarial drug treatment policies. The study aims to assess antimalarial drug resistance in Odisha using molecular markers, focusing on ACTs (ART-SP) and Chloroquine (CQ) to ensure effective antimalarial treatment policies. A total of 239 isolates of *P. falciparum* mono-infection were collected during 2018-2020 from the four different geographical regions of the state. Genomic DNA was extracted from 200 µL of venous blood and amplified using nested polymerase chain reaction. Mutations on the gene associated with CQ, MDR, ART, and SP were detected by PCR followed by Sanger sequencing. The point mutation in *Pfcr*t (K76T) was detected in 2.1%, in *Pfmdr*1 (N86Y) 3.4%, and no mutations were found in *Pfkelch*13 propeller domain. The prevalence of *Pfdhfr*, *Pfdhps* and *Pfhdfr*-*Pfdhps* gene mutations was 50.43%, 47.05%, and 49.79% respectively. The linked quintuple mutation *Pfdhfr* (IRN) *Pfdhps* (GE) responsible for clinical failure (RIII level of resistance) of SP resistance, and the A16V-S108T mutation in *Pfdhfr*, responsible for cycloguanil, was absent. The current study has shown a sharp decline in the prevalence of *Pfcr*t 76T (from 80% in 2008 to 2.1% in 2020) and *Pfmdr*1 86Y (from 85% in 2008 to 3.4% in 2020) after 10 years of CQ withdrawal. Furthermore, no significant increase in the *Pfmdr*1 copy number suggests that *P. falciparum* isolates are not under selection pressure of AQ, MQ and LF. Despite the absence of the mutations in ART, the high prevalence of the point mutations of the partner drug undermines the efficacy of SP, thereby threatening the *P. falciparum* malaria treatment policy. Therefore, continuous molecular and *in vivo* monitoring of ACT efficacy is warranted in Odisha.

Oral Presentation

## How can behavioural studies in vector control programme & Kala-azar Mukht Panchayat initiative contribute towards ending Visceral Leishmaniasis in India?

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Kala-azar is targeted for elimination on an accelerated timeline with Indoor Residual Spray (IRS) as the mainstay in vector control. This technique needs assessment for sandflies and needs to be revised as per the region and adaptation of vectors, as this is not taken into consideration, due to ignorance of vector behaviour changes and can cause huge hindrance in achieving the target for elimination. Community Engagement (CE) for disease control and health has been tested for a long time across the globe for various health programmes. Also inspired by 'TB Mukht Panchayat' (meaning 'TB free village council' in Hindi language) on 24 March 2023, banking on the system of local self-governments in the country, we propose the same strategy for Kala-azar control as "Kala-azar mukht panchayat" as we see this initiative can have huge potential to contribute to achieving targets soon. We critically analyse the vector behaviour changes needed to be incorporated for making a "tailor-made" strategy according to area and suggest a few recommendations for the way forward.

## Oral Presentation

## **Filarial infection among ineligible population(I-Pop) of Mass Drug Administration in an endemic area: Implications for filariasis elimination program in India**

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India is moving towards the Lymphatic Filariasis (LF) elimination goal in 2027. However, the success of LF elimination relies on achieving a participation rate of at least 65% during a mass drug administration (MDA) within the endemic community. Nevertheless, the ineligible population (I-Pop), such as pregnant women, children below 2 years of age, and severely ill persons, will not be covered under the MDA. The prevalence of filarial infection among the I-Pop and the possible risk of contribution of this population as a source of infection is also not known. Therefore, this study was carried out to screen the ineligible population for filarial antigenemia and microfilaremia.

The study was carried out in the LF endemic district, Yadgir in Karnataka State, India. Out of the 235 villages, 36 villages were selected randomly for the blood survey. Family registers used by the community drug distributors were referred to identify the I-Pop. We screened 921 individuals (pregnant women [88], children <2 years [282] and chronic diseases [551]) who belonged to I-Pop. Filarial antigenemia was tested using the Alere Filariasis Test Strip (FTS), and night blood smear was observed in only those individuals who were positive for the circulating filarial antigen. Overall, we observed a high level of filarial infection in the ineligible population in comparison with the recommended thresholds for defining filarial endemicity in terms of antigenemia (2%) and microfilaraemia (1%). Therefore, it is important to screen the ineligible population for filarial infection. The I-Pop may act as a potential source of infection during the post-MDA period, as they will never be treated and will be missed during the MDA. Strategies such as “test and treat” individuals may be useful to prevent the possible resurgence of infection after stopping MDA.

## Oral Presentation

## Effectiveness of Long-Term Mass Drug Administration (MDA) on Antigen and Antibody in the filariasis endemic area of Khurda & Puri, Odisha, India

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Lymphatic filariasis is a major public health problem worldwide. India contributes a larger part of it. Odisha is also sharing its part. The elimination of lymphatic filariasis began in 2004 through the Mass Drug Administration (MDA). The Mf rate, CFA rate, and disease prevalence have been reduced significantly, but they are still awaiting elimination. A cross-sectional study was conducted in Jatni CHC, Khordha district, which is endemic for lymphatic filariasis. The incidence of acute infection in the endemic area of Jatni has decreased significantly, from 13.9% in 1999/2000 to almost zero. However, there is no significant change in the Chronic patients. The rate of active infection has reduced to 2.14% and the percentage of endemic normal individuals has increased to 87.65%. As the target to eradicate the disease by 2015 has not yet been achieved, and as active infection cases (8.3% in 2022) are still prevalent, a few more rounds of MDA will still be required to bring the infection rate to zero. A group of eighty-one individuals participated in the survey in 2000 and 2022. After 22 years, it was found that the number of patients with chronic, acute, and active infections had dramatically reduced. The purposive sample of 81 follow-up individuals showed significant changes in all categories. Among these follow-up individuals, 21 acute patients became 3, and 21 chronic patients became 3 after 17 rounds of MDA. 39 individuals asymptomatic increased to 75 individuals. One individual who was previously infection negative has now tested positive for infection. When we compared these individuals at their filarial-specific antibody (IgG and IgG4) level, it was also significantly reduced over time. A certain amount of IgG antibody was found in the (infection-negative in 2000 and 2022) -/- group, which could be due to mild transmission. That suggests that T cells are producing antibodies even after the infection is cured in filariasis-endemic areas. Puri district has successfully eliminated lymphatic filariasis, as it has cleared TAS-3 according to the WHO guidelines. Instructions are provided to identify microfilaria-positive cases by provoking DEC if any symptoms of filariasis are present. One hundred one patients with acute and/or acute-on-chronic conditions were screened for active infection. We attempted to identify the antigen using the Og4C3 assay kits. However, none of them found an antigen-positive result. The filarial-like symptoms could be a secondary infection or caused by an infected mosquito bite, suggesting that a low and suboptimal transmission of infection is occurring, which can be confirmed by Wolbachia PCR.

## Exploration of Knowledge Attitude and Practice (KAP) regarding Lymphatic Filariasis and its vectors in Bangladesh

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Lymphatic filariasis is a tropical neglected mosquito-borne disease worldwide that affects the lymphatic system of the human body. It disfigures body parts, causes pain, and social stigma, and also affects economic status. 863 million people in 47 countries worldwide remain threatened by lymphatic filariasis. This study was conducted among 100 patients with confirmed LF cases. The Kobo Collect application and face-to-face interviews were done to collect qualitative and quantitative data with semi-structured questionnaires. This study found that 37% of people aged 51–60 years were infected with filariasis in this study area. The peak of elephantiasis onset has been observed in patients aged between 30 and 50 years. Most respondents were below primary education. About 8% of infected patients never heard about filariasis. Most people (72%) were aware that LF is primarily transmitted through mosquito bites. A significant portion of the patients reported (70%) experiencing mental weakness and social stigma (85%) after being affected by Filaria. The right (46%) and left leg (32%) are predominantly affected with 11% of respondents had both leg lymphedema. Moreover, the involvement of the scrotum is evident in the right (6%), left (2%), and both scrotum (1%) cases. Most patients 57% was aware about the MDA program. Village health workers (80%) and community clinics (42%) were the major sources of information regarding LF. A campaign involving village health workers and experienced doctors can provide a proper understanding of filariasis to the people. Data obtained from this study help to conduct specific sensitization campaigns and to provide locally adapted strategies to monitor national lymphatic filariasis control and elimination activities which can be applied as a model to other parts of Bangladesh as well as to the national program of LF worldwide.



## Detection of *W. bancrofti* infection among migrant population from lymphatic filariasis endemic areas – A Molecular xenomonitoring approach

R. Balasubramaniyan\*, Philip Raj Abraham, Ashwani Kumar, S. L Hoti and Manju Rahi

Migrant population from lymphatic filariasis (LF) endemic areas can pose a high risk to naïve areas and/or areas that have cleared the disease/infection. In India, a sizable population from endemic states such as Bihar, Jharkhand, Uttar Pradesh, Madhya Pradesh, Andhra Pradesh, and Odisha, which are highly endemic for LF, migrates to non-endemic or TAS-cleared areas for various purposes, including trade, construction, industry, and education. Such long-term migration and temporary settlement of population can act as a source for local transmission of LF infection, thus adding to the problem or defeating the ongoing elimination efforts. Screening for LF-infected migrants and tracking them is essential to prevent the introduction of new cases and further transmission.

The study was carried out in two different migrant settlement areas of Tamil Nadu (Tirukovilur block) and Puducherry. Night blood smears (NBS) were collected from both these migrant areas and were stained and observed under a microscope for the detection of microfilariae. Mosquitoes were collected from Mf-positive in-migrant areas of Puducherry by using gravid traps, and the collected mosquitoes were segregated and made 72 pools for DNA extraction by using a simple method followed by real-time PCR assay for the detection of *W. bancrofti* infection in vector mosquitoes. In Tamil Nadu, out of 52, none of them were found to be Mf positive in NBS. In Puducherry, out of 209, 5 migrants were detected as microfilariae (Mf) positive in NBS. Mf prevalence rate of 2.4%. In Puducherry, A total of 1800 *Culex quinquefasciatus* comprises of 72 pools (1-25). Out of 72 pools, 3 pools were detected *W. bancrofti* infection in vector mosquitoes.

In Puducherry is endemic to LF and it has successfully cleared the TAS-3; we observed a Mf prevalence rate of 2.4% which is above the critical threshold of <1%. Molecular xenomonitoring (MX) is a highly sensitive and non-invasive method for the detection of filarial infection in vector mosquitoes. Therefore, it is essential to have an active post-MDA surveillance and screening of LF infection among migrants in their settlement areas to avoid resurgence/recrudescence, and it is helpful for sustainable LF elimination and policy decision for the lymphatic filariasis elimination programme.



## Rethinking Arboviral Transmission: Emerging Roles of Secondary Vectors

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Vector-borne diseases remain a major public health challenge, with *Aedes aegypti* traditionally recognised as the primary driver of dengue, chikungunya, and Zika transmission. However, ecological shifts, climate variability, and urbanisation are reshaping vector dynamics, enabling secondary vectors to assume increasingly important roles. *Ae. aegypti* thrives in indoor habitats, ensuring perennial survival in urban landscapes, while *Ae. albopictus*, with its ecological plasticity and global dispersal, dominates outdoor environments and is becoming a co-primary vector in many regions. The recent molecular and ecological investigations on of *Ae. vittatus* in Indian metropolitan settings, Kolkata, after decades of absence, highlights its re-emergence as a potential arbovirus vector.

The study aimed to characterise *Aedes* species diversity, assess their genetic variation using mitochondrial *Cytochrome c oxidase subunit I* (COI) and nuclear *Internal Transcribed Spacer 2* (ITS-2) markers, and examine their seasonal distribution and breeding habitats. Molecular analyses identified 12 haplotypes of *Aedes aegypti*, 16 of *Aedes albopictus*, and 6 of *Aedes vittatus*. Two novel haplotypes in each species were detected for the first time globally, indicating low-to-moderate genetic diversity and evidence of gene flow within local populations. *Ae. vittatus*, a re-emerging species absent in Kolkata since 1997, was observed only during June–August.

Ecological analysis revealed perennial presence of *Ae. aegypti* in both indoor and outdoor habitats, *Ae. albopictus* populations peaking post-monsoon with dominance in outdoor sites, and seasonal detection of *Ae. vittatus* breeding close to human dwellings. This study underscore the dynamic interplay between primary and secondary vectors underscores the fluidity of vectorial hierarchies, challenging conventional surveillance and control programs that remain largely *Ae. aegypti* centric. Understanding the ecological preferences, genetic diversity, and adaptive mechanisms of these three vectors is critical for predicting outbreak patterns and designing integrated strategies that target both indoor and outdoor breeding habitats.

## Oral Presentation

## Detection and Distribution of Dengue Virus Serotypes in Aedes Larvae: A Multi-Location Study from Bhubaneswar, India

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Dengue virus (DENV) is a major vector-borne pathogen of public health concern in India, transmitted primarily by *Aedes* mosquitoes. Monitoring the presence of DENV in *Aedes* larvae provides insights into potential hotspots and risk zones for dengue outbreaks. This study aimed to detect and map the distribution of DENV serotypes in *Aedes* larvae collected from various urban locations in Bhubaneswar, India.

A total of 279 *Aedes* larvae samples were collected from 15 urban locations, including BDA near RMRC, Chandrasekharpur, Khandagiri, Lumbini Vihar, and others. Sampling sites were selected based on high mosquito density and human activity. Larvae were screened for the presence of DENV using molecular techniques, and serotyping was conducted to identify the circulating DENV serotypes. Among the 279 samples, 41 tested positive for DENV, with infections concentrated in four locations. BDA near RMRC reported 13 positive cases out of 129 samples, comprising 9 cases of DENV-1, 1 case of DENV-2, and 4 cases of co-infections with DENV-1 and DENV-2. Chandrasekharpur recorded 7 positive cases among 45 samples, all involving co-infections. In Khandagiri, 18 of 21 samples tested positive for DENV, while Lumbini Vihar reported 3 positive cases out of 5 samples, all of which were co-infections. Samples from the remaining 11 locations, including Patia, Nayapalli, and Vani Vihar, tested negative for DENV.

This study highlights the uneven distribution of DENV serotypes in Bhubaneswar, with significant hotspots identified in BDA near RMRC, Khandagiri, and Chandrasekharpur. The findings underscore the need for enhanced vector surveillance and location-specific interventions to prevent dengue outbreaks. The detection of co-infections further indicates the potential for severe disease outcomes, necessitating comprehensive monitoring and control strategies.

## Oral Presentation

## Circulating Dengue virus serotypes and full-length genome sequencing of DENV-2 in the state of Odisha

**Sabat J<sup>1\*</sup>**, Priyadarshini S<sup>1</sup>, Panda S<sup>1</sup>, Subhadra S<sup>1</sup>, Mohanty S<sup>2</sup>, Pati S<sup>1</sup>, Praharaj I<sup>1</sup>

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Dengue is a major public health challenge in India as multiple outbreaks are reported from different parts of the country every year becoming a major public health challenge. The present study provides the distribution of Dengue serotypes from all 30 districts of Odisha from 2022 to 2024, along with information on the genotype and lineages of Dengue serotype II.

A total of 2899 serum samples suspected of Dengue virus infection from 30 districts of Odisha were subjected to serotype-specific multiplexed real-time RT-PCR assay targeting the CprM gene of 4 different Dengue Virus serotypes. Whole Genome Sequencing was done on MinION MK1C using Oxford Nanopore SQK-RBK114.96 sequencing kit. Sequence merging and read mapping were done by the minimap 2 aligner. Variant calling and consensus sequences were generated using Samtools & Bcftools. Phylogenetic analysis for genotypes and lineage determination was done by the Nextstrain/ Nexclade bioinformatics tool. Serotype DENV-1 was found to be the predominant serotype in 48%, DENV-2 in 31%, DENV-3 in 5.7% and DENV-4 in 0.34% from 2022 to 2024. Prevalence of DENV-1 was detected highest rate in 54% of cases during 2024, circulating in 23 districts, followed by DENV-2 (36%) in 18 districts. DENV-3 was detected in 8 districts in 17.6% of cases in 2022. DENV-4 (n=5) was detected in 2022 from 3 districts only. Co-infection of DENV-1 and DENV-2 was detected in 14.5%. Other Co-infections of DENV-1 and DENV-3 (N=5), DENV-2 and DENV-3 (n=5) were detected in low proportion. Whole Genome Sequencing for 40 samples positive for DENV-2 serotype were detected as genotype II cosmopolitan, and lineages were identified as DENV-2 II\_F.1.1 in 85% of samples and lineage DENV-2 II\_F.1.1.5 in 15% samples.

Odisha experiences multiple outbreaks of Dengue with the circulation of more than one serotype every year. DENV-1 was found to be the predominant serotype, followed by DENV-2 circulating during 2022-2024. Genotype II (cosmopolitan) of Serotype-2 was the common genotype with the predominant lineage DENV-2 II\_F.1.1 in 85% of cases. This information helps to understand the epidemiology of circulating Dengue serotypes and genotypes, which may have implications for predicting large-scale outbreaks in the region.

## Oral Presentation

**Arboviral vectors harboring *Wolbachia*: A glimpse towards Dengue control****Mohanty I<sup>\*1</sup>, Hazra RK<sup>1</sup>**<sup>1</sup>Regional Medical Research Centre, Bhubaneswar-751023, India.

*Wolbachia*, a gram-negative obligate endosymbiont, is renowned for its reproductive manipulation in mosquitoes. Studying the presence of natural *Wolbachia* in mosquitoes is crucial for understanding its impact on inhibiting arboviral replication. This study focuses on the prevalence, diversity, infection frequencies, phylogeny, and density of indigenous *Wolbachia* strains in wild mosquito species from Odisha. Our findings confirm *Wolbachia*'s presence in *Ae. albopictus*, *Cx. quinquefasciatus*, *Cx. vishnui*, *Cx. gelidus*, *Ar. subalbatus*, *Mn. uniformis*, and *Mn. indiana*. *Wolbachia* strains were categorised into two supergroups (A and B). *Ae. albopictus*, the primary vector of dengue and chikungunya, exhibited both super-infection and mono-infection. The ovaries of *Ae. albopictus* showed the highest density of *Wolbachia* compared to the midguts or salivary glands. The densities of *wAlBA* and *wAlbB* varied in G1 mosquitoes based on sex and age. We observed that *Wolbachia* superinfection increased in females, while *wAlBA* density decreased in older males compared to *wAlbB*. Giemsa-stained squashed ovaries revealed pink pleomorphic *Wolbachia* cells with various shapes and forms. This study is designed to cover the major aspects of *Wolbachia* and its potential as a biocontrol agent in arboviral outbreaks. Understanding the potential of indigenous *Wolbachia* strains and their interactions with viruses could further help reduce the global burden of vector-borne diseases.

## Oral Presentation

**“Whole genome sequencing of DENV-2 serotype circulating in Odisha 2024”****Priyadarsini S<sup>1\*</sup>**, Sabat J<sup>1</sup>, Panda S<sup>1</sup>, Subhadra S<sup>1</sup>, Pati S<sup>1</sup>, Praharaj I<sup>1</sup>

I. ICMR- Regional Medical Research Centre, Bhubaneswar

Dengue virus (DENV), a significant mosquito-borne pathogen, poses a major public health challenge worldwide. Out of its four serotypes (DENV1-4), DENV-2 has been associated with severe outbreaks. This study aims to better understand the molecular epidemiology of the circulating DENV-2 serotype in Odisha and to manage dengue outbreaks more effectively.

A total of 23 DENV-2 positive samples, confirmed by serotype-specific multiplex RT-PCR, were included in the study. The GTnRich kit for Dengue Virus was used to generate amplicons for whole genome sequencing. cDNA synthesis was performed using LunaScript RT SuperMix. Each sample underwent two multiplex PCR reactions using six primers divided into two pools (Pool A and Pool B), each containing three amplicons to minimise interference from overlapping regions. Amplified PCR products were combined for library preparation using the Oxford Nanopore SQK-RBK114.96 (Rapid) kit. The library preparation process included steps such as barcode addition, bead cleanup with AMPure XP beads, quantification using a Qubit 4 fluorometer, and adapter ligation. The prepared library was then loaded onto a flow cell (FLO-MIN114) for sequencing. Base calling and demultiplexing were performed, and FAST5 FASTAQ files were extracted from the MinION Mk1C. The FASTAQ pass barcode files with the referred DENV-2 genome were analysed using Commander-NGS software on a Linux system. These files were merged, and reads were mapped to the DENV-2 reference sequence obtained from Nextstrain, using the minimap2 aligner. Variant calling and consensus sequence generation were done using the Samtools & Bcftools pipeline, and the final consensus FASTA files were analysed by Nextstrain – Nextclade for phylogenetic analysis.

Out of the 23 samples, 22 were identified as DENV-2 genotype II, while one sample could not be sequenced due to poor quality. From these 22 samples, 18 samples were DENV-2 II\_F.1.1 and 4 samples were DENV-2 II\_F.1.1.5 lineages. 9 samples had genome coverage of more than 60%.

This study provides valuable insights into the molecular epidemiology of circulating DENV-2 strains in Odisha. The majority of the samples were identified as DENV-2 genotype II, with the predominant lineage being DENV-2 II\_F.1.1. Continued surveillance and further genomic analysis are essential to track the evolution and provide information for public health interventions and vaccine development.

## Inter-relation between Dengue, dengue vector and climatic factor in Kolkata, India

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Dengue is the fastest-spreading mosquito-borne arboviral disease in the world, with an estimated 390 million infections annually. India accounts for nearly 33 million clinically confirmed cases each year, making it the third-most affected country globally. Within India, West Bengal has reported the highest dengue burden from 2007 to 2022. The primary vectors, *Aedes aegypti* and *Aedes albopictus*, also transmit chikungunya, Zika and other arboviruses. Kolkata, one of India's most densely populated cities, experiences frequent dengue outbreaks, making it a critical location for understanding micro-level transmission dynamics.

The present study examined the spatial distribution of dengue cases in Kolkata in 2022, correlating them with entomological indicators, breeding habitats, and climatic as well as micro-environmental factors. Dengue case data were collected from the Kolkata Municipal Corporation (KMC) and supplemented with longitudinal entomological surveys conducted in seven wards. Temperature, rainfall, humidity, and local environmental conditions such as construction sites, vacant land, garbage dumping areas, and slum settlements were recorded. Statistical analyses were performed using Excel, SPSS, ANOVA, and linear regression, with Origin software used for visualisation.

Results showed a highly uneven distribution of dengue across Kolkata. Four southern wards—109, 106, 101 and 99—recorded more than 10 cases per 10,000 population, whereas many northern wards had fewer than 1 case per 10,000. Of the 42,942 premises and 126,284 water-holding containers surveyed, 2.55% of premises and 1.06% of containers were positive for *Aedes* breeding. Plastic containers were the most common breeding site (55.7%), followed by discarded materials. *Aedes aegypti* was mainly found in domestic and peri-domestic habitats and showed the strongest correlation with rainfall at zero-month lag, while *Aedes albopictus* was mostly outdoors and correlated with rainfall at a one-month lag. Dengue cases peaked in September, one month after peak rainfall and vector density in August, indicating a strong lag-phase association.

Dengue incidence was positively linked to under-construction buildings and vacant land used as garbage dumps, but not to slum areas. The findings suggest that localised micro-environmental conditions, especially man-made breeding sites, play a greater role in dengue transmission than broader city-wide climatic factors. The presence of *Ae. aegypti* during the dry season and *Ae. albopictus* proliferation in the monsoon collectively increase post-monsoon transmission risk.

The study highlights the need for targeted pre-monsoon vector control, improved waste management, and removal of artificial breeding habitats. Such evidence-based insights can guide more effective dengue control strategies in urban India.

## Integrating Sero-Xenomonitoring into Dengue Surveillance: Early Detection of DENV Serotypes in Urban Odisha

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Dengue fever remains one of the most significant arboviral diseases globally, with an estimated 390 million infections and around 96 million clinical cases reported annually across over 125 countries. In India, dengue has become hyperendemic with co-circulation of multiple serotypes and recurring seasonal outbreaks, posing a major public health burden in urban and peri-urban settings. Odisha, a state with rapid urban expansion and climatic variability, continues to report sporadic dengue upsurges, particularly in Bhubaneswar city, despite ongoing vector control interventions.

