Chapter 4

# Bionomics and control of *Anopheles (Cellia) minimus* in the context of disappearing malaria in India

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**Abstract**

Malarial threat is receding in India and for achieving malaria-free status, vector biology is regaining its due importance for targeting species-specific interventions in place and time. *Anopheles minimus*, the major vector of malaria in north-east India, is disappearing fast evidenced by reduced levels of malaria transmission and morbidity. *An. minimus* is a species complex comprising three sibling species of which *An. minimus s.s.*, formally named as *An. minimus*, is recorded to occur in east and north-east India. It is highly anthropophilic and responsible for fulminating outbreaks of malaria evidenced by incrimination records in range of its distribution. For its control, DDT continues to be the insecticide of choice for indoor residual spraying; however, populations are highly resilient in response to residual insecticides for paradigm shift in vector behaviour from indoor resting to outdoors resulting in continued transmission. Extra-domiciliary transmission of malaria is a challenge for which newer interventions, viz., attractive toxic sugar bait, eave-tube technology, nano-synthesized pesticides, new adult repellents, oviposition deterrents need to be field-evaluated under local geo-epidemiological conditions. To keep vector populations at bay, it is advocated to upscale interventions for 'universal coverage' of human populations at risk to check malaria transmission and spread of drug-resistant malaria.

**Keywords:** Anopheles, malaria elimination, sibling-species, bionomics, vector control, outdoor transmission, India

## Introduction

India is reporting declining transmission of malaria and targeting elimination by 2027 – three years ahead of global target date of 2030 [1-4]. Given the mandate, vector biology is regaining its lost ground for being an integral component of the national control strategy. A lot of new information has been generated on malaria vectors, aided by molecular taxonomy tools, including sibling-species composition, distribution and disease transmission relationships helping formulate species-specific control interventions [5, 6]. Amidst myriad of challenges including climate change, deforestation, population migration, cross-border population movement, galloping urbanization, paradigm shift in vector behaviour, and growing menace of insecticide resistance; monitoring vector abundance and bionomical characteristics in the altered ecology has become important in different eco-epidemiological zones of the country. Amongst dominant vectors of malaria in India, *Anopheles minimus* is reckoned as the major vector in north-east region contributing ~5% of the recorded positive cases in the country annually [7]. History is replete with records of devastating malaria outbreaks characterised by high rise in cases and deaths across all age groups attributed to this dreaded vector [8-10]. Due to its medical importance, it has been the subject of extensive investigations and review for its bionomical characteristic both in pre-independent and post-independent India for formulating intervention strategies. It has become increasingly clear that containment of populations of this vector species is of paramount importance to forbid entry and spread of drug-resistant malaria and check ongoing transmission specific to northeast India (the gateway to Southeast Asia). Included in this chapter are the bionomical characteristics of this species (in brevity), highlighting significance for sustained interventions for keeping its populations below threshold for malaria elimination at national/sub-national level.

## Taxonomic considerations & distribution

*Anopheles* (*Cellia*) *minimus s.l*. is an Oriental species and has been genetically characterised to be a complex of three designated formally named species namely *An. minimus* Theobald, *An. harrisoni* Harbach & Manguin, and *An. yaeyamaensis* Somboon & Harbach with distinct bionomical characteristics and distribution records (Figure 1) [11, 12]. Of these, exclusively *An. minimus* is encountered in north-east India with records of its disappearance and re-appearance after decades in erstwhile domains of its distribution including eastern state of Odisha (formerly Orissa) [13, 14]. It is a small sized mosquito that is morphologically similar to sympatric populations of *An. varuna* and requires an experienced eye for its correct identification due to subtle morphological differences (Figure 2) [8, 15], but can be identified by molecular tools unequivocally [11, 12]. Aided by molecular tools, earlier identified populations of *An. fluviatilis*, prevalent during winter months (November – March) in Assam, are now characterised as hyper-melanic seasonal variant of *An. minimus* with history of malarial outbreaks in hill ranges (~3000 feet above mean sea level) [16].

