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ARE MALE MOSQUITOES WORTH THEIR WEIGHT? A MOSQUITO'S EYE VIEW

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*"The female is large and notorious,
She is progenitor of the race, yes,
She is also lauded for her prowess to infect,
With infirmity to countless men and women, and infants, too,
And, yet, all of her survival and glory,
Hinge along one singular, smaller Y – her better half, the male mosquito."*

The world is full of countless different species of animals, ranging from single-celled invertebrate protozoans to multicellular vertebrate mammals. The whole animal kingdom is organized into invertebrates (3-30 million species) and vertebrates (47,000 species); the former constituting over 95% of all animals on the earth. Invertebrates are important in the functions and processes of most ecosystems. They are spectacular, abundant and diverse. Over 80% of all invertebrates are grouped into the single phylum Arthropoda that includes insects, spiders, crustaceans, centipedes and millipedes. Insects, arachnids, and other terrestrial arthropods are important because together they comprise at least 75% of all the species of animals in the world now known to science. Estimates of the actual number of insects and related forms now living possibly range from 3-30 million, although many different views exist on their estimates; therefore, whatever the total, a great deal of scientific information on life comes only from invertebrates or more precisely the arthropods, particularly the insects. The dominance of insects among the world's animals is a fundamental scientific insight. Mosquitoes (Culicidae) are insects which attracted human's attention since time immemorial largely due to their potential to mediate a large number of deadly and debilitating diseases. Consequently, female mosquitoes have attained the zenith of biomedical significance, pushing the males into oblivion, notwithstanding the fact that males play an inevitable role in enhancing the transmission prowess of the *femme fatale!* In essence, without male's support, a female mosquito will aimlessly nurture a quixotic quest – the disease pathogen transmission!

Mosquitoes (app. 4000 species) occur in all regions and high altitudes within tropical and subtropical environments. This dominance means that in numbers of individuals beyond our comprehension these creatures permeate diverse and essential natural processes in Earth's terrestrial, aerial and freshwater ecosystems, contributing to the function of the natural world as a self-sustaining biological system. Mosquitoes, among which only vector species' females are hematophagous, are, in fact, an integral and complex part of the terrestrial, aerial and freshwater ecosystems with which the future of scores of other animals, including human being, and some plant species, is inextricably linked; therefore knowledge about their biology, ecology and behavioural diversity is a practical necessity. Since female mosquitoes seek blood for food and are involved in transmitting disease pathogens, only they – *the femme fatale* – are generally at the center of scientific exploration (Fig.1). Males are virtually ignored *hook, line and sinker*, and are but considered of far lesser scientific worth – a pre-empted conclusion based on biased, erratic and unsound human disposition!