This study employed a sero-surveillance-based xenomonitoring approach to detect dengue virus (DENV) in *Aedes* mosquitoes during the dry, pre-monsoon period in dengue-endemic wards of the Bhubaneswar Municipal Corporation (BMC), Odisha. A total of 44 pools of adult female *Aedes* mosquitoes were collected from four wards during January to May 2025, using standard entomological techniques. RNA was extracted from homogenised pools and screened by real-time reverse transcription (RT-PCR) using a multiplex DENV detection and serotyping kit. Twenty pools (45.5%) tested positive for DENV RNA, indicating silent viral circulation before the onset of monsoon. All four DENV serotypes (1-4) were detected, with DENV-2 and DENV-3 being predominant.

These findings provide the first molecular evidence of pre-monsoon dengue virus circulation in *Aedes* populations from Odisha, suggesting the existence of early transmission foci. The study underscores the potential of xenomonitoring as an early-warning surveillance tool, facilitating timely interventions and predictive modelling for dengue outbreaks. Integrating vector molecular surveillance with climatic and epidemiological data can significantly strengthen urban dengue preparedness and response frameworks.



## Oral Presentation

## Molecular surveillance and epidemiology of Dengue, Chikungunya and Zika from clinical samples presenting acute febrile illnesses in Bengaluru, India

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Dengue, Chikungunya, and Zika are arboviral diseases and have remained significant public health concerns in India. These diseases are transmitted by the *Aedes aegypti* and *Aedes albopictus* mosquitoes, which thrive in tropical and subtropical climates. Early detection of these viruses is challenging due to overlapping symptoms with other viral, parasitic, and bacterial infections. Clinicians usually rely on ELISA and rapid tests for detection, which are non-confirmatory. Nucleic acid-based diagnosis, although confirmatory, is not easily available for early and cost-effective implementation in low-resource settings, leading to an underestimation of the actual disease burden.

In this molecular surveillance study, we collaborated with Anand Diagnostic Laboratory in Bengaluru to understand the prevalence of arboviral illnesses such as Dengue, Chikungunya, and Zika in clinical samples presenting acute febrile illnesses. We screened 200 blood samples suspected of Dengue and Chikungunya, extracted total nucleic acid, and assessed the prevalence of Dengue, Chikungunya, and Zika, with an in-house developed, multiplex q RT- PCR based molecular assay.

We observed 54/ 200 (27 %) positivity for Dengue and 25/200 (12.5%) positivity for Chikungunya, and 9/200 (4.5%) positivity for Zika. We observed significant differences in the Prevalence of Dengue ( $\chi^2= 123.44$ ,  $df =3$ ;  $P<0.001$ ), Chikungunya (119.54,  $df =3$ ;  $P<0.01$ ), and Zika ( $\chi^2= 105.67$ ,  $df =3$ ;  $P<0.001$ ), respectively. We also observed co-infections between Dengue and Chikungunya (7%), and with Dengue and Zika (2.5%). We observed concordance in the findings upon comparing the in-house multiplex qRT-PCR assays with the commercially available individual kits for early detection of Dengue, Chikungunya, and Zika, respectively.

Molecular surveillance is the need of the hour for early diagnosis and timely clinical decisions. The study findings shall aid in understanding the actual disease incidence and prevalence and in creating appropriate diagnostic algorithms and protocols to accurately and early diagnose and mitigate Dengue, Chikungunya, and Zika in low-resource settings.



## Oral Presentation

**Development and implementation of a strategy for early diagnosis and management of scrub typhus: an emerging public health threat****Jain HK<sup>1\*</sup>**, Das A<sup>1</sup>, Dixit S<sup>1</sup>, Kaur H<sup>2</sup>, Pati S<sup>3</sup>, Ranjit MR<sup>1</sup>, Dutta A<sup>4</sup>, Bal M<sup>1</sup><sup>1</sup>Regional Medical Research Centre, Bhubaneswar, India<sup>2</sup>Indian Council of Medical Research, New Delhi, India<sup>3</sup>Regional Medical Research Centre, Bhubaneswar, India<sup>4</sup>Public Health Foundation of India, Indian Institute of Public Health, Bhubaneswar, India

Scrub typhus, caused by *Orientia tsutsugamushi*, is a re-emerging zoonotic disease in the tropics with considerable morbidity and mortality rates. This disease, which is mostly prevalent in rural areas, remains underdiagnosed and underreported because of the low index of suspicion and non-specific clinical presentation. Limited access to healthcare, diagnostics, and treatment in rural settings further makes it challenging to distinguish it from other febrile illnesses. While easily treatable, improper treatment leads to severe forms of the disease and even death. As there is no existing public health program to address scrub typhus in India, there is an urgent need to design a program and test its effectiveness for the control and management of the disease. Scrub typhus to be included as a differential diagnosis of fever cases in every tier of the public health system, Screening and diagnosis among presumptive scrub typhus cases to be enhanced, and Complete treatment of diagnosed scrub typhus cases to be ensured. Desk review, secondary data analysis and formative research were carried out for assessing the knowledge, attitude, practices and preparedness of the health system for implementation of the developed intervention. Based on the findings, an intervention package was developed for capacity building and health system strengthening for establishing diagnosis at the peripheral level of the health care system. IEC BCC activities were done for Community awareness. An evaluation survey was done post-implementation to assess the effectiveness of the developed strategy. A total of 8216 suspected individuals at the OPD of selected hospitals were screened for Scrub typhus, and more than 18% tested positive for the same. No mortality was reported because of early diagnosis among the positive cases. Knowledge among the community increased by 79%. Many healthcare staff's capacity building was done regarding disease diagnosis and management. A robust referral system, capacity building of health personnel and community awareness can help in reducing severe morbidity and mortality caused by undiagnosed fever.

## Dynamic host T cells miRNA profile during early hepatic stages of *Schistosoma japonicum* infection

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CD4<sup>+</sup> T cells play a critical role in response to *Schistosoma* infections, and accumulated data have indicated that miRNAs tightly regulate T cell activity. However, miRNA profiles in host T cells associated with *Schistosoma* infection remain poorly characterised. Therefore, we undertook the study and systematically characterised T cell miRNA profiles from the livers and blood of *S. japonicum*-infected C57BL/6J mice at 14- and 21 days post-infection. We observed 508 and 504 miRNAs, in which 264 miRNAs were co-detected in T cells isolated from blood and livers, respectively. The comparative analysis of T cell miRNAs showed that *miR-486b-5p/3p* expression was significantly downregulated and linked to various T cell immune responses, and *miR-375-5p* was highly upregulated, associated with Wnt signalling and pluripotency, Delta notch signalling pathways, etc. Whereas hepatic T cells showed *miR-466b-3p*, *miR-486b-3p*, *miR-1969*, and *miR-375* were differentially expressed compared to the uninfected control. In addition, bioinformatics analysis combined with RT-qPCR validation of selected targets associated with the immune system and parasite-caused infectious disease showed a significant increase in the expression of *Ctla4*, *Atg5*, *Hgf*, *Vcl* and *Arpc4* and a decreased expression of *Fermt3*, *Pik3r1*, *Myd88*, *Nfkbie*, *Ppp1r12a*, *Ppp3r1*, *Nfyb*, *Atg12*, *Ube2n*, *Tyrobp*, *Cxcr4* and *Tollip*. The in vitro study of isolated T cells and EL-4 cells stimulated with *S. japonicum* worm antigen (SWA) showed corroborated miRNA expression as detected by RNA-seq and in vivo validation. Overall, these results unveil the comprehensive repertoire of T cell miRNAs during *S. japonicum* infection, suggesting that the circulatory (blood) and liver systems have distinct miRNA landscapes that may be important for regulating T cell immune response. Altogether, our findings indicated a dynamic expression pattern of T cell miRNAs during the hepatic schistosomula stages of *S. japonicum* infection.

## Oral Presentation

**Duplex PCR For Simultaneous Detection of *Babesia gibsoni* and *Babesia vogeli* in canines****Manaswini Dehuri\***, Anita Dora, BN Mohanty, A.Hembram

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Canine babesiosis is a tick-borne disease caused by *Babesia* spp., which infects and destroys healthy erythrocytes, leading to mortality and morbidity in dogs. The diagnosis of babesiosis is tedious and time-consuming, especially in latent and chronic infections. In India, the two most common *Babesia* spp. are *Babesia vogeli* and *Babesia gibsoni*. Duplex PCR for simultaneous detection of *Babesia vogeli* and *Babesia gibsoni* was performed using the two primers (BAB1 F/ BAB4 R and BAG1 F / BAG1 R) at the annealing temperature of 54°C. Reaction mixture for polymerase chain reaction was prepared and was set in a final volume of 25 µl with 0.50 µl of each primer and 2 µl of template DNA. The samples which were used in the PCR assay were similar samples which was found positive for *Babesia vogeli* and *Babesia gibsoni* by single plex PCR assay. The results of duplex PCR confirmed that amplification of genomic DNA of *Babesia vogeli* and *Babesia gibsoni* yielded a PCR product of 600bp and 488 bp, respectively. Our findings suggested that Duplex PCR is a rapid, accurate, cost-effective method for *Babesia* spp. detection that is feasibly applicable for diagnosis.

## Molecular Detection of Bovine Trypanosomosis and Anaplasmosis in and Around Bhubaneswar

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The agroecological and geo-climatic conditions of India are highly favourable for the growth and multiplication of vectors like ticks and flies, which act as natural vectors of anaplasmosis and trypanosomosis. These diseases are of zoonotic importance. Blood parasites have a detrimental impact on the global bovine population, significantly hindering livestock productivity and health. This study focused on assessing the prevalence of certain bovine hemoparasites, their molecular characterisation, and associated risk factors in and around Bhubaneswar, Eastern India. A total of 106 bovine blood samples from varying breeds, ages, and sexes presented to the Veterinary Clinical Complex were examined using microscopy (Giemsa-stained thin blood smears) and polymerase chain reaction (PCR). DNA was extracted through conventional methods, with primers targeting the msp5 gene (382 bp) for *Anaplasma marginale* and VSG RoTat 1.2 gene (110 bp) for *Trypanosoma evansi*. Sequencing and phylogenetic analyses were performed on representative PCR products. Blood smear analysis revealed an overall prevalence of 21.7% (23/106), with *A. marginale*, *T. evansi*, and mixed infections showing rates of 10.38%, 3.77%, and 7.55%, respectively. All microscopy-positive samples were also confirmed by PCR, which indicated an overall prevalence of 35.8% (*A. marginale*: 17.92%, *T. evansi*: 5.66%, and concurrent infections: 12.26%). Higher prevalence was observed in cattle aged 2–4 years (16.9%), females (29.2%), and crossbred Jersey cattle (20.75%). Representative PCR products were sequenced and assigned to GenBank (OL550058: *A. marginale*; OL550059: *T. evansi*). Disease mapping for these parasites is limited in many regions of India. This study is likely the first to report molecular characterisation of *T. evansi* and *A. marginale* from this area, shedding light on potential genetic diversity among local isolates.

## Entomo-molecular surveillance of breeding sites reveals habitat-specific adaptive strategies of *Aedes* spp. in Odisha, India

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Dengue epidemiology in India is intricately linked to the ecological and molecular adaptability of *Aedes* mosquitoes. Vector adaptation to diverse aquatic microhabitats strongly influences their survival, development, and transmission potential. Understanding how environmental parameters modulate genetic and physiological adaptation in *Aedes* spp. is essential for developing sustainable and habitat-specific vector control strategies in Odisha.

This study integrated entomological field surveillance with physicochemical characterization of breeding habitats and developmental gene expression profiling to elucidate habitat-specific adaptive mechanisms of *Aedes* populations across Odisha.

*Aedes* larvae collected from diverse breeding sites in the Khordha district of Odisha were reared under controlled laboratory conditions, followed by bionomic assessments to evaluate developmental traits and habitat-specific associations. Physicochemical attributes (temperature, pH, TDS, salinity, and conductivity) of source breeding sites were analyzed. Expression of developmental genes (*Hsp70*, *Hsp83*, *EcR*, *JHEH*) was quantified using qRT-PCR, and molecular interaction networks were mapped through STRING analysis. Bionomic assessment identified discarded tyres (DT) (58.81%) and plastic receptacles (PR) (14.11%) as predominant breeding habitats, followed by cement tanks (CT) (8.87%) and earthen pots (EP) (7.85%) contributing to immature *Aedes* abundance. Larvae from DT and PR exhibited shorter developmental duration, higher pupal emergence, and greater fecundity. Physicochemical characterization showed moderately alkaline pH, elevated TDS, and variable salinity and conductivity, indicating nutrient-rich yet stress-prone environments. Gene expression profiling revealed significant upregulation of *Hsp70*, *Hsp83*, and *EcR*, along with downregulation of *JHEH* in larvae from DT and PR habitats. STRING analysis identified *Hsp83* as a key regulatory hub linking proteostasis, hormonal signaling, and detoxification pathways.

The entomo-molecular approach demonstrates that habitat-specific physicochemical variability drives developmental and molecular adaptations in *Aedes* spp., enhancing adaptive resilience and vectorial efficiency. Integrating molecular biomarkers into vector surveillance can improve predictive modeling and support sustainable, breeding site-targeted dengue control strategies in Odisha.

## Oral Presentation

## Exploring the Frontiers of Mosquito Research: A Journey through TIGS Insectary

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The TIGS insectary represents a distinctive biosafety level facility within the infectious disease domain, specifically tailored to support research endeavours centered around mosquitoes. This state-of-the-art establishment adheres rigorously to regulatory protocols, featuring a controlled environment to ensure precise conditions.

Its operational focus encompasses three key areas:

Insectary operations spanning three distinct genera of mosquito species, and the facility specializes in housing diverse mosquito lines, providing a critical resource for experiments ranging from behaviour studies to genetic investigations. This specialized area ensures a comprehensive understanding of mosquito vectors.

A well-established insect transformation facility facilitates embryonic and adult biomanipulation for knock-out and knock-down studies, and this approach opens new avenues for diverse studies in entomology.

A *Plasmodium* culture facility is instrumental in conducting various drug screenings and membrane-feeding assays to assess the inhibitory properties of different antibodies. This enables in-depth studies on parasite biology, drug testing, and the development of potential vaccines.

The Insectary Facility emerges as a basis in the pursuit of cutting-edge research on vector-borne diseases. Mosquitoes, as primary vectors, demand a dedicated facility that addresses the unique challenges and opportunities in understanding their biology and interactions with pathogens. Our goal is to collaborate with researchers and provide them with the necessary support for their mosquito-related work, including access to our facility and resources. Our platform also serves as a resource center for researchers to enhance their understanding and address various challenges related to mosquito-borne diseases.

## Cresol derivatives from *Bacillus subtilis* as natural oviposition modulator of *Culex quinquefasciatus*: a molecular docking approach

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Mosquitoes rely heavily on olfactory cues for locating suitable oviposition sites, with microbial communities in aquatic habitats playing a crucial role in producing volatile organic compounds (VOCs) that influence mosquito behaviour. In this study, we isolated *Bacillus subtilis* DHB13 from the breeding habitat of *Culex quinquefasciatus*, a major vector of several human diseases. The partial 16S rRNA gene sequence of the isolate has been submitted to NCBI GenBank with the accession number PV698100. The identity and resistance profile of the strain was confirmed through biochemical and antibiotic susceptibility tests. The bacterial suspension demonstrated a notable oviposition activity index (OAI) of 0.77, suggesting a significant attraction to gravid female mosquitoes. LC-MS analysis of the bacterial culture supernatant revealed the presence of three cresol derivatives: diisopropyl-m-cresol, 3-ethyl-p-cresol, and 6-ethyl-o-cresol. These compounds were evaluated through molecular docking against Cx. *quinquefasciatus* Odorant Binding Protein 1 (CxOBP1), a protein known to mediate olfactory-driven oviposition behaviour. Molecular docking revealed strong binding of CxOBP1 with diisopropyl-m-cresol (-6.7 kcal/mol), 3-ethyl-p-cresol (-6.2 kcal/mol), and 6-ethyl-o-cresol (-5.9 kcal/mol), indicating potential oviposition attractant activity. All three ligands were found to bind within a conserved binding pocket of CxOBP1, behavioural assays confirmed the oviposition-stimulant properties of the bacterial suspension, indicating that the detected compounds mimic natural semiochemicals such as p-cresol, previously recognized as an oviposition cue. These findings reinforce the role of microbiota in shaping mosquito reproductive behaviour through the production of volatile attractants. Moreover, they highlight the potential of using microbial VOCs as environmentally sustainable tools for mosquito surveillance and vector control. This integrative approach linking microbial ecology, chemical analysis, and mosquito behaviour provides novel insights for the development of attractant-based control strategies.

## Oral Presentation

**Ecotype-Associated Microbiome Profiling of *Aedes aegypti* and *Culex quinquefasciatus* Mosquitoes in Odisha, India****Barik T K\***, Sahu B, Nayak B, Pattanayak A K, Murmu L K

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Mosquito-borne diseases are among the primary health issues in almost all tropical and subtropical countries, including India. Surroundings of mosquitoes, particularly their breeding site, have a major effect on the structure of their microbiome. The present study was conducted to investigate the relationships between the mosquito bacterial microbiota and different ecotypes in the Ganjam district of Odisha, India. Microbiome profiles of midgut and salivary glands of the important mosquito vectors, *Aedes aegypti* and *Culex quinquefasciatus* collected from five different ecotypes namely plain, coastal, forest, hilly, and riverine were compared using full-length 16S rRNA next-generation sequencing (NGS). *Pseudomonadota* and *Gammaproteobacteria* were the predominant bacterial phylum and class in the midgut and salivary glands of both species, respectively. Significant differences existed between bacterial communities in the midgut and salivary glands, as well as between ecotypes in various taxonomic hierarchies of both species. Both the midgut and salivary glands of *Aedes aegypti* were dominated by the genera *Pseudomonas* and *Bradyrhizobium*. The predominant taxa in the salivary gland of *Culex quinquefasciatus* were *Enterobacter* and *Pseudomonas*, while *Wolbachia* dominated the midgut. These findings contribute to our knowledge of the microbiota of *Ae. aegypti* and *Cx. quinquefasciatus* mosquito vectors, emphasizing the key role of ecotype in shaping larval microbial communities and providing a foundation for further research on mosquito ecology and their management strategies.



## Characterization of Peak biting times of *Aedes aegypti* and *Aedes albopictus* using the human baited double net trap technique in the Colombo District

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The human landing catch (HLC) technique is widely regarded as the most effective method for monitoring the biting activity of *Aedes* mosquitoes. However its use is ethically unacceptable due to its high risk of human exposure to pathogens. In this study we used an alternative method, The Human- Baited Double Net Trap (HDN), to determine temporal biting activity of *Aedes aegypti* and *Aedes albopictus* the primary vectors of Dengue and Chikungunya

The study was conducted from July 2021 to July 2023 (across 12 replicates) in Bopeththa Grama Niladhari Divison (GND), located in the Gothatuwa Medical Officer of Health (MOH) area in the Colombo District. Mosquitoes were collected using human baited double net traps (HND) laced one indoor and one outdoor location on haphazardly selected premises. A single bait was used in each trap. Hourly collections were performed from 5.00 a.m.-7.00 p.m. using a mouth aspirator by a single trained collector. Temperature and Humidity were recorded during the study period. The average proportion of mosquitoes collected each hour was compared to identify peak biting times of the two species. Comparison of indoor and outdoor all data were analyzed using statistic software.

A total of 121 (79 indoor and 42 outdoor) *Ae aegypti* and 104 (13 indoor and 91 outdoor) *Ae albopictus* were collected during the study. There were two biting peaks coinciding for the two species. The morning peak was during 6-10 a.m. (total of 36 *Ae aegypti* and 44 *Ae albopictus*) and the evening peak was during 3-7 p.m. (total of 61 *Ae. aegypti* and 74 *Ae. albopictus*)

With mosquiti activity peaking in the pre-sunset hours, *Ae. aegypti* and *Ae. albopictus* catches of HDN should be performed in the hours before dark. Indoor premises are considered to be the most suitable biting place for *Ae. aegypti* mosquitoes and also outdoor premises suitable biting place for *Ae.alboictus* therefore routine *Aedes* vector surveillance and monitoring need to be streamlined for the space spraying. Essential thing both indoor and outdoor should be needed.

## Spinosad bio-larvicide: field evaluation for mosquito control in Bengaluru City, India

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Mosquito-borne diseases remain a significant public health challenge in India, particularly in rapidly urbanizing cities. With widespread insecticide resistance undermining traditional tools, novel and eco-friendly alternatives are urgently needed. Spinosad, a naturally derived bio-larvicide from *Saccharopolyspora spinosa*, offers a safer and environmentally sustainable alternative. This study evaluated the field performance and persistence of three formulations of Spinosad against key mosquito vectors.

A large-scale multi-season (monsoon, winter, and summer) field trial was conducted. WHO-guided Phase III field evaluation of three Spinosad formulations—Dispersible Tablet (DT, 7.48%), Emulsifiable Concentrate (EC, 20.6%), and Granular (G30, 2.5%)—against *Aedes aegypti* and *Anopheles stephensi*, while EC (2.5%) and G30 were tested against *Culex quinquefasciatus* in diverse urban habitats of Bengaluru, India. Reapplication was performed when larval densities in treated habitats reached control levels.

All Spinosad formulations induced rapid larval mortality, leading to a statistically significant reduction in larval density. The duration of residual efficacy varied by species, habitat, and formulation. For *Aedes aegypti*, DT remained effective for 56 days, G30 for 35 days, and EC for 28 days. For *An. stephensi*, DT and EC showed 70 and 53 days of residual activity, respectively. For *Culex quinquefasciatus*, suppression persisted for 63 days (EC, cement tanks) and 35 days (EC/G30, drains). No adverse effects were observed on non-target organisms and community acceptance surveys indicated a high willingness to support future applications.

Our findings confirm Spinosad exhibited potent larvicidal action at relatively low concentrations and maintained activity longer than conventional larvicides. Its strong bioefficacy, coupled with strong community acceptability and an eco-friendly profile, positions Spinosad as a valuable addition to integrated vector management strategies. In the era of escalating insecticide resistance, deploying Spinosad could substantially contribute to sustainable mosquito control and advance progress towards vector-borne disease control and elimination.

## Insecticide susceptibility of *Phlebotomus argentipes* and residual activity of alpha-cypermethrin sprayed on local substrates in visceral leishmaniasis endemic states of India

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Visceral leishmaniasis (VL), a parasitic disease caused by *Leishmania donovani*, is transmitted in India by the sand fly *Phlebotomus argentipes*. Although endemic in several countries, VL remains a public health concern in India, where 385 cases were reported in 2024. Despite this low count, fluctuations in case numbers remain possible due to various contributing factors. Indoor residual spraying (IRS) is the primary vector control strategy against *Ph. argentipes*. After widespread resistance to dichlorodiphenyltrichloroethane (DDT), in 2016 India replaced it with IRS of a pyrethroid, alpha-cypermethrin, for VL control. While earlier studies assessed sand fly susceptibility to insecticides at the WHO-discriminatory concentrations (DC), this study is the first to evaluate the residual efficacy of alpha-cypermethrin 5% wettable powder (WP) on different local substrates in a concurrent, multi-site investigation in VL-endemic areas of India. Conducted during 2023–2024 by three institutes of the Indian Council of Medical Research (ICMR) in the states of Bihar, Jharkhand, and Uttar Pradesh, the study assessed the susceptibility of wild *Ph. argentipes* in WHO tube tests with filter papers impregnated with DCs of DDT (4.0%), malathion (5.0%), alpha-cypermethrin (0.05%), and deltamethrin (0.05%) (prescribed for testing mosquito susceptibility), as well as alpha-cypermethrin (0.1%) (for sand flies). The residual efficacy of alpha-cypermethrin WP applied at the target dose of 25 mg/m<sup>2</sup> was tested on four common indoor substrates: cement concrete, limewashed cement, mud, and wood. *Ph. argentipes* showed resistance to DDT 4.0% but remained susceptible to malathion, deltamethrin, and alpha-cypermethrin at both mosquito and sand fly DCs. Residual efficacy of alpha-cypermethrin declined below 80% one month post-spray on cement concrete and limewashed cement substrates, and after three months on mud. On wood, mortality remained at 100% for four months post-spray.