## Seasonal abundance, infectivity and disease transmission

*An. minimus* is recorded throughout the year, however, its density is seen rising beginning March/April with the onset of pre-monsoon showers and recorded to occur in good numbers till cessation of rainy season in September/October; it occurs in low numbers during winter months (November–February) [10, 13, 17]. The disease transmission is perennial and persistent that follows suit with rising density of *An. minimus* beginning April/May marked with maximum malaria cases occurring during May till September/October (months of heavy rainfall), while the rest of the year is the period of low transmission season. *An. minimus* mosquitoes were repeatedly incriminated for most part of the year evidenced by detection of live sporozoites in salivary glands, with an overall infectivity rate of about 3% (Table 1) [13, 17]. The relative risk of infection, however, varied across landscape, the highest being in forest-fringe foothill villages in closer proximity to mosquito breeding sources and the minimal to healthcare facility within <5 km [18]. In these villages, all age groups of both sexes were recorded parasite positive mostly for *Plasmodium falciparum* (70%), the remaining were *P. vivax* cases (data not shown).

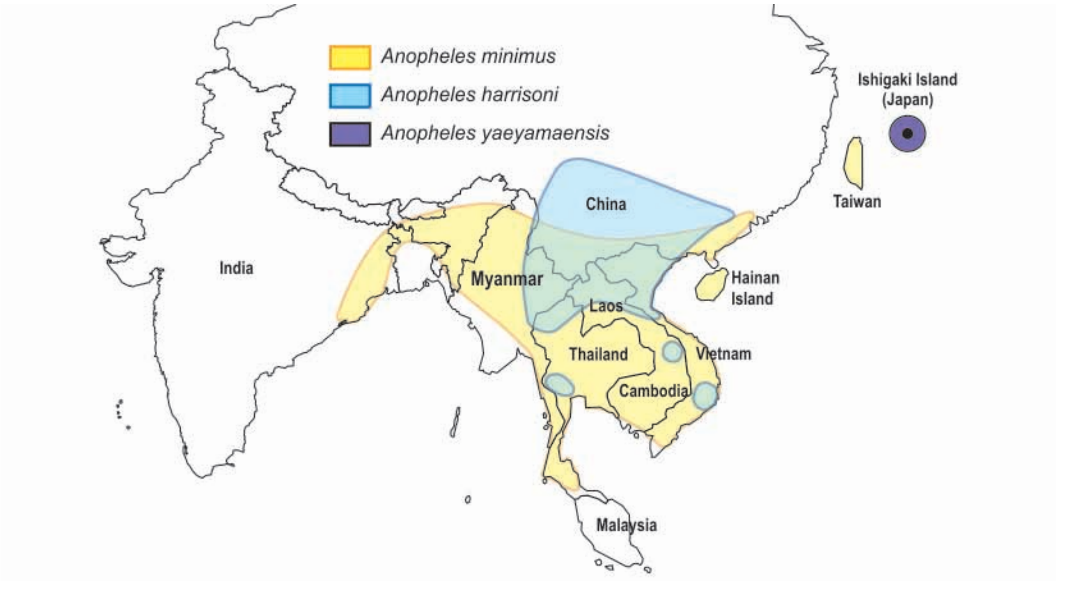


**Figure 1:** Distribution map of sibling species of the *Anopheles minimus* complex in Southeast Asia based on molecular identification. *Anopheles minimus* has a wide distribution extending from eastern to north-east India and eastwards to Myanmar, Thailand, Laos, Vietnam, Cambodia, China including Taiwan and occurs in sympatry with *Anopheles harrisoni* over large areas in southern China, Vietnam, Laos and Thailand. *Anopheles yaeyamaensis* is restricted to the Ishigaki Island of the Ryukyu Archipelago of Japan. Populations of *Anopheles minimus* are fast depleting in north-eastern states of India and seemingly have disappeared from Bangladesh [sketch map not necessarily in conformity with political boundaries].