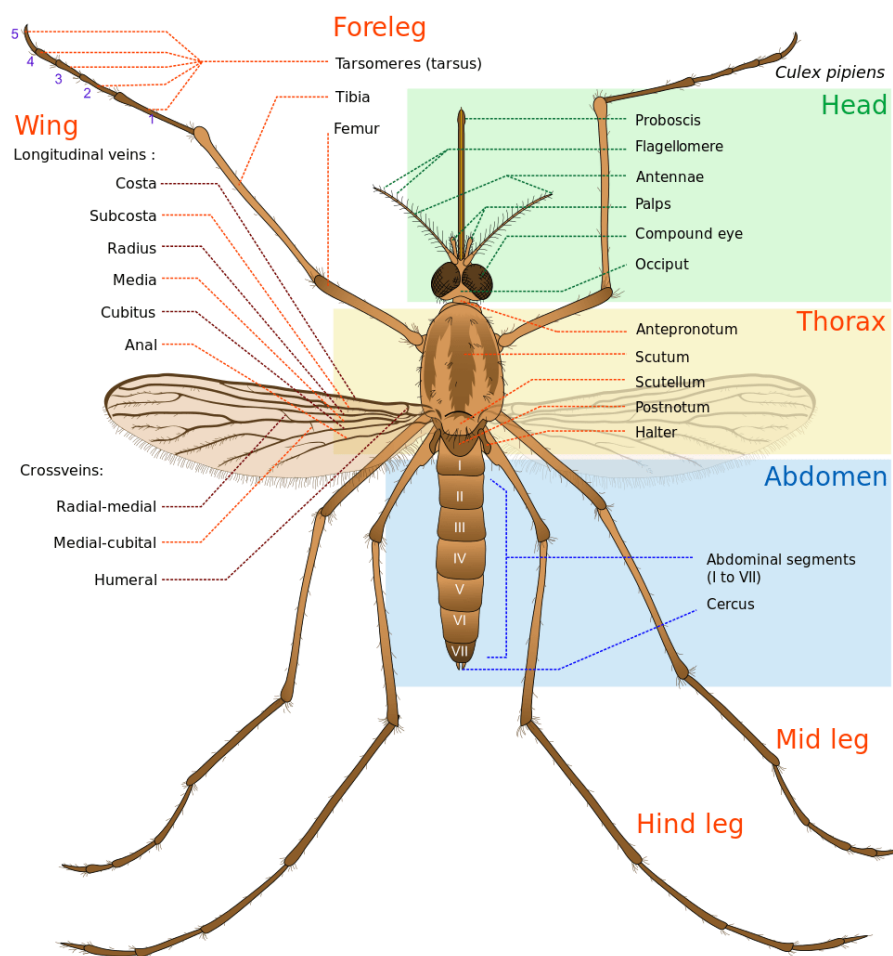


Fig. 1. Morphological features of a (female) mosquito (*Source: Free Wikipedia*).

Of all the predators that stalk the planet, mosquito has taken more lives than any other organism. Consequently, other than mosquito, no animal on earth has touched so directly and profoundly the lives of so many human beings with respect to their health, economy, social activities and tranquillity. Each year some 500 million people are affected by mosquito-borne diseases and nearly 3 million die from these. With a tiny little bite this pest, with the skill of a phlebotomist, transmits more than a hundred diseases, from protozoan (e.g., malaria) and helminthic (e.g., lymphatic filariasis) to viral infections (dengue, yellow fever, several different

kinds of encephalitides). There are over 3500 species of mosquitoes but only a small percentage of these phlebotomize and/or carry disease pathogens to humans, in fact there are more pestilent or nuisance mosquitoes than vectors. Throughout human memory, mosquitoes had been remembered by only a bad name, due largely to the role that females played to transmit pathogens of many devastating infections such as malaria, yellow fever, dengue etc., besides painful bite and blood-feed.

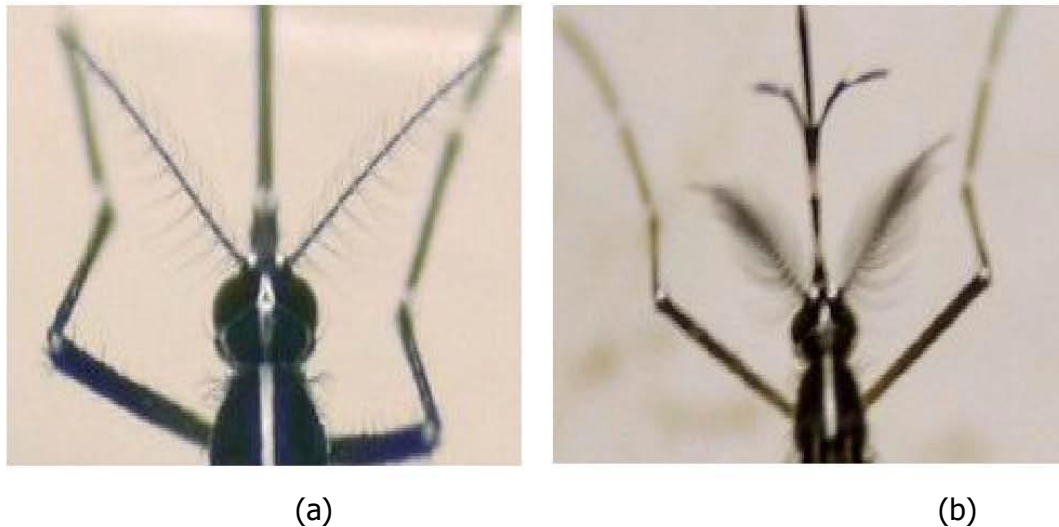


Fig 1: Mouthparts and antennal differences between female (a) and male (b) mosquitoes of *Ae. albopictus* (Source: Free Wikipedia).