Sand flies in endemic Indian states remain susceptible to key pyrethroids. Given that rural homes feature mud-plastered walls and wooden structures as preferred resting sites for *Ph. argentipes*, and considering the 3–5-month VL transmission period, one to two rounds of alpha-cypermethrin IRS, each lasting 3–4 months, could effectively interrupt VL transmission.

## Oral Presentation

## Phytogenic Fabrication and Comparative Bioefficacy of ZnO, CuO and ZnO–CuO Nanocomposites against *Aedes aegypti*: A Nano-Enabled Vector Management Approach

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The present investigation delineates the phyto-mediated, eco-benign synthesis of zinc oxide nanoparticles (Am-ZnO-NPs), copper oxide nanoparticles (Am-CuO-NPs), and zinc oxide–copper oxide nanocomposites (Am-NCs) employing aqueous leaf extract of *Aegle marmelos*, followed by comprehensive physicochemical characterization and bioefficacy evaluation against *Aedes aegypti*. Am-ZnO-NPs were fabricated via a combustion-mediated thermochemical approach under optimized reaction parameters (5% extract concentration, 70 °C, 60 min contact time, pH 10), Am-CuO-NPs via a green co-precipitation route, and Am-NCs through a biogenic hybrid nanofabrication strategy integrating ZnO and CuO crystalline phases. High-resolution characterization confirmed nanometric dimensions (~37.44 nm), phase purity, and defined crystallographic structures including hexagonal wurtzite (ZnO) and face-centered cubic (CuO) and with substantial colloidal. Biotoxicological assays revealed that Am-NCs exhibited superior entomotoxic potency, attaining minimal LC<sub>50</sub> values of 5.429 µg mL<sup>-1</sup> (larvae) and 5.695 µg mL<sup>-1</sup> (pupae), surpassing the efficacy of Am-CuO-NPs (6.497, 8.641 µg mL<sup>-1</sup>) and Am-ZnO-NPs (86.67% larval and 56.00% pupal mortality at 10 µg mL<sup>-1</sup>). Enzymatic inhibition assays and histopathological analyses elucidated pronounced suppression of acetylcholinesterase, α- and β-carboxylesterases, concomitant depletion of total soluble proteins, and profound midgut epithelial disintegration which is the indicative of neurotoxic and metabolic impairment, with maximal physiological disruption observed in Am-NC-treated cohorts. Collectively, the findings reveal Am-NCs as the most potent mosquitocidal modality within the studied series, demonstrating pronounced biocidal efficiency, enzymatic pathway interference, and tissue-level cytotoxic pathology. This investigation underscores the potential of *A. marmelos* derived nanostructures as sustainable, multifunctional nano-interventions for integrated vector management and pathogen control, aligning with global imperatives for green nanotechnology driven disease mitigation strategies.

## Development of Species-Specific Microbial Lures for the Targeted Control of *Aedes* Mosquitoes

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Mosquito-borne arboviral diseases including dengue, chikungunya, and Zika remain major public health and economic burdens in tropical and subtropical regions, with India bearing a substantial share of the global morbidity. Traditional vector control strategies are increasingly constrained by widespread insecticide resistance, ecological concerns, and limited community compliance, necessitating the development of novel, sustainable interventions.

This study explores a microbiome-based, species-specific approach to vector control by developing microbial lures derived from human skin-associated microbiota. These lures exploit volatile organic compounds (VOCs) emitted by commensal skin bacteria, which are known to modulate the host-seeking behaviour of vector mosquitoes such as *Aedes aegypti*.

Using a multidisciplinary methodology encompassing microbial culturing, gas chromatography-mass spectrometry (GC-MS) analysis, and behavioural olfactometry assays, this study aims to identify and characterise key VOC profiles that serve as attractants to *Aedes* species. The goal is to formulate ecologically safe, species-selective, and economically viable lures that can be integrated into existing vector surveillance and control systems.

This approach aligns with the principles of Integrated Vector Management and the One Health framework, offering a potentially transformative solution for vector surveillance and “attract-and-kill” traps that could significantly reduce vector densities and lower disease transmission, contributing to a reduction in dengue-associated morbidity and mortality.

## Superabsorbent composite for *Aedes* larval reduction in breeding sites

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Dengue virus (DENV) infection poses a significant public health challenge in tropical and subtropical regions of India. In the absence of dengue vaccines and antiviral drugs, integrated vector management (IVM) strategies are necessary to control the spread of DENV. Furthermore, green technologies, in addition to chemical insecticides, are needed for vector control. As *Aedes* larvae development depends on breeding site water characteristics, altering these characteristics using superabsorbent composite could reduce *Aedes* larval breeding.

This study aimed to develop an organic superabsorbent composite for vector management and reduction of *Aedes* larval breeding sites.

A composite was developed using acid extraction and characterized using XRD and SEM-EDS to evaluate structural and surface properties. Application dose determination for efficient water absorption and retention time was assessed at different temperatures and pH, followed by in-house evaluation using water samples collected from *Aedes* breeding sites.

The composite exhibited crystalline phases of sodium chloride (NaCl), iron oxides (Fe<sub>2</sub>O<sub>3</sub>), and silicon oxide (SiO<sub>2</sub>), which indicates a combination of both organic and inorganic matrix. This crystalline and amorphous combination suggests the composite is hygroscopic and reactive, making it suitable for applications. Further, elemental composition of the composite revealed carbon (32.64 %), oxygen (38.58 %), and sodium (28.78 %) as major elements. High carbon and oxygen content highlight an organic framework, while silica and sodium provide stability and desiccation potential. Water absorption and retention analyses confirmed the composite's efficiency (100%) across varying temperatures and pH. Field evaluations further validated its ability to alter breeding site water characteristics, reducing *Aedes* larval breeding.

The developed organic superabsorbent composite effectively altered *Aedes* breeding site water characteristics, demonstrating potential for sustainable vector management. High water absorption and retention properties of the composite make it an efficient approach for reducing *Aedes* larval breeding in dengue outbreaks and other areas.

## Oral Presentation

## Assessment of Household Pesticide Use Practices and Public Awareness in Urban and Rural area of Pimpri-Chinchwad: Implications for Vector Control

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Vector-borne diseases such as dengue, malaria, chikungunya, and Zika virus continue to pose serious public health challenges globally, accounting for over 17% of infectious diseases and causing approximately 700,000 deaths annually. In urban environments, household pesticide products are widely used to control pests like mosquitoes, cockroaches, and rodents. However, improper use and limited awareness for health and environmental risks associated with these chemicals remain major concerns. Present cross-sectional study was conducted in the urban & rural regions of Pimpri-Chinchwad area over two phases (July–November 2024 and June–August 2025), involving 791 residents from diverse localities. Environmental observations showed stagnant water in 95% of areas, indicating poor sanitation and ideal breeding conditions for vectors. This study highlights crucial concerns regarding household pesticide use and vector-borne diseases in urban and rural areas of Pimpri-Chinchwad. A structured questionnaire was prepared to assess knowledge, attitudes, and practices (KAP) related to household pesticide use, pest awareness, application methods, and storage behaviors. Findings of the present study revealed that mosquitoes were the most reported household pest (95%), followed by cockroaches (78%) and rats (67%). Pesticide use was nearly universal (97%), with insecticide sprays being the preferred method (90%). Despite high usage, only 34% of respondents read manufacturer instructions, and 71% applied pesticides at night. The study highlights significant gaps in safe handling practices and public awareness underscoring crucial concerns regarding household pesticide use and vector-borne diseases in urban and rural study areas. The findings emphasize the need for educational interventions and the promotion of Integrated Pest Management (IPM) strategies, which can combine with chemical, biological, and environmental approaches for sustainable pest control to reduce health risk and contribute to effective urban vector control efforts.



## Oral Presentation

## Transcriptome Analysis of *Haemophysalis flava* using Illumina Hi-Seq 4000 sequencing: De novo assembly, functional annotation and SSR marker analysis

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Hard Ticks plays a critical role in Transmitting various tick-borne diseases (TBDs) posing significant global threats to human and animal health. They are ectoparasites and directly damages the hosts, transmitting the vector borne diseases. Ticks as a vector for Crimean-Congo hemorrhagic fever, Severe fever thrombocytopenia syndrome virus SFTSV and Tick-borne encephalitis (Virus), Lyme disease, Relapsing fever, Rickettsia diseases and *Tularemia* (Bacteria). The tick, *Haemophysalis flava* is considered to be public health concern due to their role in transmitting tick borne pathogens. The detection of Severe fever thrombocytopenia syndrome virus (SFTSV) and tick born encephalitis virus (TBEV) genome fragments from *H. flava* distributed China, Japan and South Korea. So *H. flava* is the vector and reservoir of SFTSV and TBEV in South Korea. In this context a genome-level understanding of tick biology explores the molecular resources of host-pathogen interaction. The Transcriptome analysis of *Haemophysalia flava* using Illumina Hi-Seq 4000 sequencing studies of functional annotation and molecular resources annotated against public databases. The locally curated protostome databases (PANM-DB) was used to screen the putative adaptation related transcripts classified as gene families, such as angiotension converting enzymes, aquaporin, adenylyate cyclase, AMP-activated protein kinase, glutamate receptors, heat shock proteins. The simple sequence repeats (SSR) were screened using unigenes using Repeat Masker (v4.0.6) and MISA (v1.0) software tools, followed by the designing of SSRs flanking primers using Batch primer 3 (v1.0) software. The transcriptomes produced a total of 69,822 unigenes of which 46,175 annotated to the homologous proteins in the PANM-DB. KOG, KEGG and GO were also annotated from unigenes. The presence of protein kinase, Zinc-finger (C2H2-type), reverse transcriptase, and RNA recognition motif domains were also observed in unigenes. A total of 3480 SSRs were screened of which, 1907 and 1274 were found as Tri and Di nucleotide repeats respectively. The transcripts obtained and the SSR markers screened from the *H. flava* transcriptome will be essential to understanding-the molecular biology, biochemistry and biological evolution of this tick from the perspective of the tick as a vector. Additionally, this is the first large-scale screening of SSRs from the coding transcripts of *H. flava* that includes the identified primers for genetic studies.



## Oral Presentation

## Emergence of *Anopheles stephensi* and Evolving Vector Resistance: Implications for Malaria Elimination in Koraput, Odisha

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Malaria continues to pose a significant public health challenge in the tribal and forested areas of Koraput district, Odisha, despite the sustained implementation of core vector control interventions. The present study undertook comprehensive entomological surveillance, insecticide susceptibility testing, and assessments of community knowledge, attitudes, and practices (KAP) across selected sites in the district. Entomological investigations revealed the presence of *Anopheles stephensi* in Koraput for the first time, alongside the predominant vectors *An. culicifacies* and *An. fluviatilis*. Both *An. culicifacies* and *An. stephensi* populations exhibited resistance to DDT, malathion, and pyrethroids, whereas *An. fluviatilis* remained fully susceptible. Socio behavioural assessments indicated widespread outdoor sleeping habits and suboptimal use of long lasting insecticidal nets (LLINs), particularly in remote and high endemic localities. These behaviours significantly enhance human vector contact and sustain malaria transmission. The findings highlight that conventional, indoor focused interventions alone are insufficient to interrupt transmission in ecologically complex and behaviourally dynamic tribal settings. Achieving and sustaining malaria elimination in Koraput will require the integrated approach that combines targeted outdoor vector control strategies, strengthened entomological and resistance surveillance, and community driven behavioural change interventions. Addressing these multifaceted challenges is essential to accelerate progress toward India's goal of malaria elimination by 2030.



# POSTER PRESENTATIONS



## ***Plasmodium falciparum* histidine-rich protein 2 (*pfhrp2*) gene deletions and the impact of malaria camp interventions in Odisha, India**

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The persistence of *Plasmodium falciparum* (*P. falciparum*) malaria in India remains a major public health concern ahead of the country's national framework for malaria elimination (NFME) goal-2030, despite a global decline in the malaria cases and several successful in- country malaria elimination programs in place. The rapid diagnostic test kits (RDTs) based on PfHRP2 (*P. falciparum* Histidine Rich Protein2) are the mainstay of malaria diagnosis, however, recent reports confirm the proportion of false-negative PfHRP2-RDTs are on the rise, owing to the deletion in the parasite *pfhrp2* gene. The prevalence of *pfhrp2* gene deletions in 15 malaria endemic villages was investigated as part of the effectiveness trial of the malaria camps in Odisha and their impact on the interventions. Findings suggested 61.6% of subpatent *P. falciparum* infections had *pfhrp2* deletions (majority in exon 2 locus, and in febrile samples) along with identification of novel amino acid repeat motifs (231-293 amino acids) in subpatent samples without deletions (intact *pfhrp2*-Exon 2) markedly differing from those of RDT+/PCR+ samples, probably associated with suboptimal production of PfHRP2 antigen levels in the subpatent samples. Such anomalous observations (novel repeat motifs) should be further investigated in the context of *de novo* gene deletions over time, as undiagnosed *P. falciparum* infections harboring the *pfhrp2* gene deletion can act as a potential reservoir for malaria transmission. In addition, our modeling studies suggested that mass screening and treatment (MSAT) using PfHRP2-based RDTs alongside additional interventions (e.g., long-lasting insecticidal net distribution, indoor residual spraying, intensified IEC etc.) indicated lower relative risk of *P. falciparum* infections with or without *pfhrp2* gene deletions, when compared with the use of MSAT by PfHRP2-based RDTs alone. These findings collectively highlight the rationale for deploying alternative diagnostic targets/biomarkers for effective implementation of ongoing malaria elimination programs.

## Residual Malaria Transmission among PVTG Communities in HBHI Districts of Odisha: Entomological Evidence from the DAMaN Evaluation

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After two decades of remarkable progress, the global malaria trend has entered a phase of epidemiological resilience and uneven gains. While some regions move closer to elimination, others continue to face resurgence transmission driven by vector adaptability and ecological complexity. India has achieved an over 80% decline in malaria cases and 90% reduction in deaths since 2015, advancing steadily toward elimination by 2030. Yet, residual transmission persists in forested and tribal regions of Odisha, particularly in High-Burden High-Impact (HBHI) and Particularly Vulnerable Tribal Group (PVTG) areas, where ecological diversity sustains malaria receptivity. A cross-sectional entomological survey was conducted in 2024 across 40 tribal villages in Rayagada and Kalahandi districts, both of which are HBHI zones under the Durgama Anchalare Malaria Nirakaran (DAMaN) initiative by the Odisha state government. Adult mosquitoes were collected using mouth aspirators from human dwellings and cattle sheds, following standard guidelines. Morphological identification using the standard key was supported by molecular confirmation. Vector susceptibility was assessed using WHO tube bioassays and LLIN cone bioassays. A total of 3,296 adult *Anopheles* were collected, representing nine vector and eight non-vector species. *An. fluviatilis* predominated across all sites, especially in Bissamcuttack (PMHD = 9.58) and Chandrapur (PMHD = 5.59), indicating strong adaptation to hilly, forested PVTG habitats. *An. culicifacies* and *An. subpictus* were abundant in Lanjigarh (PMHD = 7.03), suggesting overlapping vector niches. Minor vectors such as *An. minimus* and *An. jeyporiensis* reflected ecological heterogeneity. Most species showed indoor resting and human-feeding behaviours. WHO bioassays showed 100% mortality to Alphacypermethrin and Deltamethrin, and LLIN cone tests confirmed full efficacy. This first entomological and insecticide resistance study in DAMaN areas provides valuable baseline evidence for Odisha's malaria control programme. The findings confirm the continued effectiveness of pyrethroid-based interventions and highlight the need to maintain strong surveillance, regular insecticide resistance monitoring, and timely replacement of LLINs. Strengthening zonal-level entomological capacity and integrating these data into program planning will support sustained malaria elimination efforts in tribal and HBHI districts of Odisha.

## Resurgence of *Plasmodium vivax* in Kandhamal, Odisha a Sickle Cell Endemic District

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Malaria is one of the major public health problems in Odisha. Among the four species of human malaria parasites, *Plasmodium falciparum* has been the dominant species, followed by *Plasmodium vivax*. This study was conducted in Kandhamal of Odisha, which is a Sickle cell endemic district. We obtained an interesting finding while conducting this study on sickle cell patients. During the follow-up of 65 sickle cell disease patients over the past three years, only one patient was found to have malaria twice — both the times caused by *Plasmodium falciparum*. Following which we analysed the incidence of malaria over the last eight years in the same area, we found that cases increased from 2018 to 2019, followed by a sharp decline up to 2021. However, malaria cases have risen again, with a noticeable increase in the incidence of *Plasmodium vivax* alongside *Plasmodium falciparum*. This sudden rise in malaria cases is a significant concern from a public health perspective. This study suggests that, although patients with Sickle Cell are at lower risk of malaria than persons without Sickle Cell, protection is not complete, and malaria is an important factor that needs to be considered.

## Global perspectives on insecticide resistance in Malaria vectors: Evidence from India and Brazil

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Malaria remains a significant public health threat in endemic regions of India and Brazil. However, the emergence of insecticide resistance poses a major challenge to sustaining malaria elimination efforts. This combined review synthesizes data from India (2017-2024) and Brazil (2021-2024) on phenotypic insecticide susceptibility in primary malaria vectors, including *Anopheles culicifacies* and *An. stephensi* in India and *An. darlingi* in Brazil. Data were gathered from peer-reviewed publications that examined vector species and insecticides tested, including dichloro-diphenyl-trichloroethane, malathion, deltamethrin, alpha-cypermethrin, permethrin, and etofenprox. The findings revealed widespread resistance, particularly to pyrethroids and DDT, with varying intensities across different regions and vector populations. In India, low-to-moderate resistance levels have been observed in *An. culicifacies* populations from several states, with evidence of emerging multi-insecticide resistance in the species. In Brazil, the first large-scale monitoring revealed resistance to deltamethrin and permethrin in most of the *An. darlingi* populations, except for a few locations, highlighting heterogeneous resistance profiles. These findings underscore the urgent need for continuous, large-scale insecticide resistance monitoring, proactive resistance management strategies, and the introduction of new-generation vector control tools to sustain malaria control and elimination goals.



## Prevalence of malaria parasite infection among the migrants in the western coastal part of India

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Malaria remains a significant public health concern in Goa, India. The state has been endemic since the first outbreak, which occurred in 1986; since then, cases have been consistently reported from urban and peri-urban areas of the state. Although the state has reported zero indigenous cases in 2024 and is approaching malaria elimination, the threat of migration associated malaria in the state remains a serious concern.

A study was conducted among symptomatic migrant labourers to determine the prevalence of *Plasmodium* species infection using microscopy and rapid diagnostic tests. The blood grouping and haemoglobin levels of the malaria patients were determined and the socio-demographic data were collected to understand the contributions of these factors to the disease transmission in this region.

In this study, *Plasmodium vivax* (61.4%) was the predominant species compared to *Plasmodium falciparum* (34.55%), and mixed infection accounted for 3.9% of the total samples collected. Males represented nearly 89.7% of reported cases with a mean age of  $29.3 \pm 13.09$ . The blood group and haemoglobin (Hb) levels of the malaria patients were determined, revealing a predominance of the B+ blood group and a mean Hb concentration of  $12.75 \pm 3.14$  g/dL. Majority of the migrant patients originating from 12 Indian states, particularly Bihar (35.7%), West Bengal (15.32%), Uttar Pradesh (15.3%), Jharkhand (11.67%), Odisha (3.6%), Karnataka (3.6%), Madhya Pradesh (3.6%), Maharashtra (2.1%), Rajasthan (2.1%), Haryana (2.1%), Assam (1.45%) and Chhattisgarh (0.7%). Almost 63% of the patients were employed as labourers on the construction sites. Common symptoms include fever, chills, headache, vomiting, and fatigue.

The predominance of *P. vivax* and the concentration of cases among migrant labourers highlight the need for continuous active surveillance, early detection, and prompt treatment of migrant workers. Sustaining ongoing parasite and vector control strategies is crucial for maintaining malaria-free status in Goa.

## Lot Testing to Improve the Quality of Malaria Rapid Diagnostic Tests in India

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Since 2010, malaria rapid diagnostic tests (RDTs) are widely used to detect malaria. The Indian Council of Medical Research-National Institute of Malaria Research performed lot testing (LT) according to WHO procedures since 2016. Lot testing is performed to evaluate the lot-to-lot variation in performance of malaria RDTs. We receive RDTs from different manufacturing companies and RDTs procured through various agencies such as National and state programmes, Central Medical Service Society etc. The storage temperature of RDTs, kit components and handling in transportations impact the results of RDT. Therefore, quality assurance is required before it reaches end-users. WHO recognized lot testing laboratory facility assure the quality of RDTs as the high level of quality plays a significant influence, particularly in regions with persistent low parasitemia. Four sets of positive quality control (QC) panels for *P. falciparum* (*Pf*) and *P. vivax* (*Pv*) and 10 negative panels tested RDTs, Since Jan 2022, A total 381 lots containing 22,098 RDT kits for malaria diagnosis were subjected to Lot Testing at NIMR. Of the 381 lots analyzed, 27 lots containing 1,566 RDTs were deferred. The QC panel for *Pf* revealed a faint Pan band in the tested lots, which is a cause for concern. The results of deferred lots were reported to concerned government agencies. Quality-compromised RDTs may lead to an incorrect diagnosis. It is critical to have a QC system in place for effective malaria management. Regular and systematic lot testing builds trust among healthcare workers and patients, encouraging wider use of RDTs, especially in rural and underserved areas and the success of lot testing underscores the need for mandatory quality assurance protocols for all diagnostic tools used in public health programs in India.

## Prevalence of malaria and associated socio-economic factors among pregnant women of hard-to-reach areas of Odisha, Eastern India: a cross-sectional study

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Asymptomatic malaria infection during pregnancy is a major public health concern that poses significant risks to pregnant women, represents a major obstacle to malaria elimination programme. This study aimed to evaluate the prevalence of asymptomatic malaria and associated socio-demographic factors among pregnant women in different districts of the state of Odisha, India. A cross-sectional hospital-based study was conducted in 14 districts from the state during the year 2018 to 2020. Using the convenience sampling method, 336 pregnant women were enrolled. The RDT & nested PCR method was employed to identify the *Plasmodium* genus and species. MS-excel was used to entered data before being uploaded to SPSS v20 for bivariable and multivariable logistic regression analysis to find the associated factors (P-value < 0.005 highly significant). Of the total 336 pregnant women who participated in this study, N=9 (2.7%) and N=21 (6.3%) were confirmed to be infected with *Plasmodium* species by nested PCR. Asymptomatic malaria during pregnancy was found to be significantly associated with not using an insecticide-treated bed net [(P-value = 0.000, AOR: .068, 95% CI [.020, 0.237], not having ITN [(P-value =0.000, AOR: 0.027, 95% CI [0.005, 0.158] using IRS spray P-value= 0.324, AOR: 0.455, 95 %CI [0.095, 2. 177] and pregnancy outcome secondigravidae P-value=0.333, AOR: 0.557 95 %CI [0.171, 1.819] and multigravidae P-value=0.796, AOR: 1.144, 95 %CI [0.413, 3.165]. The current study revealed that asymptomatic malaria is prevalent among pregnant women, with factors such as lack of insecticide-treated bed nets, absence of IRS spraying, and pregnancy outcomes contributing to the issue. Therefore, promoting the use of insecticide-treated bed nets, implementing IRS spraying, and providing health education could play crucial roles in preventing asymptomatic malaria among pregnant women in the study area.

## Differences in the malaria vector densities and their insecticide susceptibility status in a subset of villages with malaria camp interventions in Odisha, India

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The differences between the major malaria vector density and their insecticide susceptibility status were investigated in a subset of villages (n=12) in the Keonjhar district of Odisha, India where 'Malaria Camp (MC) intervention' (i.e. multi-component malaria elimination drive in inaccessible villages as part of the DAMaN program in Odisha), were assessed via a cluster assigned quasi- experimental effectiveness trial (study type 3) by the Center for the Study of Complex Malaria in India. Briefly, five villages as Arm-A (New MC), five villages as Arm-B (delayed MC) and two villages as Arm-C (Old or continued MC) were selected for the malaria vector surveillance. Though an overall inconclusive trend in the total mosquito density was revealed, the total Anopheline vector density increased in human dwellings (58-75% from 42-61%) and in cattle sheds (70-71% from 48- 68%) across all study arms; indicating statistically insignificant impact of the new or delayed interventions (n=10). However, the proportions of the major malaria vectors *An. fluviatilis* and *An. culicifacies* declined (8% from 18%; and 13% from 25% respectively) only in Arm-C villages (n=2) and specific to human dwellings - suggesting subtle impact of multiple rounds of malaria camps. *An. fluviatilis* and *An. culicifacies* exhibited possible insecticide resistance with respective observed mortalities of 97-98% and 54-90% in Arm-A and B. In Arm-C, while *An. fluviatilis* showed susceptibility to DDT and Deltamethrin, possibly due to the effects of LLIN usage and continued IRS measures, in stark contrast, *An. culicifacies* presented complete resistance to DDT and Malathion and possible resistance to Deltamethrin across all study arms. In conclusion, our investigation suggest that multiple rounds of MC interventions could be essential for effective outcomes on malaria vector control. Large scale effectiveness trial in multiple geographical areas is warranted to translate such pilot observations to reflect the real impact on the vector population.