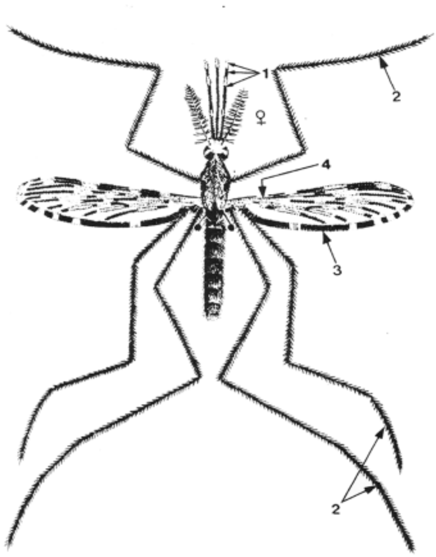


**Figure 2:** *Anopheles minimus* Theobald 1901, adult morphological distinguishing characters: (1) Apical and sub-apical pale bands equal separated by a dark band, (2) Tarsomeres without bands, (3) Fringe spot absent on wing vein 6, (4) Presence of pre-sector pale spot and humeral pale spot on the costa. These characters do not allow identification of the sibling species of the *An. minimus* complex. Source Reference [8]

**Table 1. Seasonal sporozoite infection rate in *Anopheles minimus* in the Sonapur Primary Health Centre of Dimoria Block in Kamrup district of Assam, north-east India for data based on 1989-1991. Source Reference [13]**

| Month, year | No. mosquitoes dissected for salivary glands | No. gland positive for sporozoites | Infection rate (%) |
|---|---|---|---|
| **1989** | | | |
| July | 802 | 39 | 4.86 |
| August | 284 | 12 | 4.22 |
| September | 386 | 11 | 2.85 |
| October | 114 | 8 | 7.02 |
| November | 38 | 0 | 0 |
| December | 28 | 0 | 0 |
| **1990** | | | |
| January | 106 | 1 | 0.94 |
| February | 223 | 3 | 1.35 |
| March | 282 | 2 | 0.71 |
| April | 217 | 2 | 0.92 |
| May | 328 | 11 | 3.35 |
| June | 172 | 7 | 4.07 |
| July | 52 | 1 | 1.92 |
| August | 25 | 0 | 0 |
| September | 5 | 0 | 0 |
| October | 94 | 8 | 8.51 |
| November | 227 | 7 | 3.08 |

| | | | |
|---|---|---|---|
| December | 182 | 3 | 1.65 |
| 1991 | | | |
| January | 112 | 0 | 0 |
| February | 38 | 1 | 2.63 |
| March | 96 | 0 | 0 |
| April | 245 | 7 | 2.86 |
| May | 334 | 13 | 3.89 |
| June | 245 | 17 | 6.94 |
| August | 15 | 1 | 6.67 |
| September | 30 | 1 | 3.33 |
| Total | 4680 | 155 | 3.31 |

## Host choice, resting and breeding characteristics

*An. minimus* is a formidable foe in maintaining hyper-endemic malaria throughout north-east India where it shows a strong predilection for human host (anthropophilic index >90%) making it an efficient vector substantiated by sporozoite infectivity for all months of the year ranging from anywhere from <1% to 7% (Table 1) [13, 17]. All night mosquito landing catches revealed that it actively searched for a host throughout the night beginning 19:00 hours with pronounced feeding activity during 01:00–4:00 hours that ceased at break of the dawn. The mosquito biting rate per person night (02–23) and entomologic inoculation rates (0.12–0.71) varied between locations representative of low-to-moderate transmission intensities [18]. It is largely an endophilic and endophagic (resting and feeding indoor human dwellings) species recorded resting indoors often in lower half of the walls in darker corners of the house (much away from sunlit areas) and seen exit at break of the dawn. Houses made of split-bamboo with thatched roofing is the preferred resting habitat opposed to RCC (Reinforced Cement Concrete) structures [19]. The flight range of this mosquito is estimated to be just about one km resulting in focal disease outbreaks often with case clusters in given locality yielding more than one case per household (Figure 3). It is a perennial species breeding in slow-flowing foothill seepage water streams in all seasons/months [20]. Larvae of *An. minimus* are recorded breeding along the shaded grassy banks sans sunlit areas in good numbers for most part of the year including winter season both in hills (~3000 feet above mean sea level) and valleys (Figure 4).

**Figure 3:** Top: Resting habitat - typical housing made of split bamboo and thatched roofing; mosquitoes are collected in good numbers inside house premises in dark/ shady corners resting on clothes, umbrella and other articles. Bottom: Infants and children are the vulnerable population groups with record of multiple cases within single household.