## Insight into the genetic diversity of *Plasmodium vivax* isolates circulating in Goa, India

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Goa state is endemic for both the species of *Plasmodium* i.e. *Plasmodium vivax* and *Plasmodium falciparum*. *Plasmodium vivax* exhibits genetic diversity, existing in VK210 and VK247 subtypes. The present study was carried out to understand the prevalence of *Plasmodium vivax* subtypes circulating in Goa state.

*Plasmodium vivax* patients were recruited at the Goa Medical College and the blood sample was collected followed by the informed consent of the patient. Samples were analyzed using PCR-RFLP by targeting the CSP gene. Further, the patient gametocytaemia and parasitaemia were compared and Genetic diversity was analysed using appropriate software.

PCR-RFLP analysis of 110 samples revealed that 93.6% were VK210, 3.6% were VK247 and 2.7% found mixed infections of VK210 and VK247. Among VK210-infected patients, parasitaemia levels ranged from 0.039% to 1.7%, while VK247-infected patients exhibited parasitaemia levels between 0.545% to 1.346%. The study also observed fluctuations in VK210 case numbers, peaking in August (22 cases) and July (16 cases), with fewer cases recorded in October, November, and December (2-3 cases). Furthermore, parasite metrics displayed seasonal variation, with elevated average parasitemia noted in March, April, and August, along with higher average gametocytemia levels in May and June. Several polymorphic characteristics of the VK210 variant were observed.

The predominant *Plasmodium vivax* subtype circulating in Goa is VK210. The Genetic diversity and seasonal dynamics of *P.vivax* VK210 highlights the importance of targeted intervention strategies and surveillance efforts in malaria control initiatives.

## Development of gene-based diagnostic method for detection of neglected malaria species: *Plasmodium knowlesi* and *Plasmodium cynomolgi*

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Zoonotic malaria is an emerging public health threat, particularly in regions with human-primate interactions. The imminent risk of zoonoses of non-human malaria parasites is not far from reality in India, as has been observed in the case of *Plasmodium knowlesi* (*Pk*), and so is possible with *P. cynomolgi* (*Pc*), already reported in Southeast Asia. Microscopy, often used for malaria diagnosis, is time-consuming and less efficient at sub microscopic parasitaemia, we propose to develop a diagnostic method for detecting non-human Plasmodium.

We conducted *in silico* analyses to identify a suitable target gene for the diagnosis of non-human malaria parasites. We have designed Primers for this gene and evaluated using bioinformatics tools, followed by PCR amplification. Cross-reactivity was assessed against other malaria parasites, and multiplexing was performed to enhance detection. The assay will undergo sequencing and serial dilution to evaluate sensitivity and specificity. Validation will be carried out using a large set of samples, and the results will be compared with reported diagnostic methods for non-human malaria parasites.

We have performed experiments and check to ensure that primers are amplifying the desired target sequence efficiently and specifically. We have checked cross reactivity of our primer with different species of malaria parasite namely *Plasmodium falciparum* (*Pf*), *Plasmodium vivax* (*Pv*), and *Plasmodium knowlesi* (*Pk*).

Non-human Plasmodium parasites (*Pk*, *Pc*, *P. fieldi*, *P. inui*) have been reported in Malaysia, with *Pk* and *Pc* also found in India. *Pk* has been detected in human hosts and vectors across Indian states. The proposed diagnostic tool will aid in detecting, identifying, and monitoring *Pk* and *Pc*, contributing to malaria control and elimination by targeting reservoirs.

## Emerging Threat of *Plasmodium Vivax* in Mizoram

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Mizoram is a malaria hyperendemic state with *Plasmodium falciparum* being the predominant species. However, the state is experiencing a paradigm shift in malaria epidemiology with *Plasmodium vivax* emerging as a major contributor to the malaria burden. This study aims to investigate the epidemiological trends and control challenges associated with the emergence of *P. vivax* in Mizoram. Malaria surveillance data for the past few years (2020-2025) were collected from the National Center for Vector-Borne Diseases Control (NCVBDC), Government of India, and the Integrated Disease Surveillance Programme (IDSP) of the Government of Mizoram. This study employed a retrospective data analysis to investigate the epidemiology of *P. vivax* in Mizoram. The study findings indicate a significant shift in malaria epidemiology in Mizoram, with *P. vivax* accounting for over 47.83% of reported cases in 2025 (up to June), as compared to 16.57% in 2020. Alarming, the Mamit district in Mizoram has seen a steep increase in *P. vivax* cases, from 15.77% to 71.20% in 2020 and 2025 (as of June), respectively. This could be due to the emergence of drug-resistant *P. vivax* parasites. Furthermore, the increase in *P. vivax* poses additional challenges due to its capacity for relapse from dormant liver stages, complicating disease management. Treatment is often limited by the risk of hemolysis in individuals with G6PD deficiency, thereby posing a significant challenge to malaria control efforts. This study underscores the need for targeted interventions, including enhanced surveillance, improved diagnostic capabilities, and effective treatment strategies. Further research is needed to understand the dynamics of *P. vivax* transmission in Mizoram and inform evidence-based policy decisions.

## **Integrated Metabolomics and Network Pharmacology to decipher the latent Mechanisms of green synthesized silver nanoparticles against Malaria**

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Despite intensive attempts to eliminate malaria, drug and insecticide resistance has rendered the disease a serious public health concern. Thus, it is necessary to explore novel mechanisms of action for future antimalarials that are not impacted by current drug resistance. The sea algae *Sargassum tenerrimum*, which has several biological uses and contains bioactive chemicals that help with the environmentally friendly synthesis of silver nanoparticles, is used in the present study. Metabolomics is used to determine the metabolic changes in the Ag-ST-treated and infected mice models. Network pharmacology and molecular modelling studies confirmed Ag-ST's interactions with the target protein. According to the metabolomics study, arachidonic acid is connected to Ag-ST's therapeutic effects. The network pharmacology and molecular modelling studies exemplified that PTGS2 is the primary target for Ag-ST nanoparticles. The study suggests that the Ag-ST can decrease malaria-induced inflammation by modifying arachidonic acid pathways.



## Understanding malaria transmission patterns through multi-level surveillance data in Meghalaya, Northeast India

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India has achieved significant reductions in the malaria burden in recent years. However, considerable heterogeneity persists in different regions; for instance, the South Garo Hills district in Meghalaya remained under an intensified control phase (API > 2). The study aimed to identify emerging hotspots of malaria across blocks and sub-centres in Meghalaya and to examine the role of environmental factors responsible for transmitting malaria cases. Malaria surveillance data were collected by the Meghalaya National Vector Borne Disease Control Programme (NVBDCP). Climatic factors, including temperature, precipitation, and humidity, were obtained from the NASA Power Portal. In Meghalaya, the seasonal trends of malaria transmission were analysed using the time series technique. For detecting spatial hotspots, the Getis-Ord\* method will be employed. Subsequently, the Distributed Lag Non-Linear Model (DLNM) was employed to examine the relationship between malaria cases and climatic factors. Malaria cases in South Garo Hills follow a seasonal pattern, with the peak season during the monsoon period. The highest mean monthly malaria case count was found in Baghmara block, while the lowest number of cases was observed in Gasaupara block during the monsoon period. In 2017, major malaria hotspots were concentrated in the Chokpot block; however, with the latest trend, the Baghmara and Rongara blocks have been identified as critical hotspot regions in Meghalaya. DLNM revealed that higher temperatures, increased precipitation, and high humidity can significantly increase the risk of malaria in South Garo Hills, and are measured by the cumulative Incidence Rate Ratio (IRR) across different exposure values. These findings will inform targeted interventions that enable focused resource allocation. Furthermore, by integrating climatic factors, potential outbreaks can be anticipated, allowing preventive measures to be taken in advance and thereby reducing the risk of transmission.

## Evidence based strategy to meet the malaria elimination challenges in Angul district, Odisha, India by 2030

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Malaria remains a major health problem in many parts of Odisha and India. Angul, a centrally located district of Odisha state is under the complexity of rural, industrial/mining and forested dynamics with population immigration and outmigration. Once, Angul district was inflicted with high malaria burden having over 90% falciparum infection. Due to state specific strategy, malaria case load reduced to a very low level during 2015 to 2022 following a rising trend since 2023 as per malaria elimination by 2030 as per NFME, 2016. The present study aims to provide analysis of malaria prevalence, incidence and associated deaths, shedding light on the evolving burden of the disease in Angul district which would help in understanding the malaria epidemiology and challenges towards the malaria elimination.

Secondary data from the malaria program along with intervention inputs like LLIN, IRS and DAMaN in Angul district were analyzed between 2015 to 2025 and Annualised indicators were calculated for 2025 taking the program data from January to August. Excel based software was used for data analysis & interpretation. GIS software was used for identifying malaria hotspots.

Malaria declined since 2015 to 2019 followed with a slight rise in 2020 & 2021. Malaria decline was historically low in 2022. But since 2023 onwards there is an inclining trend. In 2015, number of malaria cases was 29853 which reduced to 43 in 2022 i.e. 99% decline. In 2024 malaria cases increased to 548 (1174% increase). Similar trend was observed in the DAMaN areas. In 2017, DAMaN program of Angul district reported 2700 malaria cases which reduced to 13 in 2022 (99% decline), but then increased to 39 in 2024 (300% incline). By analyzing the routine & DAMaN data, the district VBD office could identify the sub-center wise villages reporting high number of malaria cases after the rapid declining phase. To combat malaria in the villages strategic intervention plan has been designed.

This study highlights the unstable malaria situation in Angul district which poses a challenging situation towards malaria elimination by 2030. Data driven evidences could help to focus with a district specific plan for malaria elimination in Angul district. The learning and output of the study and evidence of the planned intervention would help towards malaria elimination drive in Angul and other districts with similar malaria situation.

## Bridging the Malaria Diagnostic Gap: The Promise of Point-of-Care Testing in Resource-Limited Settings

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Malaria still remains a significant public health disease despite several advanced technologies developed and employed in last 5 decades. According to the WHO *World Malaria Report 2024*, an estimated 263 million cases and ~597,000 deaths globally in 2023. India contributed nearly one-third of cases in the South-East Asia region, and exited WHO's HBHI group in 2024. Despite advances in control strategies, program by national and international agencies and recent developed vaccines, diagnostic and surveillance systems remain critical bottlenecks in malaria elimination in low- and middle-income settings.

A comprehensive literature review was conducted, analysing peer-reviewed journal articles, WHO reports, related to malaria diagnostics including POCT in mainly focusing on middle income and low-income countries. All the studies were collected through targeted searches in different databases like PubMed and Scopus using keywords and a detailed methodologies has been prepared by using bioinformatic tools and wet lab standard protocol.

The study highlighted several challenges related to malaria diagnosis and the use of Point-of-Care Testing including low parasite count present, limited health worker availability in resource limited areas, the limitations in the timely access to the results from conventional method and the challenges experienced in new developed method and their availability and affordability.

The need of highly sensitive and specific POCT is important for the effective malaria diagnosis and surveillance in middle- and low-income countries, while challenges to improve its diagnostic accuracy, affordability, and accessibility using high end technological solutions is possible. POCT will be a promising alternative to overcome these challenges. our country like India having several issues like healthcare access, emerging resistance, and availability of new diagnostic methods will be vital for achieving effective malaria control. Future directions include integrating artificial intelligence (AI) for automated interpretation and digital reporting, which can enhance the speed of surveillance and accelerate India's progress toward malaria elimination by 2030.

## Malaria vector Bionomics in highly endemic district Dantewada, Chhattisgarh

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Malaria continues to be a significant health problem in India. 80% of the malaria reported in India is from the tribal, hilly and forested areas. Malaria is a significant health issue in Chhattisgarh state, especially in the forested tribal areas. Despite the decrease in Prevalence in Chhattisgarh, few districts like Dantewada still report very high malaria incidence. Vector control is one of the essential components of the malaria elimination program in endemic areas. For successful implementation of any vector control tool knowledge about vector behaviour and bionomics is essential. The study aimed to study bionomics and behaviour of malaria vectors in Dantewada. Adult mosquito were collected from indoor as well as outdoor. Vector was collected by ,and catch total catch and human landing collection. Vector surveys were carried out in pre-monsoon, monsoon and post-monsoon period for vector density and their bionomics. *Anopheles* species are morphotyped and sibling species of *An. fluviatilis* were identified using PCR. All the female anophelines were tested for sporozoite positivity and blood meal sources using PCR. A total of seven different *Anopheles* species were collected in the survey. *An. subpictus* (n- 3234) is the predominant species followed by *An. culicifacies* (n-1023) and *An. fluviatilis*(n- 216). Indoor resting behaviour is more common as compare to outdoor. In indoor, the vector were resting mainly in human dwelling and cattle shed. Sporozoite positivity of *A. culicifacies* in pre-monsoon is 0.4854%, in monsoon 1.3245%, zero sporozoite positivity in post-monsoon season. Blood meal source of all *Anopheline* mosquito is mainly cow, in addition they also fed on human and goat. *An. culicifacies* and *An. fluviatilis* are the key malaria vector in district Dantewada of Chhattisgarh. Vectors were having indoor resting behaviour with preference to human dwelling compared to Cattle sheds.

## Managing Malaria Upsurge in Purulia, West Bengal: A Three-Dimensional Strategy linking Epidemiology, Entomology and Awareness Generation

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Purulia district, one of the important malaria-endemic regions of India and the second most tribal-dominated district of West Bengal, witnessed a notable malaria upsurge during 2024, with 76.06% of cases concentrated in six blocks—Arsha, Baghmundi, Balarampur, Bandwan, Jhalda-I, and Jhalda-II. Epidemiological analysis revealed that 65.25% of the cases were due to *Plasmodium falciparum* and 34.75% to *P. vivax*, with the peak incidence occurring in July–August. Approximately 51% of the total cases were recorded among tribal communities, reflecting their disproportionate vulnerability. Early entomological alerts were generated in June 2024, when higher per man-hour density (PMHD) values and larval densities were observed.

Entomological and environmental surveys were carried out following NVBDCP guidelines. Adult mosquitoes were collected during early morning hours (05:00–07:00 h) from cattle sheds and households using aspirators and test tubes, while larval surveys targeted natural habitats such as pond edges, puddles, low-running streams, and rivulets, as well as peri-domestic containers including PVC drums, storage tanks, plastic sheets, and unplanned drainage systems. The surveys revealed four vector species: *Anopheles culicifacies* (35.16%), *An. vagus* (32.97%), *An. annularis* (24.18%), and *An. fluviatilis* (7.69%), most of which were endophagic and endophilic in nature. High larval density and elevated per man-hour density (PMHD) generated entomological alerts, while observations showed suboptimal LLIN utilization due to hot and humid conditions. Mixed breeding of *Anopheles*, *Aedes*, and *Culex* larvae was also recorded in several habitats.

To address the upsurge, a three-dimensional strategy was implemented: epidemiological surveillance with early detection and prompt treatment, entomological interventions including source reduction and bio-larvicide spraying, and community awareness generation through IEC/BCC activities such as live demonstrations of *Anopheles* mosquitoes. These measures enhanced awareness of LLIN use, larval source management, and personal protection, and successfully curtailed the outbreak within two fortnights. The experience highlights the effectiveness of integrating epidemiology, entomology, and social mobilization in managing malaria in tribal-dominated endemic districts.

## Poster Presentation-18

***In vitro and in vivo* evaluation and synergistic adjuvant effect of Noscapiene as a potential antimalarial vis-à-vis Dihydroartemisinin (DHA) against *Plasmodium falciparum* 3D7, *P. falciparum* clinical isolate Pf140/SS and *P. berghei* ANKA**

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The emergence of artemisinin resistance has necessitated the urgent exploration of alternative antimalarials with enhanced efficacy and safety profiles. This study evaluates the antimalarial and synergistic adjuvant potential of the natural compound Noscapiene against *Plasmodium falciparum* (Pf) both clinical and lab adopted strains and *Plasmodium berghei* Anka (PbA). Noscapiene exhibited potent antimalarial activity, outperforming dihydroartemisinin (DHA) with IC<sub>50</sub> values of 7.68±0.88 nM/mL and 5.57±0.74 nM/mL against Pf 3D7 and clinical isolate Pf140/SS, respectively. In Pb-infected Wistar albino rats, Noscapiene achieved >95% inhibition after a 4-day suppressive test. It demonstrated minimal hemolysis, low cytotoxicity, and high cell viability, with a CC<sub>50</sub> of 1748 nM/mL in J774.A.1 macrophages. Mechanistic studies revealed Noscapiene significantly inhibited Pf Falcipain-2 (PfFp-2) expression, a critical cysteine protease in malaria pathogenesis, with enhanced efficacy in clinical isolates. Furthermore, Noscapiene's synergistic adjuvant potential was assessed in an *in vitro* co-culture model of Pf-infected erythrocytes and THP-1 monocytes. Treatment with Noscapiene and DHA significantly reduced pro-inflammatory cytokines TNF- $\alpha$  and IL-6, while increasing anti-inflammatory IL-10 levels (p<0.01). Gene expression analysis demonstrated downregulation of MyD88 mediated NF- $\kappa$ B signaling components, suggesting modulation of inflammation in severe malaria. This is the first comprehensive report on Noscapiene's dual antimalarial and immunomodulatory effects, showcasing its ability to effectively inhibit parasite growth, reduce inflammation, and enhance host immunity. These findings highlight Noscapiene's promise as a safer, potent alternative or adjunctive therapy to DHA in malaria treatment. Further validation through multi-centric clinical studies is warranted to establish Noscapiene as a novel therapeutic agent against malaria.

## Potential of Secondary malaria vectors in malaria transmission dynamics across North-eastern states of India

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Malaria is endemic in the North eastern regions of India, in which a variety of ecological variables promote diverse vector species. The major primary malaria vector in this region has long been considered to be *Anopheles minimus* and *Anopheles baimaii*, both closely associated with forest ecosystems. The variety of secondary malaria vectors have grown increasingly important for sustaining residual malaria transmission. In the states of Assam, Tripura, Meghalaya and Mizoram, this study investigates the distribution and occurrence of different secondary malaria vectors, particularly *An. culicifacies*, *An. annularis*, *An. vagus*, *An. subpictus*, *An. nivipes/ phillipinensis*. In Goa, India, *A. subpictus*, which is considered to serve as a malaria vector, occurred in the presence of *P. vivax* and *P. falciparum* in the midguts and salivary glands. According to entomological surveys that occurred within a variety of ecological surroundings, particularly in Assam, several secondary vectors that are widely distributed, usually coexist with primary species, have been identified as sporozoite positive and demonstrate zoophilic and exophagic adaptive behaviour. Their seasonal abundance during the monsoon and post- monsoon periods, when malaria transmission is at its peak. The evolving behavior of secondary vector allows to avoid typical indoor- based interventions like IRS and LLINs. As of 2017, insecticide status of *An. culicifacies* recognised in Assam showed they were resistant to DDT and possibly resistant to Deltamethrin. The results indicate that secondary vectors still maintain low- level transmission in areas of focus even though the prevalence of primary vectors has reduced as the consequence of additional intense vector control approaches. Secondary malaria vectors are critical for the dynamics of malaria transmission when primary malaria vectors are either inactive or under control. This study emphasises how critical secondary malaria vectors have grown to the entire effort to eradicate malaria and indicates the need for further investigations regarding species- specific, more effective approaches to control. These outcomes highlighted the importance of enhanced insecticide susceptibility evaluations, vector surveillance, and integrated vector management strategies that target the primary and secondary malaria vectors in northeastern India.



## Harnessing Drug Repurposing for Multi-Target Antimalarial Development: Bridging Molecular Innovation and India's Elimination Goals

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Drug resistance in *Plasmodium falciparum* poses a major barrier to malaria elimination, diminishing the effectiveness of frontline therapies and driving the need for innovative solutions in antimalarial discovery. Drug repurposing offers a strategic advantage by leveraging clinically established compounds with known safety profiles, thus accelerating the drug development pipeline. In this study, we adopted a multi-target inhibition framework aimed at simultaneously disrupting key parasite pathways to reduce the risk of resistance emergence.

A curated set of approved and late-stage investigational compounds was computationally screened against essential *Plasmodium* proteins spanning multiple biological functions: glycolytic enzymes (*Pf*FBP, *Pf*PGM1), mitochondrial electron transport components (cytochrome bc<sub>1</sub>, *Pf*MQO), epigenetic regulators (*Pf*PMRTs) and metabolic regulators (*Pf*PMT, *Pf*LPL3, *Pf*GluPho). Pharmacophore modeling and comparative docking revealed recurring scaffolds capable of dual or triple target engagement, supporting the concept of mechanistic redundancy and strengthening the barrier to resistance.

Hits were prioritized using a combined metric of predicted binding affinity, ADME-Tox profiling and pharmacokinetic parameters such as half-life optimization and minimization of cardiac liabilities. Preliminary findings identified several non-antimalarial drug classes with promising multi-target binding potential. These candidates are currently progressing to in vitro evaluation against *P. falciparum* strains to validate their efficacy and guide lead optimization.

By integrating drug repurposing with a multi-target inhibition strategy, this work not only accelerates the identification of safe and effective antimalarial candidates but also introduces a paradigm shift in drug discovery—transforming the fight against malaria from single-target interventions toward resilient, multi-site therapeutic solutions that support both control and elimination goals.



## Exploring realistic target(s) of malaria elimination through novel approaches to empower the ongoing malaria elimination efforts in India: A mythological inspiration from Ramayana

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Current approach of Malaria elimination involves multifaceted approach (e.g. MSAT, RDT, ACT, LLIN, IRS, IEC etc.), however it is severely challenged by major scientific bottlenecks as well as social and implementation barriers, e.g. Drug/Diagnostic/ Insecticide resistance, inaccessibility, lack of Community Engagement, insufficiency in surveillance, etc. Given the present context of malaria elimination in the face of these multiple challenges, a novel approach (hypothesis) inspired from the famous Indian mythology Ramayana is presented here. When Lord Rama had aimed at multiple heads of Demon king Ravana to eliminate him, he couldn't succeed until the mystery of his life was revealed to him by Bibhishana (Ravana's youngest brother). Likewise, there is an urgent need for exploring and adopting realistic target(s) of malaria elimination which include hidden parasite reservoirs, new parasite/vector species, biomarker-gene deletion(s), Asymptomatic malaria, conducive environmental/climatic factors - all of which, hypothetically compared with the Demon's navel which was the real target as it contained the 'Juice of immortality' (Amrit Kalash). Similar to unravelling of the real mystery of Ravana's death, a combinatorial approach targeting the underlying challenges for malaria elimination can be adopted to empower the ongoing malaria elimination efforts. Lord Rama had used 31 arrows (10 for Heads, 20 for Arms) before deploying the ultimate weapon, Brahmastra (01) targeting for the Naval containing Amrit Kalash. Likewise, for the complete elimination of the deadly vector-borne disease, the arrows of Lord Rama could be the following approaches: (a) introducing or strengthening Molecular diagnostics, (b) discovering alternative biomarkers, (c) deploying digital real-time Microscopy, (d) Enhancing Vector Surveillance with species ID, Hotspots ID using AI tool/Drones, (e) Environmental and climate studies, (f) Smartphone cum local community-led outbreak information and quick response measures etc, to target simultaneously the Vectors (Amrit Kalash) and destroy the Parasite (Amrit), to achieve the elimination goal.