**Figure 4:** Top: A typical village hamlet in the forest-fringe at risk of malaria transmitted by *Anopheles minimus* in north-east India. Bottom: Typical larval habitat of *Anopheles minimus* in foothill perennial seepage water stream marked with grassy banks. Households located nearer to breeding habitat (<1 km) are at greater risk of malaria

## Vector Control

*An. minimus* is reported to be highly susceptible to DDT despite decades of its application in areas of its occurrence over space and time [10]. It has innate abilities evading exposure (behavioural resistance) to the sprayed surfaces and tend to maintain extra-domiciliary transmission by changing its resting habitat from indoors to outdoors. Due to repeated applications of DDT during 1960s, it was believed to have disappeared, but resurfaced decades apart (due to inadequate spray coverages years together) resulting in fulminating outbreaks across its range of distribution [21]. The advent of insecticide-treated netting materials, i.e., long-lasting insecticidal nets (LLINs) proved to be big boon for its effectivecontrol disrupting transmission. Based on field evaluation of this technology, LLIN-based intervention was held appropriate against *An. minimus* transmitted malaria and well received by the communities and programme official alike [22]. With the increasing coverage of LLINs, the populations of *An. minimus* are once again depleting in erstwhile domains of its distribution corroborated by evidences of reducing levels of transmission [10,23]. The populations of this species are presently at lowest ebb and scarce restricted to remote areas rendered inaccessible due to some logistic reasons, viz., recurrent flash floods, poor communication, insurgency in the preceding years. There is strong body of evidence that the niche, thus, vacated by *An. minimus* is being accessed by *An. culicifacies s.l.*, which is multi-resistant to available insecticides including pyrethroids [24-26]. It is the high time to ensure blanket coverage of the populations at any risk by appropriate interventions to ward off this vector species below threshold.

## Priority areas of research

Vector control programme in India largely rests on indoor residual spraying (IRS) and distribution of LLINs in communities most at risk. The largest challenge in vector control, however, is the emerging paradigm shift in mosquito behaviour towards outdoor transmission [27]. Mosquitoes are shifting outdoors in response to application of indoor residual insecticides rendering less amenable to control interventions [28]. *An. minimus* is one such classic example stymieing the control authorities by phenomenon of disappearing and re-appearance decades apart. Little is known of its outdoor population resting characteristics, which needs to be investigated for spearheading interventions. North-east is experiencing rapid ecological changes on account of population migration, deforestation and developmental project activities permitting sub-structuring of vector populations. *An. minimus* is proven genetically diverse evidenced by nucleotide diversity suggesting population expansion and possible existence of other sibling species with obvious implications for control interventions [29, 30]. There is an urgent need to devise appropriate technologies that are community-based and doable to capture residual as well as outdoor resting populations. Development of newer interventions like attractive toxic sugar baits, eave-tubes technology, nano-synthesized pesticides, new adult repellents, oviposition deterrents; all need to be evaluated in local ecological transmission settings possibly as component of integrated vector management for containment of vector populations [31]. Equally important would be to prioritize: (i) vector surveillance (that remained neglected), which should be the cornerstone activity to check unusual build-up of vector density [32],

(ii) monitor current insecticide susceptibility status, (iii) ecological succession by other vector species and their bionomical characteristics, (iv) promote information, education and communication activities for enhanced community participation and compliance, and above all (v) seek political commitment for sustained allocation of resources for 'universal coverage' of interventions to end malaria for good. For malaria elimination in India, it is of utmost importance to invest heavily strengthening interventions in north-eastern corridor for reducing populations of this vector species at sub-optimal levels averting impending disease outbreaks and spread of drug-resistant malaria.

## Conclusions

*An. minimus* is an invincible mosquito vector with characteristics of its innate ability to avoid sprayed surfaces and establish extra-domiciliary transmission. However, with the available current tools for vector control, *An. minimus* is seen fast disappearing in north-east India. Nevertheless, for outdoor transmission control (an emerging paradigm in Southeast Asia), field-evaluation of newer interventions targeting residual populations and spotting the residual malaria foci should be the priority [33]. What concerns most is the ecological succession by yet another vector, *An. culicifacies s.l.*, which is multi-resistant to available arsenal of insecticides. We strongly advocate 'vector surveillance' and 'universal coverage' of evidence-based interventions for control of vector populations to defeat malaria.