## Harnessing Structural Biology and Computational Tools for the Discovery of Broad-Spectrum Antimalarials

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Malaria continues to pose a major public health burden in India despite significant advances in control strategies, with emerging drug resistance in the *P. falciparum* and *P. vivax* threatening existing therapeutic options. To contribute towards India's malaria elimination programme, our research focuses on the rational design of novel antimalarials employing a multi-target inhibition strategy. Instead of restricting drug action to a single protein, we are exploring pharmacophores and scaffolds that can selectively engage multiple essential enzymes or proteins with overlapping binding pocket features. This approach not only increases the likelihood of achieving higher efficacy but also reduces the risk of resistance development.

Through *in silico* pharmacophore modelling, molecular docking, and comparative binding site analysis, we have shortlisted several *Plasmodium* targets of high therapeutic relevance, including enzymes of the glycolytic pathway (*Pf*FBP, *Pf*PGM1), mitochondrial electron transport chain (cytochrome bc<sub>1</sub>, *Pf*MQO), and metabolic regulators such as *Pf*PMT, *Pf*LPL3, and *Pf*GluPho. Common structural motifs interacting across these targets were identified, enabling the construction of prototype scaffolds capable of dual or triple binding activity. Lead prioritisation was guided by potency, drug-likeness, and predicted pharmacokinetic properties, with particular emphasis on extending half-life and overcoming cardiotoxic liabilities such as QTc prolongation.

This multi-target scaffold-based strategy provides a promising framework for next-generation antimalarials, combining rational design with resistance-prevention mechanisms. Ongoing efforts include optimisation of hits into leads, *in vitro* validation, and integration of ADME-Toxicity profiling. By aligning molecular innovation with national elimination goals, our work underscores the potential of multi-target inhibitors as viable candidates in India's roadmap towards malaria elimination.

## Molecular Aspects of Artemisinin Drug Resistance in *Plasmodium falciparum* Isolates from Odisha: A Decade of Post-Malaria Drug Policy, Routine Surveillance for Elimination

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Effective drug policies require continuous monitoring of resistance to assess the spread & effectiveness of resistance. Estimation of the spread and advancement of *Plasmodium falciparum* artemisinin-resistant parasites can be done by probing polymorphisms in the blades of the *Pfkelch13* propeller domain. Hence the present study was undertaken to assess the prevalence of artemisinin drug resistance markers associated with treatment failures on *P. falciparum* isolates, during the year 2018 and 2020, after ten years of the introduction of new drug policy in Odisha.

Nested PCR was performed in 239 *P. falciparum* mono-infection isolates for amplification of the propeller region of the *Pfkelch13* gene (from blade 1 to blade 5 and partially blade 6; 849-nucleotide base pair, bearing ART-resistant SNPs-Y493H, R539T, I543T, P441L, F446I, G449A, N458Y, P553L, R561H, V568G, P574L, C580Y, A675V) have shown quality amplification in 168 (70.3%) samples, which were selected for DNA sequence analysis (Sanger Sequencing methods).

None of the validated mutations associated with artemisinin resistance was found in the *P. falciparum* parasite population circulating in Odisha during the present study. However, presence of six silent/synonymous mutations that were not coding any proteins, which were not reported earlier in India, has been detected, indicating that *P. falciparum* population in Odisha are unique.

No non-synonymous mutations have been observed during the present study despite occurrence of silent/synonymous mutations, not reported earlier in the Indian sub-continent indicating no direct threat to the consequence of ART on the *P. falciparum* isolates in Odisha, which is the only drug to combat uncomplicated *falciparum* malaria. More attention is required on account of the surveillance study for the complete elimination of malaria in Odisha. Field-based survey along with low-cost molecular diagnosis, genotyping of markers related to causing clinical failure of ART should be carried out along with a large number of sample sizes.

## **Malaria Treatment Seeking Behaviour and Related Factors Among Koya Tribe in Malkangiri District: A Qualitative Study**

**Rojalin Maharana\***, Pradeep K Panda, Kalpana Panigrahi, Paramjot Panda

Malaria deaths increased by 12% globally in 2020 compared with 2019, to an estimated 627,000. According to the WHO malaria report, India has occupied the world's fourth-highest number of malaria cases and deaths. Around 95% of the country's population is living in malaria-endemic areas. In the current study in Odisha, many tribes live mainly on hilltops and foothill villages. Koyas (70%) is the predominant tribe in the Malkangiri district. Therefore, this study was conducted to determine the knowledge and practices of the Koya Tribe towards treatment-seeking behaviour on malaria illness and the level of awareness and perception of ITNs as a preventive measure against malaria and develop a framework for the policy makers to understand the culture, practice of tribes for the malaria elimination from tribal pocket. A community-based cross-sectional qualitative study was done in the Kalimela Block of Malkangiri District, Odisha, with a sample size of 30 participants that were purposively selected based on a list of hotspots obtained from the National Vector Borne Disease Control Program Unit cell. In-depth interviews were conducted among 18 participants/stakeholders (Medical Officer - 3, ANMs/Staff Nurse - 3, ASHA workers - 3, Adolescents - 3 and Malaria Survivors - 6). In addition, two FGDs were conducted - one among males (6) and one among females (6). Semi-structured interview tools were used to elicit information from various participants/stakeholders. For qualitative analysis, MAXQDA20 Version software was used to generate codes and do the thematic analysis. The qualitative analysis showed several barriers to malaria treatment-seeking behaviour, such as severe economic, social, and psychological distress, lack of transport, environmental hygiene and health-related distress as evidenced by various themes narrated by the malaria patients and different stakeholders.

The following nine themes were identified based on thematic analysis: (a) Lack of awareness of malaria treatment-seeking, (b) Traditional cultural beliefs on malaria treatment, (c) Unhygienic environment, (d) Poor access to service to LLIN and ITNs, (e) Delay in diagnosis, (f) Lack of transportation, (g) Language barriers, (h) Fear of out-of-pocket expenditure and (i) Not prioritizing health/Negligence. The study recommends that the government should provide awareness regarding malaria treatment to the people to decrease the number of malaria cases and provide the LLIN and ITNs as a preventive measure to the people of the Koya tribe.

## **Determinants of Cross-Border Malaria Transmission in India's Northeastern Region: Implications for Targeted Control**

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The Northeastern Region (NER) of India shares borders with Bangladesh and Myanmar, areas known for artemisinin treatment failures. This study aimed to identify key loci of malaria transmission in the NER and associated risk factors, aligned with India's target to eliminate malaria by 2030.

Retrospective malaria epidemiological data at state and district levels were analyzed to pinpoint critical loci. Land use and land cover data, meteorological factors, drug resistance profiles, and malaria vector dynamics in regions bordering Bangladesh and Myanmar were evaluated to identify risk factors contributing to persistent malaria transmission.

Nine districts from Mizoram, Tripura, and Meghalaya were identified as crucial loci. These districts border Bangladesh's Chittagong Hill Tract and Myanmar's Chin State, sharing similar geographical and climatic conditions that facilitate malaria transmission, predominantly *Plasmodium falciparum* (~78% to ~98%). Key environmental factors, including forest cover (~61% to ~93%), relative humidity (~70% to ~85%), and temperature (~20°C to ~25°C), were conducive to vector and parasite proliferation.

Shared epidemiological and environmental conditions, coupled with porous international borders, pose a significant risk of cross-border malaria transmission and the importation of artemisinin-resistant parasites into the NER. Establishing regional malaria intervention sentinel sites in these critical areas is essential to enhance case detection, monitoring, and response systems.

## Regulation of Hemoglobin Uptake Genes in Artemisinin-Resistance *Plasmodium falciparum* strain

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Malaria remains a significant global health challenge, with *Plasmodium falciparum* being the most causative agent. While artemisinin-based combination therapies (ACTs) are the cornerstone of malaria treatment, the emergence of artemisinin resistance poses a significant threat to malaria resurgence. The present scenario demands the development of newer or alternative therapeutic strategies. This requires understanding the molecular mechanisms underlying artemisinin resistance. To accomplish this, the mechanism behind hemoglobin uptake and its connection to kelch13 mutation must be investigated. As artemisinin is activated by heme, a by-product of hemoglobin digestion, it can be hypothesised that by modulating hemoglobin uptake, the parasite may reduce heme production, thereby inactivating artemisinin and enhancing its survival. The study focuses on the gene expression analysis of key proteins involved in hemoglobin uptake, including coronin, ubiquitin-binding protein 1 (UBP1), AP-2 $\mu$ , and EPS-15, in wild-type and mutant strains with a focus on the Kelch 13 C580Y mutation associated with artemisinin resistance. The primers were designed, RNA was extracted, and quantitative PCR was conducted to analyze the expression of selected hemoglobin uptake protein genes in both wild-type 3D7 and C580Y mutant strains. The results show a downregulation of genes involved in hemoglobin uptake, suggesting that resistant parasites reduce hemoglobin uptake, which may contribute to their survival. This study has the potential to identify key biomarkers that could be targeted to increase heme production and enhance artemisinin activation, improving its efficacy against the parasite.

## Role of Anopheles Vector in Changing Scenario of Malaria Epidemiology

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The global fight against malaria has seen a paradoxical challenge, emerge a rising proportion of asymptomatic *Plasmodium* infections in areas previously dominated by symptomatic cases. These silent reservoirs contribute to sustained transmission, evade routine detection, and complicate elimination efforts. While human and parasite factors have been studied extensively, the role of *Anopheles* vector dynamics in this epidemiological shift remains underexplored. This study investigates how changes in *Anopheles* mosquito species composition, behavior, and ecological adaptations may be influencing the transition from symptomatic to asymptomatic malaria infections in endemic regions. This study was conducted in two phases (2016-2019 and 2024- Present) to assess temporal changes in *Anopheles* vector dynamics and their association with the emergence of asymptomatic malaria infections in Kalahandi district. Comparative analysis allowed for identification of evolving vector behaviors and their epidemiological implications. Mosquitoes were collected using standard techniques and identified morphologically and molecularly. Vector infection rates were determined through PCR. Concurrently, human participants were screened for *Plasmodium* infections using RDTs and PCR, with clinical status documented to distinguish symptomatic from asymptomatic cases. Preliminary findings indicate a notable shift in vector species dominance, with secondary or outdoor-biting *Anopheles* species gaining prevalence in areas with higher asymptomatic infection rates. These species exhibited lower sporozoite rates and altered biting behavior, potentially leading to lower inoculum loads and delayed host immune response. Increased insecticide resistance was also associated with vector behavioral changes, influencing transmission patterns. Evolving *Anopheles* vector dynamics appear to play a significant role in the increasing burden of asymptomatic malaria. These findings highlight the need to adapt vector surveillance and control strategies to address the silent threat of asymptomatic carriers, particularly in the context of malaria elimination programs.



## Prevalence of haemoglobinopathies and G6PD deficiency among particularly vulnerable tribal groups residing malaria endemic areas of Odisha, India

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Inheritable diseases like Sickle cell anemia, Thalassemia and G6PD deficiency are most prevalent among Indian tribal groups in malaria-endemic regions. There is a lack of epidemiological information specific to Particularly Vulnerable Tribal Groups (PVTGs), which is crucial for developing and implementing appropriate malaria control strategies. Because Odisha reports the highest number of malaria cases in the nation, the current community-based study sought to determine the prevalence of haemoglobinopathies and G6PD deficiency among the 13 PVTGs there. This cross-sectional study was conducted among 13 PVTGs in 12 districts, in an estimated sample size of 1461, using a two-stage sampling method. Detection of haemoglobinopathies was done by the variant analyser. Screening of G6PD deficiency was carried out using DPIP method followed by quantification using spectrophotometry. The PCR-RFLP technology was used to determine variant of G6PD deficiency. GAP-PCR was used for detecting the  $\alpha$ -thalassaemia mutation respectively. The diagnosis of malaria was done by Pf-PAN RDT as point of care followed by nPCR for confirmation and Plasmodium species identification. The prevalence of sickle cell heterozygotes (AS) was 3.4%, sickle cell homozygous (SS) 0.1%,  $\beta$ -thalassaemia heterozygotes 0.3%, Hb S/ $\beta$ -thalassaemia compound heterozygote 0.07%, Hb S- $\alpha$ -thalassaemia 2.1%, G6PD deficiency 3.2% and malaria 8.1%. In case of  $\alpha$ -thal,  $\alpha\alpha/\alpha$ -3.7 gene deletion was most frequent (38%), followed by  $\alpha\alpha/\alpha$ -4.2 (18%) and  $\alpha$ -3.7/ $\alpha$ -3.7 (4%). The frequency of G6PD Orissa (131C  $\rightarrow$  G) mutation was found to be 97.9% and G6PD Mediterranean (563C  $\rightarrow$  T) 2.1%. Around 57.4% of G6PD deficient individuals and 16% of the AS were found to be malaria positive. In conclusion, the PVTGs of Odisha have a high frequency of alleles like G6PD deficiency, Hb S, and  $\alpha$ -thalassaemia associated with malaria. The moderate to high frequency of G6PD deficiency suggests the need for G6PD deficiency diagnosis and management especially for malaria positive cases, as well as mandatory newborn screening programmes, prenatal screenings, and counselling for these genetic disorders, especially Hb S, for the PVTGs.



## ***In vitro* Identification and analysis of Immunoprophylactic Candidate for Visceral Leishmaniasis**

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VL is a pathophysiological condition of *Leishmania* and is regulated by cellular immunity. Dominance of MHC polymorphism and antigen variability is a factor in HLA-specific epitope discovery.

Screening of MHC class-I and II-restricted epitopes of *Leishmania donovani* identified a consensus HLA A02:01, HLA-DRB1\*01:01-restricted epitopes, respectively. Based on *in silico* screening, followed by population coverage analysis, and HLA cross-presentation ability, the five best epitopes in each class were evaluated. *In vitro* assays to evaluate the immunogenicity of these peptides by isolating peripheral blood mononuclear cells (PBMCs) from healthy donors and VL patients with respect to HLA Type were performed. Peptide stimulation, proliferation, and cytokine secretion assays are being conducted.

The analysis revealed that the set of epitopes presented by diverse MHC class-I and II alleles has maximum population coverage using IEDB software. IL-10Pred signifies the immunopathogenic nature of the shortlisted peptides. Selected epitopes have the potential to generate significantly higher proinflammatory cytokine (CD4<sup>+</sup>, CD8<sup>+</sup> IFN- $\gamma$ ). This comprehensive approach allowed us to identify potential Th1 stimulatory peptides that could effectively stimulate a protective immune response, thereby highlighting their potential as vaccine candidates.

The Protein fragment identified in this study can be further evaluated for the development of a prophylactic or therapeutic study. Stimulation with the cocktail of epitopes against the VL-treated subject will provide an enhanced CD4<sup>+</sup> and IFN- $\gamma$  level. The selected epitopes have the potential to generate Th1 immune responses rather than Th2, which is important for suppressing leishmania infection.

## Poster Presentation-30

**“Molecular Xenomonitoring for detection of *Brugia malayi* in Odisha: Evidence from a pilot entomological study.”**

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Lymphatic filariasis (LF), also known as elephantiasis, is a mosquito-borne parasitic disease, which manifests with chronic morbidity, including lymphoedema and hydrocele, due to lymphatic dysfunction and obstruction<sup>1</sup>. Odisha has historically been endemic for LF, with infections primarily attributed to *Wuchereria bancrofti* and *Brugia malayi*. Notably, 98.4% of *B. malayi* cases in the state have been reported from coastal districts like Bhadrak and Baleswar, where the presence of numerous rivers, tributaries, and seasonal flooding provides favourable breeding habitats for *Culex*, *Mansonia* and *Anopheles* mosquitoes<sup>2,3</sup>. The first report of a *B. malayi* focus in Odisha was from Chudamani, Balasore, in 1989<sup>4</sup>. Despite the implementation of mass drug administration (MDA) under the National Filaria Control Program, evidence of ongoing transmission in these districts highlights the importance of ensuring more sensitive, non-invasive and scalable techniques for surveillance. Molecular xenomonitoring (MX), which detects filarial DNA in mosquito vectors, has emerged as a valuable tool for post-MDA monitoring, particularly when human infection prevalence is low. MX provides an early warning signal of residual or recrudescence transmission and has been recommended as a complementary surveillance strategy alongside transmission assessment surveys (TAS) for at least 10 years in the post-elimination phase<sup>5,6</sup>. Additionally, MX can be used to detect parasite carriage in non-vector species and to mapping of co-endemic areas, thereby enhancing integrated surveillance frameworks<sup>3,7</sup>.

This pilot study conducted entomological surveillance in two coastal districts of Odisha, Puri and Jagatsinghpur, both of which had passed TAS-III, indicating sustained interruption of LF transmission. Vector species collected and identified, included three potential filariasis vector species: *Mansonia annulifera*, *Mansonia uniformis*, and *Cx. quinquefasciatus*. Female mosquitoes were pooled (10-25 female mosquitoes per pool) and analyzed using polymerase chain reaction (PCR) assays for filarial DNA. Notably, *B. malayi* DNA was detected exclusively in *Mn. annulifera*, confirming its role as the primary vector species in the region. Maximum likelihood estimation (MLE) indicated an infection prevalence of 1.23%-5.74% (95% CI), exceeding the World Health Organization's provisional threshold for elimination<sup>5</sup>. No evidence of infections was detected in *Mn. uniformis* or *Cx. quinquefasciatus*. These findings establish the role of *Mn. annulifera* as the principal vector sustaining *B. malayi* transmission in Odisha and highlight the importance of integrating entomological evidence into post-MDA surveillance frameworks. Strengthening MX, expanding longitudinal entomological surveillance, refinement of molecular diagnostics, and programmatic capacity strengthening to ensure the sustainability of LF elimination efforts. Such an integrated surveillance approach, coupled with community engagement and health system preparedness, represents a more feasible pathway to sustain the gains of lymphatic filariasis elimination efforts.

## In silico evaluation of neem phytochemicals as modulators of pro-inflammatory cytokines: potential host directed adjuncts for lymphatic filariasis

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Lymphatic filariasis (LF) is a debilitating neglected tropical disease caused by *Wuchereria bancrofti*, *Brugia malayi*, and *B. timori*. It remains a major public health challenge across 72 endemic countries that causes chronic morbidity through lymphatic dysfunction, fibrosis, and immune dysregulation. Pro-inflammatory cytokines and chemokines, including TNF- $\alpha$ , IL-6, IL-1 $\beta$ , MIP-1 $\alpha$ , and MIP-1 $\beta$ , are central to LF pathology by amplifying inflammation and promoting tissue remodeling. Although mass drug administration (MDA) effectively eliminates filarial parasites, it fails to reverse established pathology, highlighting the need for host-directed adjunctive strategies. Neem (*Azadirachta indica*), a traditional medicinal plant, is rich in phytochemicals with immunomodulatory properties. In this study, molecular docking was performed to evaluate the interactions of nimbolide, azadirachtin, and nimbin with key inflammatory mediators. Nimbolide consistently exhibited the strongest binding affinities, particularly against IL-6R (−7.8 kcal/mol), IL-1 $\beta$  (−7.5 kcal/mol), and TNF- $\alpha$  (−7.5 kcal/mol). Azadirachtin showed selective high affinity for TNF- $\alpha$  (−7.1 kcal/mol), while nimbin demonstrated moderate, broad-spectrum interactions (−5.7 to −6.8 kcal/mol). LigPlot+ analysis revealed stable hydrogen bonding and hydrophobic interactions, most notably for nimbolide. BOILED-Egg predictions indicated good gastrointestinal absorption for nimbolide and nimbin, with poor blood–brain barrier penetration, suggesting systemic efficacy and limited CNS exposure. Despite limited drug-likeness and absorption criteria of azadirachtin, neem phytochemicals, and nimbolide show potential in modulating cytokine-driven pathology in lymphatic filariasis. These computational findings support further experimental and pharmacokinetic evaluation of neem-based adjuncts to enhance MDA outcomes in endemic regions. Neem-derived compounds thus represent affordable, plant-based candidates for adjunct therapies to complement MDA and improve LF morbidity management in endemic regions.

## To see the clinical utility and cost effectiveness of dual drug versus triple drug for Lymphatic filariasis

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Lymphatic filariasis (LF), commonly known as elephantiasis, remains a significant neglected tropical disease (NTD) and a leading cause of chronic disability worldwide. Globally, over 120 million people are affected, with nearly 40 million suffering from disfiguring and debilitating clinical manifestations such as lymphoedema, hydrocele, and elephantiasis and India bears a major share of the global LF burden. Odisha remains LF endemic, with notably high antigen detection rates and longstanding clinical disease. Mass drug administration (MDA) with dual-drug regimens such as diethylcarbamazine plus albendazole (DA) has been central to reducing the burden of lymphatic filariasis (LF) worldwide. But the World Health Organization's 2017 recommendation to use a triple-drug regimen of ivermectin, diethylcarbamazine, and albendazole (IDA) has given us a chance to expedite the elimination of LF. Randomized trials and community-based studies show that IDA achieves significantly higher microfilariae clearance than DA (84.0% vs. 61.8% at 12 months), while maintaining a similar safety profile and overall tolerability; programmatic modelling indicates that IDA's superior efficacy reduces the number of MDA rounds needed, which shortens program duration and lowers overall costs, even though per-round delivery costs are comparable and broader economic reviews confirm that preventive chemotherapy for LF is still very cost-effective, with IDA providing more value in speeding elimination timelines. All of these findings highlight the clinical utility and cost-effectiveness of IDA over DA, making it a strong option for countries addressing persistent hotspots or seeking to fast-track elimination goals.

## Evaluation of antiviral potential of traditionally used plants against chikungunya virus

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CHIKV is an arbovirus transmitted by *Aedes* mosquitoes that causes febrile illness with debilitating arthralgia. The growing emergence of Chikungunya virus (CHIKV) infection has raised a major public health concern worldwide, due to several outbreaks in different countries and the absence of vaccines or licensed drugs. To combat the spread and severity of such infection, extensive research is underway to develop drugs against the virus. Several Indian medicinal plant varieties have shown potent antiviral activity and are traditionally used to treat virus-induced fever and arthralgia. In this light, this study aims to evaluate the antiviral potential of leaf extracts from traditionally used plants. Each extract's *in vitro* safety and anti-CHIKV activity were examined using Vero cells, which showed >70% cell viability at 24 hours post-treatment. The antiviral efficacy of the most effective extract E38, was validated and found to inhibit CHIKV infection efficiently by reducing the cytopathic effect. Next, the impact of the extract on viral RNA and protein levels was evaluated, which showed approximately 90% and 95% inhibition in E1 and nsP2 gene expression, respectively, 96% and 98% reduction in viral protein E2 and nsP2, respectively, and a reduction of 90% in new viral particle formation *in vitro*. Further, it can also inhibit 80-85% of viral infection at different treatment conditions, i.e. Pre-treatment, during or Co-treatment and post-treatment. This means E38 can inhibit viral entry or attachment, can regulate host pathways, or can also have a direct effect on the viral life cycle. A similar result was observed in the physiologically relevant mouse myoblast cell line, i.e. C2C12. Altogether, these findings indicate an efficient inhibitory capacity of E38 in abrogating CHIKV infection *in vitro*. However, further experiments are necessary to understand the inhibition mechanism, the *in vivo* evaluation and developing novel therapeutics for CHIKV infections.

## Differential Transcriptional Modulation of Defensins and other Innate Immune Response Effectors by Indian isolates of *Chikungunya virus*

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Chikungunya virus (CHIKV) infection has been associated with increased morbidity and mortality, especially post its resurgence in 2006, with presentation of various atypical symptoms. Its re-emergence and the subsequent periodic outbreaks indicate the consistent evolution of the virus due to the incorporation of mutations in its genome. This can potentially elicit a differential induction of innate immune response of varying degrees, which has been poorly studied in the case of CHIKV. Of the vast spectrum of innate immune response effectors, our targets of interest are Defensins, Interferon lambda and CHIKV-specific microRNAs, for which the information regarding their modulation is scarce.

Leveraging an immunocompetent epithelial model cell line to define the varying internalisation and replication of CHIKV and differential induction of innate immune response effectors, we demonstrate that CHIKV outbreak isolates (CHIKV 2006, 2016, 2022 and 2024 outbreak isolates) have differential internalisation and replication dynamics and trigger expression of Defensins, Interferon lambda (IFN-λ) and CHIKV-specific microRNAs in an isolate-dependent manner. By utilising the epithelial cell line model, we found that CHIKV 2024, despite having the highest internalisation efficiency, shows delayed exponential replication initiation, with an unusual “Eclipse phase”. Gene expression analysis of the selected innate immune effectors revealed the modulation of DEFB2 and DEFA5 genes, the trends of IFN-λ genes coinciding with the timepoint at which each virus isolate was at its highest replication rate and CHIKV 2006 infection caused heightened overexpression of CHIKV-specific microRNAs. Bioinformatic analysis revealed the differential hybridization of CHIKV-binding microRNAs. Protein-protein interaction analysis suggests the potential interaction of DEFA5 and DEFB2 with CHIKV envelope glycoprotein.

This study provides transcriptional insights regarding the isolate-dependent modulation of innate immune response by CHIKV, as a result of its evolutionary adaptation. It also provides preliminary evidence regarding the usage of Defensins and CHIKV-interacting microRNAs as peptide-based and RNA-based therapeutics, respectively.

## Eco-epidemiological studies of Chikungunya in West Bengal, India

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Chikungunya, a mosquito-borne viral (RNA Alphavirus, Togaviridae family) disease - primarily and primitively zoonotic in origin, is a growing public health concern in different parts of the world including India. First reported in Kolkata in 1963 and then it caused outbreaks in southern and western states of India between the mid 1960s and early 1970s, followed by a prolonged lull period after 1973. It resurged in 2005–2006, triggering widespread epidemics in India.

Historically, Chikungunya had been thought to be an urban disease. Owing to rapid urbanization, global warming and other environmental changes, it has spread to suburban and rural areas. *Aedes aegypti* and *Aedes albopictus* have long been recognized as the principal vectors of Chikungunya. Several investigators have indicated the epidemiological importance of *Ae. vittatus*, although it has been underestimated and largely neglected. All of the above-mentioned vectors have been reported in Kolkata and its adjoining suburban areas.

Data from the National Vector Borne Disease Control Programme (NVBDCP) for 2018–2024 revealed 79,388 confirmed and 914,766 suspected chikungunya cases across India, with cyclical outbreaks peaking in 2022 and 2024. In West Bengal, NVBDCP data (2018–2025) reported 4,626 suspected and 468 confirmed cases, with notable increases in 2022 (1,533

suspected; 148 confirmed) and 2024 (1,734 suspected; 163 confirmed). A significant difference between suspected and confirmed cases in West Bengal (mean yearly difference: ~771 vs. 78;  $t = 2.37$ ,  $p < 0.05$ ) have been found, consistent with national trends. Vertical transmission of the virus in *Aedes* vectors could be one of the reasons for the maintenance of the virus in nature in inter-epidemic periods especially in the urban areas. Chikungunya's symptomatic similarities with dengue causing confusion about its clinical diagnosis, requiring laboratory confirmation. Sustained virus and vector surveillance and monitoring are necessary for preventing future chikungunya outbreaks, if any, enabling early detection and targeted interventions.



## Evaluation of the Dengue NS1 Ag Strip for Detection of Dengue Virus Antigen in Wild-captured *Aedes aegypti* and *Aedes albopictus*

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Dengue fever is currently one of the most important mosquito-borne diseases that affects humans in terms of morbidity and mortality. Surveillance is a critical component of any dengue prevention and control programme. The objective of this study was to evaluate the efficacy of a commercial Dengue NS1 Ag kit in detecting dengue virus (DENV) antigens within infected wild-captured *Aedes aegypti* and *Aedes albopictus* mosquitoes. Furthermore, the study aimed to assess the applicability of the Dengue NS1 Ag kit as a xenomonitoring tool. This research was conducted at the Insect Rearing and Experimental Station, a facility situated within the Jahangirnagar University campus in Savar, Dhaka, Bangladesh, during the period from May to November 2024. The research focused on the collection of *Aedes aegypti* and *Aedes albopictus* from five major dengue-affected areas within Dhaka City: Uttara, Mirpur 1 and 2, Mohammadpur, Gulshan, and Shyamoli weekly using light traps and gravidtraps. Following the identification of the collected *Aedes* species, the specimens were pooled, homogenised, and centrifuged for the NS1 test. Total 1570 *Aedes* spp. mosquitoes were collected during the study. Out of the total, 880 (56%) were *Aedes albopictus*, while 690 (44%) were *Aedes aegypti*. A total of 157 pools were created in this study, and 35 pools (22%) tested positive for the dengue virus. The result was obtained within 1-2 hours, depending on the amount of antigen present on mosquitoes. The result clearly indicated that the Dengue NS1 Ag kit is effective for detecting dengue virus in wild-captured mosquitoes and can be used as Xenomonitoring tool. The deployment of the kit in the field can contribute to the reduction of dengue transmission by raising public awareness regarding the presence of viral mosquitoes and encouraging the adoption of preventive or control measures.



## Entomo-Virological Surveillance of Arboviral Infections and Transovarial Transmission in *Aedes* Mosquitoes

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*Aedes* mosquitoes and the viruses they transmit are a major public health concern, as the emergence and re-emergence of arboviral infections like Dengue, Chikungunya, and Zika have become an international concern. Punjab is reportedly highly endemic for dengue fever, and the cases of chikungunya are also reported (NCVBDC, Punjab). Recent cross-sectional entomological surveillance reveals both known vectors of these three arboviral diseases, i.e. *Ae. aegypti* and *Ae. albopictus* were prevalent, though the transmission dynamics of the circulation of these arboviruses in the mosquito population are still poorly understood. Therefore, the present study was undertaken to standardise, detect and determine the infection of these three arboviruses in the *Aedes* mosquitoes in close proximity to the confirmed patient by NCVBDC, Punjab. Both immature stages and adults of *Aedes* were collected from and around the patient's house from August 2021 to November 2022. After identification of the species of *Aedes*, the mosquito pools were homogenised and subjected to RT-PCR for arboviral detection. A total of 316 *Aedes aegypti* mosquitoes, i.e. 68 pools, were tested for the presence of these three arboviruses. Of 68 pools, 27 consisted of wild *Aedes*, with arboviral positivity in 48.1 % (13/27) of pools. Among wild *Ae. aegypti* pools, positivity of DENV mono-infection, CHIKV mono-infection and DENV+CHIKV co-infection were noted in four, five and three pools, respectively. One wild pool of *Ae. albopictus* was positive for DENV-1. Similarly, *Ae. aegypti* F0 (adult *Aedes* developed from immatures) pools showed 58.5% (24/41) positivity for arboviruses. F0 *Aedes* showed positivity in three, fourteen and seven pools for DENV-2, CHIKV and DENV+CHIKV, respectively, suggestive of TOT. DENV serotypes and CHIKV from 35 patients' serum samples were matched with strains isolated from *Aedes*, and correlation was observed in nine instances. This is the first report of the detection of DENV and CHIKV from the wild-caught *Aedes* mosquitoes from Punjab, India, which demonstrated the presence of transovarial transmission of dengue and chikungunya virus in the districts of Punjab. Therefore, the screening of field *Aedes* species will provide us with useful information which may help the policy makers to predict the high-risk areas for DENV, CHIKV and ZIKV before there is any upsurge in the human cases.

## Dengue Vector Breeding Dynamics and Insecticide Susceptibility Patterns of *Aedes* Mosquitoes in Nagapattinam and Thiruvarur districts, Tamil Nadu, India

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In Tamil Nadu, dengue cases have always fluctuated across various districts. To identify the epidemiological and entomological causes, an investigation was conducted in the Cauvery delta districts of Nagapattinam and Thiruvarur. The entomological surveillance was carried out in selected blocks. Discarded plastic containers were identified as the primary breeding sources, followed by grinding stones, discarded tires, utensils, paper cups, coconut shells, cement water tanks, water storage containers, and Syntex tanks. Three species of *Aedes* mosquitoes were found in the surveyed districts. Co-breeding of *Anopheles* (An.) *varuna* with *Aedes* (Ae.) *aegypti* was observed in Nagapattinam district, originating from the Syntex tank. *Ae. vittatus* (non-vector) was also detected in some areas of Thiruvarur district. *Ae. albopictus* was the predominant species in both districts. A larvicidal test was performed using Temephos 50% EC. The data from entomological surveillance and larvicidal tests were analysed. Insecticide resistance test was also carried out for Deltamethrin (0.05%), Cyfluthrin (0.15), and Malathion (5%) using the WHO standard protocol. *Ae. albopictus* exhibited a higher mortality rate than the *Ae. aegypti* for Temephos 50% EC. For Insecticide insecticide-resistant test of the *Ae. albopictus* was susceptible with 100% mortality, and possible resistance was observed in *Ae. aegypti* against all three insecticide papers. This study investigates the current insecticide susceptibility status of the wild population of *Ae. aegypti* and *Ae. albopictus* from Nagapattinam and Thiruvarur districts in Tamil Nadu, India, against two major classes of insecticides: Organophosphate and Pyrethroids. The findings aim to support public health authorities and vector control personnel in formulating more effective vector management strategies. Specifically, the data generated can inform the development of resistance management plans by identifying insecticide classes that remain effective and highlighting areas where resistance may compromise control efforts.

## **The Financial Burden of Dengue and Its Coping Mechanism by Household: A Mixed-Method Study in Khordha District, Odisha**

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Dengue fever, transmitted by *Aedes Aegypti* mosquitoes, ranges from mild illness to severe conditions like Dengue Fever and Dengue Hemorrhagic Fever. It poses a substantial global health threat, with India significantly contributing to its burden. Rapid urbanisation and climatic factors facilitate its spread, particularly in states like Odisha. This study aimed to assess the financial burden imposed by dengue on households in Bhubaneswar, Odisha, and explore the coping strategies adopted by affected families, addressing a critical gap in understanding dengue's socioeconomic impact.

A case-based cross-sectional study was conducted in Bhubaneswar from May to July 2022. Data from 200 confirmed dengue patients were collected, encompassing socio-demographic details, health facility choices, and expenditure specifics. In-depth interviews were conducted with a subset of participants. Data analysis involved descriptive statistics, bivariate analysis, and linear regression.

Most patients were urban residents, predominantly males in the 21-40 age group. Private healthcare incurred significantly higher costs than government facilities (INR 21,859 vs. INR 1,225). Out-of-pocket expenditure (OOPE) averaged INR 5,567, predominantly affecting females, older age groups, higher-income individuals, and married participants. Most households (93%) utilised savings, while only 3% relied on health insurance.

Dengue fever inflicts a considerable financial and emotional toll on households in Odisha. Addressing this burden necessitates enhancing public awareness about available government schemes, regulating private healthcare costs, strengthening public healthcare infrastructure, and providing psychosocial support.

## Clinical Outcomes and Serotype Distribution of Primary and Secondary Dengue Infections in Bhubaneswar, Odisha during 2023–2024

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Dengue infection caused by four distinct serotypes is a global public health challenge. This study provides detailed clinical outcomes of primary and secondary dengue infections and the current circulation patterns of dengue serotypes in Bhubaneswar, Odisha.

Between June 2023 and December 2024, 1,986 suspected dengue samples were tested for dengue NS1 antigen using the ELISA assay. Of the 494 NS1-positive cases, 269 samples were further tested for anti-Dengue IgM and IgG antibodies via ELISA to differentiate between primary and secondary infections. Detailed clinical histories were recorded, and viral loads were quantified using pan-dengue qRT-PCR. Serotyping of pan-dengue-positive samples was performed using multiplex real-time RT-PCR to identify circulating DENV serotypes.

Based on the IgM/IgG (N-269) ratio, 193 cases (71.7%) were classified as primary dengue infections, while 76 (28.2%) had secondary infections. Secondary infections were associated with a significantly higher hospitalisation rate (22.3%) compared to primary infections (7%) due to severe clinical presentations. The rash occurrence was higher in secondary infections (34%) than in primary infections (19%).

Among hospitalised patients, 11 from primary and 9 from secondary infections required platelet transfusions. The median platelet count among dengue patients was 125,000/ $\mu$ L. Secondary infections showed a lower median platelet count (100,000/ $\mu$ L) compared to primary infections (135,000/ $\mu$ L). The median viral load among pan-dengue PCR-positive samples was 9,688 copies/mL. Primary infections exhibited a higher median viral load (17,819 copies/mL) than secondary infections (3,917 copies/mL).

From the Pan-dengue real-time PCR positive samples, 202 could be serotyped. DENV1 was the most prevalent (54.4%) serotype, followed by DENV2 (24.75%). Co-infections of DENV1-DENV2 serotypes were seen in 20.29% pan-dengue-positive samples. Sequence analysis will add further details of the circulating genotypes and lineage.

Despite low viral load, secondary dengue infections are associated with thrombocytopenia, indicating the severity of the same. The study also identified the circulation of two dengue serotypes (DENV1-2), with DENV1 being the predominant serotype. Co-infections between serotypes indicate the need for continuous surveillance and genomic epidemiology to track the evolutionary pattern of circulating strains.

## Components of Biological Essential Oil (*Eucalyptus citriodora*) with *Aedes aegypti* Acetylcholinesterase: In Silico Molecular Docking and ADME Analysis

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The transmission of dengue by *Aedes* is closely associated with human occupancy in urban settings. The main goal of our research is to reduce the ongoing reliance on synthetic insecticides. Essential oils (EO) are known to be safe for the environment and to be effective against a wide range of insects and pests. *Aedes* population reduction is getting harder, nevertheless, as a result of growing resistance to the various pesticide families. The in-silico study of *Eucalyptus citriodora* essential oil components against *Aedes aegypti* Acetylcholinesterase (AChE) and the effectiveness of EOs against *Aedes aegypti* mosquitoes were examined. The most prevalent chemicals in eucalyptus oil, according to GCMS analysis 3-Carene (14.1%), Eucalyptol (12.3%), O-Cymene (12%), Longifolene (8%), Trans pinane (5.5%),  $\alpha$ -Pinene (5.5%), (+)-4-Carene(5.3%),  $\gamma$ -Terpinene(4.7%), Camphene,(1R,4S)-(+)-(4.2%), L- $\alpha$ -Terpineol(3.7%), (1R)-2,6,6Trimethylbicyclo[3.1.1]hept-2-ene(3.2%), Longifolene-(V4)(2.3%), (-)- $\alpha$ -thujene(2.2%), m-Cymen-8-ol(2.2%), Terpinen-4-ol(2.1%) and (-)-Car-3-en-2-one(1%). *Aedes aegypti* Acetylcholinesterase (AChE) is molecularly docked with all of these chemicals. The ADMET of all these chemicals are done to predict the absorption, distribution, metabolism, excretion, and toxicity (ADMET) properties of all compounds. According to the current research, EO may be a viable bio-insecticide that works well to inhibit the growth of both adult and larvae of *Aedes aegypti* in addition to being a safe and affordable alternative to harmful insecticides. The aim of this study was to review all the in-silico strategies that have been used to find new chemicals active on *Aedes aegypti*. From the in-silico study, out of the discussed 16 chemical compounds (ligands), Longifolene, Longifolene-(V4) and L- $\alpha$ -Terpineol are showing the minimum atomic contact energy value interaction with Acetylcholinesterase (AChE) of *Aedes aegypti*. ADME shows five chemicals have high GI absorption capacity. Longifolene is not permeant to the BBB, but L- $\alpha$ -Terpineol is permeant. Therefore, L- $\alpha$ -Terpineol can be used as a potent larvicide to control *Aedes aegypti*.

## Association of CD14<sup>++</sup>CD16<sup>+</sup> intermediate monocytes with dengue severity among pediatric population in Odisha, India

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Dengue virus (DENV) infection presents diverse clinical manifestations, partly mediated by host immune responses. Innate immunity serves as the primary defence against dengue in pediatric patients, with monocytes playing a pivotal role in this response. Pediatric dengue patients (n=160) were recruited from KIMS Hospital, Bhubaneswar, India. DENV infection was confirmed via IgM ELISA and RT-PCR, and classified as dengue fever (DF), dengue with warning signs (DWS), or severe dengue (SD) per WHO guidelines. Secondary infections (DENV-IgG) were excluded. All cases were DENV-2 serotype infections. Of the cases, 114 (71.25%) were classified as DF and 46 (28.75%) as DWS & SD. Monocyte subpopulations were analysed using flow cytometry with CD14-APC and CD16-PE antibodies. Classical monocytes (CM) accounted for 73% in DF and 65.6% in DWS & SD cases. Intermediate monocytes (IM) were notably higher in DWS & SD cases (30.55%) compared to DF (12.53%), while non-classical monocytes (NCM) comprised 8.05% and 6.63% in DF and DWS & SD cases, respectively. The significant increase in IM in DWS & SD cases suggests their potential role in dengue severity.

## Combating Dengue: Advances in Diagnosis and Vaccination

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Dengue virus infection is a significant global health problem, and this vector-borne disease is transmitted by *Aedes aegypti*. It affects 50 million people, resulting in about 50,000 hospitalised with Dengue hemorrhagic fever (DHF) and a case fatality rate of about 5% to 10%. Dengue is endemic in at least 100 Asian countries, the Western Pacific, the Americas, and Africa. The dengue virus belongs to the Flaviviridae family and has four serotypes (DENV1, DENV2, DENV3 and DENV4). Its genome encodes three structural proteins (Capsid protein C, membrane protein M and envelope protein E), glycoproteins and seven non-structural proteins (NS1, NS2A, NS2B, NS3, NS4A, NS4B and NS5). Antibody-Dependent Enhancement (ADE) and early diagnosis are crucial for effective management and treatment. Current diagnosis methods, including ELISA and reverse transcription polymerase chain reaction, have limitations. Detecting nonstructural protein 1 (NS1) antigen has emerged as a promising approach. Overall, DENV NS1 antigen was detected in 98% of infected mosquitoes or pools versus 79% for RT-PCR and 20% for virus isolation. It indicates that NS1 is an excellent analyte for DENV diagnosis. Currently, the vaccine CYD-TDV, also known as Dengvaxia and other types of vaccines are under research. This review highlights the importance of NS1 detection in diagnosis, the need for advanced diagnostic kits, progress and weaknesses of various types of vaccines, their future advances and perspectives.



## Aberrant acute-phase reactants of inflammation and systemic complement levels attribute to varying grades of dengue disease severity

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Dengue virus (DENV) infection poses a serious health burden across the tropical and sub-tropical regions. Dengue manifestations are protean, ranging from asymptomatic and subclinical to severe disease with plasma leakage and organ dysfunction. The role of complement factors and acute-phase proteins in clinical dengue remains nebulous.

The enrolled 156 participants, 114 were confirmed clinical dengue cases and 42 were healthy controls. Serological profiling, estimation of serum acute-phase reactants, clinico-laboratory parameters and viral load were performed for patient classification, viz., DWS+ (n=35), DWS- (n=74) and SD (n=5) (based on varying grades of severity) in accordance with the 2009 WHO guidelines. Measurement of complement factors (C1Inh, C1q, C2, C3a, C3b, MBL, C5a, and CR1) was performed using commercial ELISA, and their concentrations were correlated with various acute-phase proteins, clinical laboratory parameters, grades of dengue disease severity, and platelet counts.

Serological classification revealed 104 patients were IgM positive, 35 were IgG positive, 24 were NS1 positive, and 26 were secondary dengue samples. Complement analysis showed a significant alteration in early classical pathway proteins, viz., C1Inh, C1q, and C2. The levels of downstream products and CR1 remained largely unchanged across both varying grades of dengue severity and primary/secondary stratification. MBL showed significant variation across the study groups but was consistent within primary and secondary dengue samples. Univariate analysis revealed NS1 positivity, IgG positivity, age, urea, and CR1 as factors associated with the severity, but further multivariate analysis showed CR1 as the only independent predictor that correlated negatively with dengue severity. Platelet counts showed a negative association with RDW and basophils, and a strong positive correlation with uric acid levels.

Our findings demonstrate that aberrant complement activation contributes to varying grades of dengue severity. Moreover, CR1 may serve as a possible predictor of dengue severity.



## Sporadic and Outbreak investigation of Japanese Encephalitis in the state of Odisha, India

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Japanese encephalitis virus (JEV) is an important cause of viral encephalitis in Asia, estimating about 100000 clinical cases globally each year, with approximately 25000 deaths. In Odisha, cases of Japanese Encephalitis reappeared in 2012. Sporadic cases of JE were laboratory confirmed every year; while 2012, 2013, 2015, and 2016 faced small to large JE outbreaks. This study emphasizes the surveillance of outbreak and sporadic cases of JEV from 2012-2024.

In coordination with IDSP and NVBDCP, VRDL at ICMR-RMRC, Bhubaneswar conducted field investigation as well as received samples for suspected AES cases from different districts, govt and private hospitals of Odisha. Anti JE IgM antibody was detected by using JE IgM MAC ELISA kit from NIV, Pune.

During 2012-2017, a total of 832(Blood and CSF) samples were received from outbreak sources which included 12 field investigations and its follow ups. The outbreaks were mostly reported from tribal dominated districts; Malkangiri, Mayurbhanj, Keonjhar, Nabarangpur, Rayagada, Koraput, Nuapada, and Sundergarh. Out of these 832 samples 205(24.5%) were JE IgM positive. Major affected age group was 6-15 years with positivity of 33% followed by  $\leq 5$  years (25%). Highest number of outbreak samples were received from Malkangir during Sept-November, 2016(n=187) and seropositivity was detected in 61 (32.6%) cases. 2026 samples (993 Serum 1285 CSF) were received from sporadic hospital cases and 140(6.9%) were positive for JE IgM including 19 CSF samples(1%).

No samples have been received from AES outbreaks after 2017. From 2018-2024, total of 3641 samples were tested with overall positivity of 10% and 3.8% in CSF. The most affected age group was 31-45 years (15.8%) followed by 16-30 years (15%).

We recorded several AES outbreaks from 2012-2016 in our state. However, in endemic areas, sporadic cases of JEV infection might occur throughout the year. As a response to the laboratory confirmation of JEV as the etiology of AES outbreaks, JEV vaccination was implemented in 2017 in 13 endemic districts of Odisha and now several districts are given under routine immunization programme. The continuous laboratory investigation has supported timely public health action and case management in the state.

## Telmisartan an AT1 receptor blocker drug via AT1/PPAR $\gamma$ pathway impedes JEV infection

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In Asia, Japanese encephalitis virus (JEV) is one of the most common pathogens responsible for viral encephalitis. The case fatality rate is around 20-30% whereas in 30-50% of cases, it causes irreversible damage to neurons of the infected individual. Vaccines are available, but the high cost and requirement of multiple doses make it more challenging for availability in poor communities. To date, there is no specific antiviral drug present in the market for the treatment of JEV; only supportive care is available. The activation of Angiotensin Type 1 (AT1) receptor by Ang II results in the formation of reactive oxygen species and inflammatory cytokines associated with viral encephalitis. Therefore, blocking AT1R activation can be an important therapeutic strategy to combat encephalitis. Thus, in the current study, an approved AT1 receptor blocker drug, telmisartan, is repurposed against JEV. TM remarkably reduced JEV titer, RNA and protein (NS3) in the BHK-21 cells. The IC<sub>50</sub>, CC<sub>50</sub> and Selectivity Index of TM were found to be 24.68 $\mu$ M, >350  $\mu$ M and >14.18, respectively. Interestingly, the viral load was significantly reduced in pre, co and post-TM treatment conditions; however, most efficiently (80%) in post-treatment. The level of AT1 was decreased, and PPAR $\gamma$  was increased, followed by decreased JEV NS3 level upon TM treatment in BHK-21 cells. Further, in JEV-infected and TM-treated RAW264.7 cells, there was a significant decrease in levels of inflammatory markers like p-IRF-3, COX-2 and p-NF- $\kappa$ B, suggesting its mechanism of action through the AT1/PPAR $\gamma$  axis. Lastly, the anti-JEV potency of TM was assessed in Balb/c mice model. The reduced disease score, viral NS3 protein and histological changes define the anti-JEV property of TM *in vivo*. Thus, the preclinical potency of TM indicates its relevance for repurposing against JEV.