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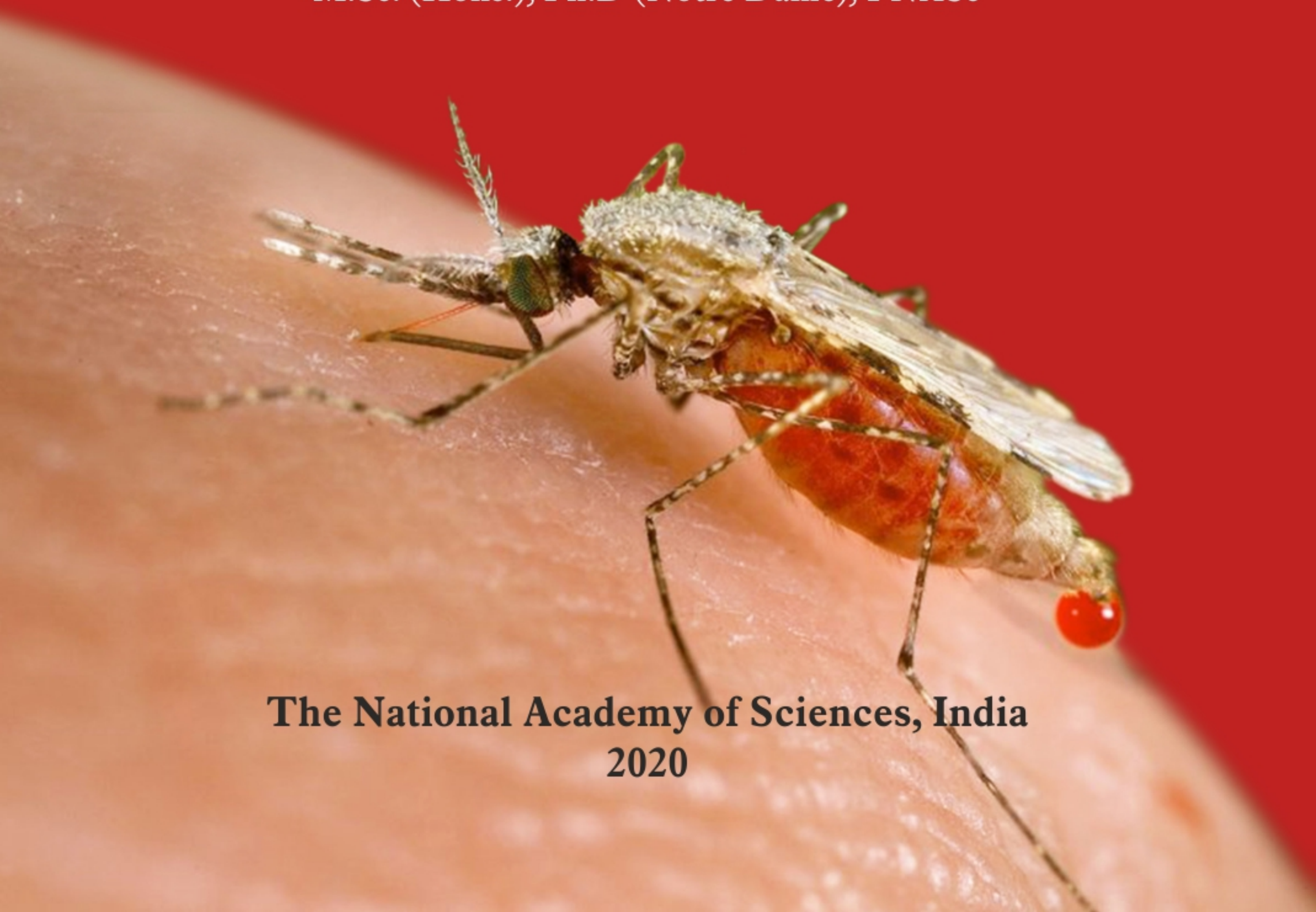

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