## Prevalence of Scrub Typhus Among Acute Kidney Injury (AKI) Cases in Odisha

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Scrub typhus has been reported from various parts of India and has recently been identified as one of the neglected zoonotic diseases of public health importance. Scrub typhus-associated acute kidney injury (AKI) is an important cause of morbidity and mortality in tropical and sub-tropical regions. Increasing incidences of AKI in India, along with Odisha, are a serious concern. Despite being endemic in Asia with an estimated one million cases occurring annually, scrub typhus is a highly underdiagnosed and under-reported cause of hospitalisation. So, the study was undertaken to find out the association of scrub typhus with AKI, which was one of the least known and least investigated diseases in Odisha at that time. During the study, 140 blood samples of AKI patients being admitted to the Department of Nephrology, SCB Medical College, Cuttack during 2017-18 were examined. The detection of scrub typhus was done using the immunochromatographic (ICT) technique, which was further confirmed by Scrub Typhus-specific IgM ELISA. Out of the 140 AKI Samples, 36% (n=50) samples were found to be positive for scrub typhus, in which 68% were male and 32% were female. The detailed clinical feature and response to treatment of each patient was documented. The disease was found to be more prevalent among people who were less educated, with low family income and living mostly in rural areas. Our study highlighted that scrub typhus should be part of the differential diagnosis of acute febrile illness with AKI.

## Scrub typhus, a serious public health challenge in Mizoram

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Scrub typhus remains a significant neglected tropical disease in India, accounting for one-fourth of the febrile cases reported in the country. It is a vector-borne zoonotic disease caused by *Orientia tsutsugamushi* and transmitted by chigger mites. The disease is characterized by non-specific febrile illness, which often leads to underdiagnosis and underreporting in routine health systems. In recent years, North East India has emerged as one of the country's hotspots, particularly Mizoram, with recurrent outbreaks and increasing case reports. Building on the previous study covering 2018–2022, the present analysis focuses on cases reported in 2023 to 2024 to understand evolving epidemiological patterns. Retrospective secondary data from the Integrated Disease Surveillance Programme (IDSP), Health & Family Welfare Department, Mizoram, were analyzed using spatiotemporal mapping and statistical methods to identify patterns of incidence and demographic distribution. The analysis revealed that scrub typhus cases continued to be disproportionately high in urban and peri-urban areas and predominantly affected adults, reflecting the ongoing vulnerability of the working-age population. A notable rise in scrub typhus cases was observed in the study when compared to 2018, with new case reports emerging in villages with previously low case counts, highlighting the increase and ongoing expansion of the disease within the state. These findings underscore the need for strengthened diagnostic capacity, timely surveillance, and public awareness initiatives to address the continued burden of scrub typhus.

## DNA barcoding, molecular screening, and histopathology of rodents/shrew reservoir hosts of scrub typhus in the Cauvery Delta District, Tamil Nadu

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Reservoir rodents play a pivotal role in the ecology of scrub typhus. The trombiculid mites they carry are the vectors responsible for transmitting *Orientia tsutsugamushi*. Accurate identification of rodent species, diversity assessment, and determination of pathogen prevalence are crucial for effective scrub typhus disease surveillance. In this study, rodents were trapped from multiple sites in the Thiruvavur Cauvery Delta district of Tamil Nadu, and species were identified morphologically using taxonomical keys and molecularly using DNA barcoding of the mitochondrial cytochrome b gene. Phylogenetic analysis was performed to validate species identification. Rodent tissue samples of liver, spleen, lung, kidney, and brain were screened for *Orientia tsutsugamushi* using nested PCR. Histopathological evaluation using hematoxylin and eosin staining was done. A total of 128 rodents representing four species of *Rattus rattus*, *Bandicota indica*, *Suncus murinus*, and *Millardia meltada* were analysed. DNA barcoding confirmed species identity and sequences clustered with reference sequences in the phylogenetic tree. PCR screening for *O. tsutsugamushi* did not yield positive amplicons, and histopathological examination revealed no significant lesions. This study provides baseline data on rodent species diversity and their screening for *O. tsutsugamushi* in the Cauvery Delta District. The integration of DNA barcoding, molecular screening, and histopathology offers a comprehensive paradigm for rodent surveillance and contributes to understanding potential reservoirs of scrub typhus in the Thiruvavur district of Tamil Nadu.

## Prevalence of Canine Babesiosis and Tick Infestation in and Around Bhubaneswar

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Canine babesiosis is a significant and potentially life-threatening tick-borne disease caused by intra-erythrocytic protozoan parasites of the genus *Babesia*. In the Indian subcontinent, canines are predominantly infested with *Rhipicephalus sanguineus* and *Haemaphysalis* species of ticks. One of the most detrimental effects of a tick bite is the release of neurotoxins in the tick's saliva, which can cause tick paralysis, systemic illness, and hypersensitivity reactions. This study focused on assessing the prevalence of canine babesiosis and tick infestation, their molecular characterisation, and associated risk factors in and around Bhubaneswar, Eastern India. A total of 200 canine blood samples from varying ages and sexes presented to the Veterinary Clinical Complex were examined using microscopy (Giemsa-stained thin blood smears) and polymerase chain reaction (PCR). DNA was extracted through conventional methods, with primers targeting the 18s RNA gene fragment (460 bp). Blood smear analysis revealed an overall prevalence of 16.5% (33/200). All microscopy-positive samples were also confirmed by PCR, which indicated an overall prevalence of 24.5%. Ticks were collected from the dogs and processed, and identified. 55 (27.5%) dogs were found positive for tick infestation with *Rhipicephalus sanguineus*. Higher prevalence was observed in dogs aged less than 1 year (39.6%), males (38.3%), and in the rainy season (40%). This study reports the molecular characterisation of *Babesia gibsoni* from this area, shedding light on potential genetic diversity among local isolates.

## Chandipura Virus Outbreak in Gujarat 2024: Epidemiological Insights and Public Health Response

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The Chandipura virus (CHPV), a negative-sense RNA virus belonging to the family *Rhabdoviridae*, was first identified in 1965 from febrile patients in Maharashtra, India (Bhatt & Rodrigues, 1967). Since then, sporadic outbreaks have been documented in Maharashtra, Madhya Pradesh, Andhra Pradesh, and Gujarat (Sapkal et. al., 2018). During June-August 2024, Gujarat experienced a notable increase in Acute Encephalitis Syndrome (AES) cases among children under 15 years. The initial confirmed CHPV case was reported from Sabarkantha district, followed by clusters in Panchmahal and Vadodara districts (WHO, 2024).

*Phlebotomus papatasi*, a sandfly species, has been implicated as a potential vector in Gujarat (WHO, 2024). In response, vector control interventions were implemented, including indoor residual spraying with alphacypermethrin 5% WP. Community-level information, education and communication (IEC) campaigns, sensitisation of healthcare workers, and strengthening of referral mechanisms ensured rapid case detection and management (NCDC, 2024). Concurrently, a joint outbreak response team comprising experts from the National Centre for Disease Control (NCDC) and Indian Council of Medical Research (ICMR) was deployed for epidemiological and entomological investigations (NCDC, 2024).

Between June and August 2024, Gujarat reported 261 AES cases, of which 61 tested positive for CHPV using IgM ELISA (Kapoor & Gulati, 2005). Human, rodent, and cattle samples were also collected to assess transmission dynamics. These findings highlight the continued public health threat posed by CHPV, particularly in endemic regions. Sustained entomological surveillance, early case detection, and robust outbreak preparedness are essential to mitigate future outbreaks and improve our understanding of CHPV epidemiology in India.

## Diagnosis and case management of west African sleeping sickness

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Dengue, *Leishmaniosis*, and African *Trypanosomiasis* (sleeping sickness) are serious diseases that the World Health Organisation (WHO) characterises as lacking effective control measures. All three diseases affect substantial populations. Globally, the WHO estimates that 500,000 cases of DHF occur annually. The prevalence of sleeping sickness is estimated at 300,000 people, with 60million people considered to be at risk. WHO estimates the number of deaths caused by sleeping sickness as currently in the range of 50,000 to 100,000 per year. Human African *trypanosomiasis* (HAT) is caused by protozoan parasites of fatal diseases transmitted by endemic infected tsetse flies in sub-Saharan Africa by two subspecies of *Trypanosoma brucei gambiense* (92%) and *Trypanosoma brucei rhodesiense* (8%). It is transmitted by an insect vector, tsetse flies, and causes an epidemic outbreak. The vectors for sleeping sickness have a much lower reproductive potential and could be eliminated over large areas, given adequate organisation and surveillance. African trypanosomiasis is caused by parasites transmitted by tsetse flies (*Glossinidae*). The most important are forms of *Trypanosoma brucei* that infect humans and livestock, and *T. congolense* and *T. vivax* that infect only livestock. Human infection causes severe disease known as sleeping sickness, which is acute in the case of infection with *T. brucei rhodesiense* but more chronic with *T.b. gambiense*. Parasites are transmitted by the bite of infected tsetse flies. They multiply locally in extracellular spaces, producing a characteristic lesion or chancre. The parasites circulate in blood and lymph, resulting in waves of parasitemia with episodes of fever, often accompanied by chills, rigour, malaise, prostration, and weight loss. Febrile episodes become less severe as the disease progresses, and after a variable period, the parasites invade the central nervous system and cerebrospinal fluid, leading to the late stage, with meningoencephalitis typically accompanied by severe and protracted headache, apathy, sleep disorders, irritability, and antisocial behaviour. The clinical features of late-stage sleeping sickness can resemble AIDS. Untreated disease causes relentless deterioration in cerebral function, with patients becoming increasingly difficult to rouse and passing into coma and death. Sustained control efforts have reduced the number of new cases by 97% in the last 20 years. This review listed the diagnostic methods to detect different stages of West African sleeping sickness. Lab test, Physical examination, blood smear test, tomography analysis, lymph node bioscopy, serology by IFAT process, plasma detection of parasite nucleic acid by PCR analysis would provide insight into the disease and case management.



## Duplex PCR For Simultaneous Detection of *Babesia gibsoni* and *Babesia vogeli* in canines

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Canine babesiosis is a tick-borne disease caused by *Babesia* spp., which infects and destroys healthy erythrocytes, leading to mortality and morbidity in dogs. The diagnosis of babesiosis is tedious and time-consuming, especially in latent and chronic infections. In India, the two most common *Babesia* spp. are *Babesia vogeli* and *Babesia gibsoni*. Duplex PCR for simultaneous detection of *Babesia vogeli* and *Babesia gibsoni* was performed using the two primers (BAB1 F/ BAB4 R and BAG1 F / BAG1 R) at the annealing temperature of 54°C. Reaction mixture for polymerase chain reaction was prepared and was set in a final volume of 25 µl with 0.50 µl of each primer and 2 µl of template DNA. The samples which were used in the PCR assay were similar samples which was found positive for *Babesia vogeli* and *Babesia gibsoni* by single plex PCR assay. The results of duplex PCR confirmed that amplification of genomic DNA of *Babesia vogeli* and *Babesia gibsoni* yielded a PCR product of 600bp and 488 bp, respectively. Our findings suggested that Duplex PCR is a rapid, accurate, cost-effective method for *Babesia* spp. detection that is feasibly applicable for diagnosis.

## Efficacy & Residual Effect of Eight *Bacillus thuringiensis* var. *israelensis* products assayed against *Culex quinquefasciatus* (Diptera: Culicidae) & Interactive Effects of These Microbial larvicides on Mosquito Oviposition

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Since the discovery of the larvicidal property of *Bacillus thuringiensis* var. *israelensis* (Bti) and *Bacillus sphaericus* (Bs), various commercial formulations of these agents have been developed and evaluated. High efficacy, specificity, low risk of development of resistance, long shelf-life, transportability, and the safety to nontarget organisms of these microbial agents have been well documented. In the laboratory, eight microbial mosquito larvicidal products consisting of Bti & Bs were bioassayed against larvae of *Culex quinquefasciatus*. During this study, four different concentrations of Bti were set with four replicate. The magnitude of oviposition and the size, shape and number of eggs per egg raft were determined after gravid *Cx. quinquefasciatus* oviposited on water treated with Bti and on untreated water. Efficiency and residual effect assessments of the formulations were compared and the time to induce 100% mortality after exposure to serial concentrations of the larvicide varied according to the dose & formulation. Among the granular formulations of Bti of the 8 Bti assayed Aquabac WDG was the most effective which caused 100% mortality of the larval population, after 24 hr. of the first application & Mosquito dunk showed lowest mortality. Mosquiguard2 caused at least 70% mortality until 28 days which is the highest residual effect shown and Aquabac<sup>xt</sup> showed at least 60% mortality until 7 days after treatment which is the lowest, respectively. Oviposition result showed different degrees of oviposition preferences on experimental and control cups. *Cx. quinquefasciatus* tended to lay a lower number of eggs for tests groups than the control group throughout the whole test duration. Between the test groups the highest number of eggs (92±6.6) were laid by *Cx. quinquefasciatus* in water treated with Mosquito dunk & lowest number of eggs (0) on Shandong ganon agrochemical. These Microbial larvicides could be appropriate for controlling mosquito population particularly in areas experiencing high insecticide resistance or outdoor biting mosquitoes.

## Stage-Specific Bacterial Community Dynamics in *Culex quinquefasciatus* from Odisha, India

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Mosquitoes and interactions with their microbiota play a critical role in their development, physiology, and vector competence, which vary greatly depending on mosquito species, developmental stages, and environmental factors. Among different mosquito vectors, *Culex quinquefasciatus* is a potent vector of diseases such as West Nile fever, lymphatic filariasis, Japanese encephalitis, etc. However, microbial profiles associated with various life stages of field collected *Culex quinquefasciatus* remain underexplored. In this study, the dynamics of the microbial communities inside and across life stages of mosquitoes collected from Ganjam district in Odisha, India, were investigated and analyzed using full-length 16S rRNA next-generation sequencing. *Pseudomonadota* was the dominant phylum in *Culex quinquefasciatus* in all developmental stages. *Alphaproteobacteria* was the dominant bacterial class in larvae and pupae of *Culex quinquefasciatus*. However, *Gammaproteobacteria* was the dominant bacterial class in both adult males and females. *Microbacteriaceae* and *Anaplasmataceae* are the major bacterial families in the larval stage of *Culex quinquefasciatus* mosquitoes. *Anaplasmataceae* was the dominant bacterial family in pupae. *Enterobacteriaceae* was the dominant bacterial family in both adult male and female *Culex quinquefasciatus* mosquitoes. *Wolbachia* was the dominant genus in the larval and pupal stage, and *Enterobacter* was the dominant bacterial genus in the adult *Culex quinquefasciatus* mosquitoes. This study contributes to our knowledge of the dynamics and composition of bacterial communities across the developmental succession of *Culex quinquefasciatus*. The finding reveals prominent bacterial candidates that may be used to create microbe-driven control measures meant to reduce the spread of vector-borne diseases.

## Toxicity of brown seaweed *Spatoglossum asperum* J. Agardh to the larvae of vector mosquitoes

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Seaweeds abundant in bioactive compounds have been shown to affect insect's activity through intoxication. In the present study, the methanolic extract of the brown seaweed, *Spatoglossum asperum* was analysed for its bioactive constituents, and tested for larvicidal property against the early third instar of *Aedes aegypti*, *Anopheles stephensi* and *Culex quinquefasciatus* at dosages of 50, 100, 150 and 200 mg/L for 24 hours. The seaweed extract reported LC<sub>50</sub> values of 2.56, 2.85 and 3.32 mg/L against *Aedes aegypti*, *Anopheles stephensi* and *Culex quinquefasciatus*, respectively. GC-MS studies confirmed the presence of major insecticidal bioactive compounds, viz., nonadecane, cyclododecane, diethyl phthalate, pentadecanoic acid, phytol, oleic acid, vaccenic acid, octadecanoic acid, hexadecenoic acid and citronellol. This study suggests that the larval mortality be due to one or more of the active chemical constituents reported above.

## Adaptive flexibility of *Anopheles minimus* and *An. baimaii* subsequent to the deployment of long-lasting insecticidal nets in north eastern states of India

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The extensive use of long-lasting insecticidal nets (LLINs) and indoor residual spraying (IRS) has led to significant reductions in malaria-related deaths and morbidity. Nonetheless, these malaria vector control measures have significantly altered the composition and number of *Anopheles* species in the endemic area. This research aims to investigate the potential entomological factors contributing to the poor efficacy of LLINs, focusing on an LLIN-based vector control intervention implemented in communities along the Bhutan-Assam border. Three malaria-endemic communities were chosen in Orang PHC throughout the research period. The indoor and outdoor collections were conducted using the CDC light trap, total capture technique, and aspiration tube method. During the research period, *Anopheles culicifacies*, *Anopheles minimus*, *Anopheles kochi*, *Anopheles nivipes/phillipiensis*, *Anopheles baimaii*, *Anopheles vagus*, *Anopheles maculatus*, *Anopheles nigerimus*, and *Anopheles annularis*. In Anopheline species, *An. nigerimus* and *An. maculatus* exhibited high prevalence in CDC trap collections, whereas *An. vagus* was predominant in hand capture collections. *An. culicifacies* were recovered from both livestock sheds and human dwellings in the plane ecotype. The average quantity of *An. minimus* collected from light traps across three distinct ecotypes (Plain, foothills, and deep woodland) was analyzed. The documented vector species, *An. minimus* s.l., was obtained in indoor (human residence) traps and seldom in cow sheds. The quantity was small in the rural plain village. This settlement is next to a river valley. *An. Baimaii* was observed only in cow sheds during the rainy season and in deep forests and slopes, exhibiting pronounced zoophilic behavior, according to HBI findings. Moreover, the diurnal resting behavior of *An. minimus* has shown its behavioural heterogeneity. The current research demonstrates heterogeneity in the resting and feeding behaviors of malaria vectors. The results revealed a need to develop supplementary vector control instruments targeting malaria vectors that exhibit behavioral flexibility, in order to maintain malaria prevention in Assam, a northeastern state of India.

## From Data to Decisions: Intelligent Tools for Vector Surveillance

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Vector-borne diseases continue to have a significant impact on public health, especially in tropical and subtropical regions. Effective control requires robust surveillance systems capable of detecting early warning signals, guiding rapid response, and ensuring efficient resource allocation. Traditional surveillance methods, such as larval indices, household surveys, and manual reporting, can provide useful insights, but they are frequently limited by delayed data flow, limited geographic coverage, and insufficient integration with health decision-making. These limitations highlight the critical need for smarter, more intelligent systems capable of bridging the gap between data collection and actionable public health strategies.

Currently, digital innovations are increasingly being used to strengthen surveillance frameworks. Mobile reporting platforms allow frontline workers to transmit data from communities in real time, reducing information flow delays. Molecular diagnostic tools, automated vector identification techniques, and climate-linked data modeling improve the accuracy and timeliness of identifying vector populations and potential transmission risks. The incorporation of GIS, remote sensing, and satellite imagery into surveillance systems enables the mapping of ecological determinants such as breeding habitats, rainfall, and vegetation indices, providing health officials with early warnings for targeted interventions.

Future tools are expected to enhance these capabilities via intelligent, interconnected systems. Smart traps equipped with biosensors, drones for habitat mapping, and Internet of Things devices will allow for continuous and large-scale monitoring. Predictive models based on artificial intelligence and big data analytics will not only predict outbreak risks, but will also aid in the development of cost-effective, location-specific interventions. Cloud-based platforms and interactive visualization dashboards will make complex data more accessible to decision-makers, thereby promoting evidence-based policies and timely community engagement.

The shift from traditional to intelligent surveillance marks a paradigm shift in vector-borne disease control. By combining current digital innovations with future-ready intelligent tools, health systems can improve preparedness, detect outbreaks earlier, and implement more equitable disease prevention strategies. Finally, smarter surveillance converts unstructured data into meaningful decisions, allowing for proactive control measures that protect communities from the growing threat of vector-borne diseases.

## **Community acceptance and perception of drone usage for vector surveillance and control: A descriptive study in Delhi**

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The usage of drones for control of vector borne diseases is a relatively new domain in usage of drones in public health. The knowledge, perception and acceptance of this newer technology by the community are of paramount importance for its successful testing and future applications. The present study aims to assess the community acceptance and perception regarding usage of drones for vector surveillance and its control. The study will be conducted on a pre-defined community in West Delhi using a pre-designed questionnaire using online tools. The sample size was estimated assuming 50% of the participants have a positive perception about the drone usage, and 95% confidence with 0.05% margin of error. The sample size thus achieved was 385. Descriptive and analytical statistics were used for the analysis of data using appropriate statistical tests and programs. The participants were generally aware of the mosquito borne diseases and control measures to be taken for prevention. However, there was a significant proportion of the population who had never heard of drone usage for vector surveillance and control. A few participants were sceptical regarding drone usage and the success thereof, for the intended purpose.

## Mosquito fauna and their seasonal dynamics in relation to vector-borne diseases in Delhi, India

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The growing threat of vector-borne diseases (VBDs), including malaria, dengue, and chikungunya, has become a pressing public health concern in urban areas like Delhi. Urbanisation and favourable mosquito breeding conditions exacerbate the issue. Understanding mosquito vector composition, seasonal dynamics, habitat alteration, distribution, and insecticide susceptibility is crucial for effective VBD control.

This study aims to conduct a comprehensive faunistic surveillance of mosquito species in Delhi, focusing on their diversity, distribution, and seasonal abundance. By analysing vector dynamics and seasonal trends, the study identifies peak transmission periods to support targeted vector control strategies.

Surveillance was conducted in six ecologically diverse sites: urban colony, industrial area, riverbank, area near the Aravalli Hills, resettlement colony, and an outskirt village. Sampling occurred during pre-monsoon (Mar-May), monsoon (Jun-Aug), post-monsoon (Sep-Nov), and winter (Dec-Feb). Adult mosquitoes were collected using methods such as resting collection, spray sheet collection, and light-trap collection. Identification of species was carried out using standard morphological keys.

A total of 6342 adult mosquitoes were collected across the four seasons from March 2024 to February 2025. The specimens belonged to three genera—*Culex* (49.93%), *Aedes* (32.84%), and *Anopheles* (17.12%)—comprising 11 species. Species composition and abundance showed clear seasonal variation, with higher species diversity and abundance during the monsoon and post-monsoon periods, while reduced activity was observed in the winter months.

This study highlights critical seasonal trends in mosquito ecology and the importance of targeted interventions. Ongoing vector incrimination and upcoming seasonal data will further elucidate vector dynamics and their connection to climate factors, guiding climate-resilient vector management strategies in Delhi.



## Changing behaviour of *Aedes* mosquitoes: A New Threat in Vector Control?

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Vector surveillance is essential for understanding local transmission risks and designing effective control strategies. In arid regions of Western Rajasthan, India, malaria vectors have traditionally received more research attention; however, *Aedes* mosquitoes are important due to their role in dengue, chikungunya, and Zika transmission. *Aedes* mosquitoes typically display anthropophilic and day-biting behaviours, making them less responsive to light traps used at night. During routine landing and light trap collection, *Aedes* mosquitoes were also collected during late night as incidental findings, suggesting possible changes in their behaviour.

Entomological surveys were conducted across selected districts in Western Rajasthan between December 2021 and November 2023. Mosquito collections were carried out using Light Trap collection and human landing catch methods. Collected mosquitoes were identified morphologically using standard taxonomic keys. Both malaria and *Aedes* species were recorded.

Light trap collections from Jhalawar, Sanchore, and Bikaner yielded *Aedes aegypti* and *Aedes vittatus*, whereas human landing catches from Barmer and Sanchore districts recorded the presence of *Aedes albopictus* from the rural area. *Aedes aegypti* mosquitoes were found in light trap collections during late night hours, despite their typical diurnal behaviour. Additionally, human landing collections confirm their active host-seeking behaviour. The simultaneous presence in both collection methods suggests possible behavioural plasticity influenced by artificial illumination, host availability, or microclimatic conditions in the arid environment.

The incidental detection of *Aedes* mosquitoes highlights an emerging change in behavioural patterns in Western Rajasthan. Their appearance in light trap catches indicates potential adaptation to late evening hour activity, while HLC collections reaffirm their human-biting tendencies. Future studies should systematically target *Aedes* surveillance using both methods to assess the behavioural adaptation, host dynamics, and climatic variation in shaping their behavior. Recognising these shifts is critical for strengthening vector surveillance and developing adaptive control measures against arboviral diseases in arid regions.

## Enhancing Diagnostic Accuracy in Vector Surveillance: Integrating Geometric Morphometrics in Medical Entomology to Differentiate *Aedes* (Diptera: Culicidae) Vectors

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Vector-borne diseases (VBDs) pose serious implications for human health and well-being presenting numerous challenges to global health security. Addressing these challenges necessitates a well-designed strategic surveillance plan that is efficient in terms of time, cost, and labour. Accurate identification of vectors prevalent in an area is crucial for understanding disease transmission dynamics and for implementing effective vector control programs. Geometric morphometrics (GM) is a reliable diagnostic tool that quantifies phenotypic variations based on landmark coordinates, enabling species identification including cryptic species complexes, intraspecific variations, and assessing population variations due to environmental effects and other factors. The two-dimensional structure of a mosquito wing, with its vein margins, cross-veins, and bifurcation points, facilitates the digitization of landmarks with reduced errors. The present study differentiated three vector mosquito species of *Aedes*—*Aedes aegypti*, *Aedes albopictus*, and *Aedes vittatus*—which are significant vectors of several deadly arboviral diseases using wing GM. The metric data generated after landmark digitization were processed to perform complex analyses, including Generalized Procrustes Superimposition (GPA), Principal Component Analysis (PCA), Canonical Variate Analysis (CVA), and Discriminant Function Analysis (DFA), which significantly differentiated these three species. The first two principal components of the PCA (PC1 = 32.94% and PC2 = 21.69%) together explained 54.64% of the variation in wing shape among the three species. The CVA resulted in three segregated clusters for each species. The pairwise cross-validation reclassification using DFA showed 95% accuracy. GM is an inexpensive technique for rapid and precise species identification across diverse geographical regions and mosquito populations. Thus, it holds great potential for application in vector surveillance and management strategies.

## Poster Presentation-63

## Environmental Information Awareness Capacity Building and Livelihood Programme (EIACP) Centre on Climate Change and Public Health

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The Environmental Information Awareness Capacity Building and Livelihood Programme (EIACP) Centre on Climate Change and Public Health is committed to understand and mitigate the impacts of climate change on public health, with a particular focus on vector-borne diseases. The Centre actively studies the interaction between climatic factors and disease epidemiology, providing real-time and evidence-based information to the public through its official website. As part of its initiatives, the Centre has meticulously developed comprehensive databases on vector-borne diseases, including a mosquito species identification database and a spatial database of mosquito-borne diseases. The mosquito species identification database provides critical insights into species-specific behaviors, enabling the design of targeted and effective intervention strategies. The spatial database contains detailed maps depicting the long-term spatial distribution and trends of various mosquito-borne diseases up to the present, offering a robust resource for tracking disease dynamics and identifying high-risk areas. These databases serve as foundational resource for research and analysis. To better understand the relationship between climate variability and disease transmission, the Centre has conducted detailed studies assessing how climatic factors such as temperature, rainfall, humidity, soil moisture influence the spread of these diseases. Advanced studies have been undertaken to project the risks of dengue and chikungunya under varying climate change scenarios based on Representative Concentration Pathways (RCP), providing critical insights into potential future health burden. Leveraging machine learning and deep learning techniques, the Centre has developed robust climate-based disease forecasting models tailored for Kerala, Delhi, and the Northeast region, of India. These models offer precise and actionable predictions, empowering public health officials to implement timely and targeted interventions to mitigate disease outbreaks. The Centre's research findings have been published in various newsletters and peer reviewed journals and earned citations from the global scientific community. Beyond research, the Centre has actively engaged in community outreach by organizing a series of awareness programs on the effects of climate change on public health, with a particular emphasis on vector-borne diseases. These initiatives aim to educate communities about preventive measures and enhance their resilience to the health impacts of a changing climate.

## **Contrasting Patterns of Mosquito Breeding Habitats across Urban, Peri-Urban and Rural Settings of Purulia District, West Bengal: Implications for Vector Control**

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Purulia district, situated entirely within the Chota Nagpur plateau, represents a distinct geo-morphological and ecological landscape of West Bengal. The district is endemic for malaria and lymphatic filariasis, with dengue cases also being reported in recent years. To characterize the ecology of mosquito breeding habitats in this unique setting, larval surveys were conducted across three contrasting environments: urban (Purulia Municipality Ward No. 10, Jhalda Municipality Ward No. 08), peri-urban (Purano Jhalda, Dulmi), and rural (Maguria, Balarampur). Approximately 60–70 households from each setting, along with their surrounding environments, were surveyed at bi-monthly intervals for one year. Containers were classified as natural or man-made and further grouped as domestic, peri-domestic, or outdoor. Analysis of larval indices revealed pronounced seasonal variation, with House Index (HI) and Container Index (CI) reaching their highest values during September–October, followed by July–August. Peri-domestic containers consistently contributed the maximum proportion of positive habitats across all settings throughout the year. Domestic containers showed significant increases during monsoon and post-monsoon in urban and peri-urban areas, but remained relatively low in rural sites. Scattered thunderstorms in summer months also generated a substantial number of positive containers. Natural habitats—including pond edges, rivulets, stone pockets of streams, culverts, slow-running streams, and unmanaged drainage systems—emerged as major breeding grounds during dry and summer seasons, with the potential to sustain high vector densities. These findings demonstrate that mosquito breeding in Purulia follows distinct spatial and temporal patterns shaped by ecological and climatic conditions of the plateau region. Vector control in such endemic areas must therefore adopt season-specific and habitat-targeted interventions. Pre-monsoon cleaning and larviciding can reduce vector build-up, while intensified larval source management during monsoon and post-monsoon should focus on peri-domestic and man-made containers. Tailored strategies based on ecological settings can substantially strengthen vector management in Purulia and comparable plateau districts.

## **Towards Comprehensive Malaria Control: Entomological Insights and Future Prospects For The Daman Programme.**

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Malaria, caused by Plasmodium parasites and transmitted by female Anopheles mosquitoes, remains a major public health threat. Vectors acquire the parasite from infected individuals (host) and later transmit it to healthy individuals. The World Health Organization (WHO) recommends integrated malaria control through early case detection, prompt treatment and effective vector control measures to reduce the spread of the disease. In Odisha, the National Vector Borne Disease Control Programme (NVBDCP) launched a program acronymic DAMaN (locally translating to Durgama Anchala re Malaria Nirakarana), which aimed to strengthen malaria surveillance in hard-to-reach endemic areas with rough terrain. The malaria cases in such villages often went undetected due to sub-optimal surveillance and the presence of many asymptomatic and afebrile cases, which act as focal points for ongoing transmission. DAMaN's strategy included rapid detection and diagnosis of the disease by Mass Screening And Treatment (MSAT), distribution of Long-Lasting Insecticidal Nets (LLINs) and robust community mobilization by field-level workers.<sup>1</sup> With the adoption of these interventions, there was a decline in malaria cases, accompanied by a significant reduction in cases in high-endemic areas.<sup>2</sup> However, to ensure the program aligns with the WHO's Global Technical Strategy for Malaria (2016-2030) target of zero indigenous cases by 2030,<sup>3</sup> further improvements are needed, especially in vector surveillance and management. A major gap is the absence of an initial baseline entomological data, which are required to characterize the local vector species composition, detect sibling species and assess climate change impact on vector biology.<sup>4</sup> Studies on vector behaviour need to be strengthened to understand their biting behaviour, host-seeking patterns and resting preferences for the optimization of LLIN deployment. Studies on possible insecticide resistance also need to be taken up, to prevent failure of current interventions.<sup>5</sup> Incorporating Malaria Molecular Surveillance (MMS) tools, could strengthen DAMaN's capacity in tracking insecticide and drug resistance, characterizing and mapping transmission chains, and detecting the sub-patent infections thereby addressing the hidden asymptomatic reservoir sustaining transmission.<sup>6</sup> A comprehensive entomological framework integrated with advanced molecular surveillance will ensure that DAMaN not only retains recent success but also advances the state towards total malaria elimination.

## Marine extremophile bacteria: a comprehensive approach to eco-friendly mosquito control

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Mosquitoes are primary vectors of several life-threatening diseases, posing persistent global health challenges. Conventional insecticides, though widely employed, have progressively lost effectiveness due to the emergence of resistant mosquito populations, highlighting the urgent need for sustainable alternatives. Interestingly, many mosquito species, have shown the ability to adapt to brackish and saline water habitats, indicating a degree of salt tolerance in their larval stages. This ecological adaptability further complicates control measures in coastal and mangrove-associated regions. In this study, salt-tolerant bacterial isolates were isolated from marine soil samples collected from the Sundarbans, West Bengal, India. Among them, *Halobacillus marinus* MB201 was identified as the most potent strain with strong larvicidal potential against three medically important mosquito species: *Aedes albopictus*, *Culex quinquefasciatus*, and *Anopheles stephensi*. Toxicity bioassays revealed LC<sub>50</sub> values of 1.868 mg/L, 2.709 mg/L, and 3.676 mg/L, respectively, demonstrating broad-spectrum efficacy. Morphological characterization using scanning and transmission electron microscopy confirmed MB201 as a rod-shaped, Gram-positive bacterium. Liquid chromatography-mass spectrometry (LC-MS) analysis identified key metabolites including nigakilactone F, nicotine, and notably batrachotoxin. The latter, a well-documented modulator of voltage-gated sodium channels, emerged as the principal factor inducing larval paralysis and subsequent mortality. Supporting this, *in-silico* molecular docking studies showed strong interactions between batrachotoxin and sodium channel proteins, with a binding energy of -9.2 kcal/mol, thereby confirming its neurotoxic mechanism. Importantly, biosafety assessments using lecithinase and hemolysis assays indicated no pathogenic traits, underscoring MB201's safety for non-target organisms and humans. Its salt tolerance further enhances its applicability in vector-prone, saline-affected environments. Collectively, these findings establish *H. marinus* MB201 as a promising, eco-friendly, and effective biological larvicide. The study not only underscores the potential of extremophilic marine bacteria in vector control but also highlights their relevance under climate-driven ecological shifts, where conventional strategies may prove inadequate.

## Mosquito fauna and their seasonal dynamics in relation to vector-borne diseases in Delhi, India

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The growing threat of vector-borne diseases (VBDs), including malaria, dengue, and chikungunya, has become a pressing public health concern in urban areas like Delhi. Urbanisation and favourable mosquito breeding conditions exacerbate the issue. Understanding mosquito vector composition, seasonal dynamics, habitat alteration, distribution, and insecticide susceptibility is crucial for effective VBD control.

This study aims to conduct a comprehensive faunistic surveillance of mosquito species in Delhi, focusing on their diversity, distribution, and seasonal abundance. By analysing vector dynamics and seasonal trends, the study identifies peak transmission periods to support targeted vector control strategies.

Surveillance was conducted in six ecologically diverse sites: urban colony, industrial area, riverbank, area near the Aravalli Hills, resettlement colony, and an outskirt village. Sampling occurred during pre-monsoon (Mar-May), monsoon (Jun-Aug), post-monsoon (Sep- Nov), and winter (Dec-Feb). Adult mosquitoes were collected using methods such as resting collection, spray sheet collection, and light-trap collection. Identification of species was carried out using standard morphological keys.

A total of 6342 adult mosquitoes were collected across the four seasons from March 2024 to February 2025. The specimens belonged to three genera—*Culex* (49.93%), *Aedes* (32.84%), and *Anopheles* (17.12%)—comprising 11 species. Species composition and abundance showed clear seasonal variation, with higher species diversity and abundance during the monsoon and post-monsoon periods, while reduced activity was observed in the winter months.

This study highlights critical seasonal trends in mosquito ecology and the importance of targeted interventions. Ongoing vector incrimination and upcoming seasonal data will further elucidate vector dynamics and their connection to climate factors, guiding climate-resilient vector management strategies in Delhi.



## Insecticidal Paint: An alternate integrated vector management strategy for mosquito control

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Vector-borne diseases (VBDs) remain one of the most significant public health challenges globally, contributing to high morbidity and mortality. They account for approximately 17% of the total global burden of communicable diseases, resulting in around 0.7 million deaths annually. The primary vectors responsible for transmitting these diseases include mosquitoes from the genera *Anopheles*, *Aedes*, and *Culex*, carriers of debilitating diseases such as malaria, dengue, chikungunya, Zika, and filariasis in tropical and subtropical regions.

Effective management of VBDs hinges significantly on vector control strategies. Among the available interventions, chemical control using insecticides has been a cornerstone approach due to its efficacy in reducing vector populations and minimizing disease transmission. However, the alarming increase in insecticide resistance among mosquito populations compromises the long-term sustainability of these measures. In this context, innovative strategies that address resistance while ensuring safety and efficacy are imperative.

Insecticidal paints offer an innovative, sustainable solution for vector control, aligning with the Global Vector Control Response principles. These paints, infused with insecticides, create long-lasting protective barriers against disease-carrying vectors when applied to walls and surfaces. With residual efficacy lasting more than 24 months, they minimize the need for frequent reapplications, ensuring cost-effectiveness. Their ease of use—similar to traditional paints—promotes widespread acceptance in homes, schools, and public facilities. Versatile and efficient, insecticidal paints provide a user-friendly, broad-spectrum defense against multiple vector species, supporting research and innovation in disease prevention while addressing community health needs.

Our study demonstrated the efficacy of nano paint infused with alphacypermethrin, a synthetic pyrethroid, offering over 24 months of protection against key mosquito vectors: *An. stephensi*, *Ae. aegypti*, and *Cx. quinquefasciatus*. Nano-encapsulation ensures controlled release, prolonging effectiveness and reducing resistance risks. Safe for non-target organisms, this formulation addresses ecological concerns. Insecticidal paints provide a sustainable, cost-efficient alternative to traditional vector control, targeting vectors at resting sites to reduce density and disease transmission. By minimizing reliance on routine spraying, they support sustainable development goals and offer a long-term solution to vector-borne diseases.



## Frugal Innovation: Application of Origami-Based Foldscope in Integrated Pest Management and Sustainable Insecticide Use

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The emergence of low-cost, portable diagnostic tools offers significant use for enhancing pest surveillance and pesticide application in Integrated Pest Management (IPM). This study investigates the use of an origami-based paper microscope, the *Foldscope*, as a field tool for early detection, monitoring, and assessment of insect pests and plant pathogens with implications for more precise insecticide implementation (1). The Foldscope, capable of magnifications in the range of  $\sim 140\times$  to  $\sim 2000\times$ , enables farmers and extension agents to identify pest species, life stages (larvae, eggs), and associated pathogen structures in situ, without the need for elaborate lab infrastructure. The study assesses how such early detection can influence action under IPM—thus reducing unnecessary insecticide applications, lowering costs, and minimizing ecological and health risks associated with chemical control. The use of Foldscope for monitoring allowed quicker detection of infestation onset by several pest types (including aphids and lepidopteran larvae), enabling timely intervention. Comparative plots under traditional monitoring vs. Foldscope-aided monitoring showed 25–40% reduction in insecticide use, with no significant yield loss (2). This work also discusses limitations such as the requirement for minimal training, lighting, mounting of samples, and durability under harsh field conditions, and proposes integration into farmer training and extension services. Foldscope's role as a frugal, scalable sensor in IPM systems offering benefits for sustainable pest control and responsible insecticide use.

## Molecular Taxonomy of Prevalent Mosquito Vectors in Odisha Via DNA Barcoding.

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Mosquitoes are important vectors of various life-threatening diseases, including malaria, dengue, chikungunya, and filariasis, causing significant public health challenges globally. In Odisha, with diverse ecosystems, the identification and monitoring of mosquito species are crucial for vector control and disease prevention. Traditional morphological identification of mosquitoes is often constrained by the need for expert taxonomists, the rubbing-off of the scales from their body surface and the morphological similarities among species. DNA barcoding, a molecular tool utilizing a specific standardized fragment of the mitochondrial cytochrome c oxidase I (COI) gene, offers a reliable, efficient, and accurate approach for species identification. This study focuses on the DNA barcoding of mosquitoes prevalent in various ecological zones of Odisha. Field surveys were conducted to collect mosquito specimens, followed by morphological identification to the genus level. Genomic DNAs were extracted from individual specimens, and the COI region was amplified using polymerase chain reaction (PCR). Sequencing of the amplified products by the Sanger sequencing method, subsequent analysis by using Mega-X software facilitated species-level identification and phylogenetic assessments. Preliminary results have revealed the presence of several key vector species, including *Anopheles*, *Aedes*, and *Culex*. The integration of DNA barcoding with traditional taxonomy has enhanced the resolution of identification of species that play a role in disease transmission. These findings underscore the importance of molecular tools in vector surveillance programs and contribute to the development of targeted vector control strategies in Odisha. This study emphasizes the utility of DNA barcoding as a cornerstone in the integrated vector management framework, enabling precised identification and monitoring of mosquito species to mitigate vector-borne diseases effectively.

## Rethinking Arboviral Transmission: Emerging Roles of Secondary Vectors

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Vector-borne diseases remain a major public health challenge, with *Aedes aegypti* traditionally recognized as the primary driver of dengue, chikungunya, and Zika transmission. However, ecological shifts, climate variability, and urbanization are reshaping vector dynamics, enabling secondary vectors to assume increasingly important roles. *Ae. aegypti* thrives in indoor habitats, ensuring perennial survival in urban landscapes, while *Ae. albopictus*, with its ecological plasticity and global dispersal, dominates outdoor environments and is becoming a co-primary vector in many regions. The recent molecular and ecological investigations on *Ae. vittatus* in Indian metropolitan settings, after decades of absence highlights its re-emergence which is also a potential arbovirus vector. Evidence of novel haplotypes, shared lineages across continents, and strong gene flow suggests that globalization and trade facilitate introductions and expansions of these species. Ecological analysis revealed perennial presence of *Ae. aegypti* in both indoor and outdoor habitats, *Ae. albopictus* populations peaking post-monsoon with dominance in outdoor sites, and seasonal detection of *Ae. Vittatus* breeding close to human dwellings. This study underscore the dynamic interplay between primary and secondary vectors underscores the fluidity of vectorial hierarchies, challenging conventional surveillance and control programs that remain largely *Ae. aegypti*-centric. Understanding the ecological preferences, genetic diversity, and adaptive mechanisms of these three vectors is critical for predicting outbreak patterns and designing integrated strategies that target both indoor and outdoor breeding habitats.

## Impact of Ivermectin exposure on larvae and adult *An. stephensi* and *An. culicifacies*

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Increasing insecticide resistance and behavioral shifts in malaria vectors necessitate novel control strategies. This study determined the potential of ivermectin as a contact toxin against *An. stephensi* and *An. culicifacies*.

Laboratory-reared late third stage larvae and adult female mosquitoes were exposed to varying concentrations of ivermectin through larval, topical, and bottle bioassays. Mortality data were used to estimate LD<sub>50</sub> and LD<sub>90</sub> values for each species. Ivermectin caused complete larval mortality at higher concentration within 24 hours whereas at lower concentration within 48 hours. In adult mosquitoes, ivermectin showed strong dose-dependent toxicity in both *Anopheles stephensi* and *An. culicifacies*. Topical and bottle bioassays revealed that *An. culicifacies* was slightly more susceptible compared to *An. stephensi*. Mortality increased rapidly with dose, resulting into 100% within 24-48 hours at higher concentrations.

Ivermectin demonstrates potent contact toxicity against major malaria vectors. These results support its potential as a dual-action tool i.e. killing mosquitoes and potentially reducing malaria transmission.

## Strengthening Vector Control and Community Engagement to Combat Dengue and Chikungunya in Koraput, Odisha: Insights from Entomological Surveillance

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Dengue and Chikungunya have emerged as major public health concerns in India, with their transmission dynamics closely linked to the abundance and distribution of *Aedes* mosquito vectors. In view of the reported arboviral cases in Koraput district, Odisha, an entomological surveillance study was conducted to assess the presence of vectors, breeding indices, and insecticide susceptibility status. Systematic larval surveys were conducted across affected and peripheral localities. Both *Aedes aegypti* and *Ae. albopictus* were detected, indicating the coexistence of primary and secondary vectors in the region.

The House Index (HI), Container Index (CI), and Breteau Index (BI) exceeded the WHO recommended thresholds, indicating a high transmission potential and an elevated risk of arboviral outbreaks. Insecticide susceptibility assays performed on *Ae. aegypti* populations demonstrated resistance to organophosphate and pyrethroid insecticides, suggesting reduced efficacy of routinely used vector control interventions.

These findings underscore the urgent need to strengthen vector management strategies by adopting an Integrated Vector Management (IVM) framework that combines chemical, biological, and environmental approaches. Enhanced Information, Education, and Communication (IEC) initiatives promoting community participation and behavioural change are equally vital in reducing breeding sites and improving public awareness of dengue and chikungunya prevention. Sustained entomological surveillance and resistance monitoring are crucial for preventing future outbreaks and supporting resilient, evidence based vector control programmes in Koraput.

## Altered cuticle protein profiles associated with insecticide resistance in *Anopheles stephensi*

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Reduced malaria cases has been mainly attributed to the use of vector control interventions which include the use of insecticides. The mosquito cuticle has been hypothesized to aid insecticide resistance by reducing or slowing insecticide uptake. Cuticle proteins (CPs) play a major role in insecticide resistance in mosquitoes by affecting the penetration of the insecticides, thereby reducing effectiveness of the insecticides. Several CP families (CPR, CPAPn, CPXL, CPF and CPLCG) belonging to different sub - group protein families in cuticle have been identified, yet their role in Indian malaria vectors remains poorly understood. To determine the role of cuticle proteins in deltamethrin resistance development in *An. stephensi*.

In this study, insecticide susceptibility assay was performed using WHO's adult mosquito insecticide susceptibility test on F1 adult female *An. stephensi* against the deltamethrin insecticides. Total RNA was extracted using the Trizol method for both resistant and susceptible mosquitoes followed by cDNA preparation. Expression was studied by the qRT-PCR analysis.

Expression of cuticular genes CPR127, CPR63 and CPLCG5 was determined using gene specific primers. Amplified products were visualised on gel. Further, real time-PCR results analysis has confirmed the differential expression levels of the genes in resistant compared to susceptible female mosquitoes.

Understanding the differences in expression of these CPs in the cuticle of resistant and susceptible mosquitoes will highlight their role in the development of insecticide resistance by vectors and may facilitate the development of insecticides with greater specificity and potency.

## Unveiling Drug Targets in *Plasmodium falciparum* 3D7: A Computational Approach

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To find potential therapeutic targets in *Plasmodium falciparum* 3D7, we studied 2,670 essential genes listed in the Database of Essential Genes (DEG). BLAST (Basic Local Alignment Search Tool) program was used to screen for the key human non-homologous genes of the parasite. The metabolic processes and pathways of these genes were further examined using the Panther and Kyoto Encyclopedia for Gene and Genome (KEGG) database. Five important genes were found through this investigation. No authorized or investigational medications that target the enzyme pyridoxal 5'-phosphate synthase subunit (pdx2) were found by a subsequent search in the Drug Bank database. A subsequent search in the Drug Bank database revealed no existing drug molecules for the enzyme pyridoxal 5'-phosphate synthase subunit (pdx2) are available. Protein-protein interaction network prediction and literature review indicated that pdx2 plays a critical role in the biosynthesis of pyridoxal 5'-phosphate (PLP) in the parasite, highlighting its potential as a therapeutic target.

## Phytochemical-Based Drug Discovery for DENV RNA Helicase Inhibition: Insights from Molecular Docking and Dynamics

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Dengue is the most important emerging viral disease of human beings in the world today. A day time mosquito, *Aedes aegypti*, is the principal vector in India. The objective of proposed study to design and screen a potential phytochemical by using in silico method against ns3 of DENV. A total of 26 derivative of phytochemical Gedunin were selected as ligand which retrieved from Pubchem database. Tetrahydrogedunin was shown to be the most promising drug by evaluating its physicochemical features, toxicity analysis, and docking scores. Based on its molecular structure, 100 drug-like derivatives were designed by Data Warrior software. Using the SwissADME and PreADMET servers, the drug-likeness and absorption, distribution, metabolism, excretion, and toxicity (ADMET) of these phytochemicals were also predicted. Derivative 42 showed effective druglikeness based on the ADMET and drug-likeness prediction results. Based on the docking study derivative number 42(-10.0kcal/mol) was selected exhibiting high binding affinity when compared to standard drug ST-610 (-9.6kcal/mol). The structural stability was evaluated using molecular dynamics simulation between derivative 42 and the RNA helicase enzyme by 50-nanosecond along with MM-GBSA analysis. Proposed study also exploits the novel inhibitory action of derivative 42 against DENV RNA helicase enzyme.



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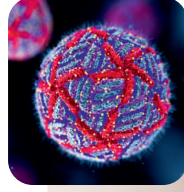
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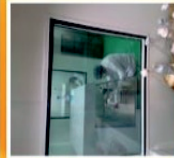
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