Male mosquitoes remained in literary oblivion until recently when they were considered a blessing in disguise, i.e., using males, either as sterile or lethal gene carriers etc. for the control of vector mosquito populations; the discovery of a protein E20 in males which is necessary for successful ovulation in females; the prospects of saliva from male mosquitoes in developing/extracting local anesthetics such as heparin (... mosquitoes secrete saliva that contains biological substances, including anticoagulants that counteract a host's hemostatic response and prevent blood clotting during blood feeding. One of the important anticoagulants is Heparin (e.g., *Aedes togoi*) present in the salivary canal, salivary gland, and midgut of male and female mosquitoes. Although, female mosquitoes contain higher concentrations of heparin than male mosquitoes, it is nevertheless important to signify males otherwise considered of no worth; Ha et al. (2014); mosquitoes including males pollinate plants to help to ensure that different types of plant life thrive (when mosquitoes pollinate plants, especially the aquatic ones around which they spend much of their lives, they help perpetuate these plants. These plants provide cover and shelter for other animals and organisms); and, of course, mosquitoes seemingly representing a considerable biomass of food for wildlife on the lower rungs of the food chain etc. etc.!

Mosquitoes are bisexual and are easily distinguishable from each other morphologically, behaviourally and genetically, as succinctly described in Fig 2 and Table 1.

Table 1. Structural differences between males and female mosquitoes

BASIS OF COMPARISON	MALE MOSQUITO	FEMALE MOSQUITO
Antennae	Male mosquitoes have bushy fine hairs on their long antennae (proboscis) which help them with hearing and in sensing their potential mates' wing beats.	Female mosquitoes' antennae are less bushy and contain several blood odour receptors that help her target blood sources so that they can feed off their host.
Body Size	Male mosquitoes are relatively smaller than female mosquitoes.	Female mosquitoes are relatively larger than male mosquitoes.
Buzzing Sound	A male mosquito does not make a buzzing sound, or at best the sound is inaudible to human ears.	A female mosquito make an annoyingly audible buzzing sound when close to ears.
Lifespan	A male mosquito has a relatively shorter lifespan than the female mosquito. A male mosquito average lifespan is between one and three weeks.	A female mosquito has a relatively longer lifespan than the male mosquito. A female mosquito average lifespan is between two and four weeks
Human Contact	Male mosquitoes do not love human blood and they will typically avoid human contact.	The female mosquito loves to feed on human blood and thus they transmit diseases such as malaria, encephalitis and yellow fever.

The battle to control disease transmitting vector mosquitoes is among the greatest public health challenges of our time. Anopheles mosquitoes cause nearly 200 million cases of malaria annually, and kill approximately 600,000 people each year. *Aedes aegypti* and *Aedes albopictus* transmit dengue viruses, which cause more than 100 million infections, as well as emerging viral threats such as chikungunya and Zika. Many mosquito-borne pathogens have no commercially licensed vaccine or cure; even for those with a cure, such as malaria, drug resistance poses a serious challenge. The most effective tools against mosquito-borne pathogens therefore remain those focused on regulating vector populations rather than those strategies that target mosquito and focus on killing adults or larvae.

Male mosquitoes are as important as their counterparts; some sterling examples specific to the biology of the male mosquito briefed here in infuse interest in the young minds who are exploring mosquito science by delving deep into their basic attributes:

- (1) A deep investigation into reproductive biology of mosquito, *Aedes aegypti*— the vector of dengue, Zika, and other viruses, with special reference to (a) proteins, (b) transcripts, and (c) genetic architecture of seminal fluid and sperm may assist efforts to control wild mosquito populations by manipulating reproduction (Fig. 3). The sperm and seminal fluid proteomes represent a valuable tool for investigations of mosquito

biology and the development of molecular targets by which mosquito populations may be suppressed (Ethan, 2016).

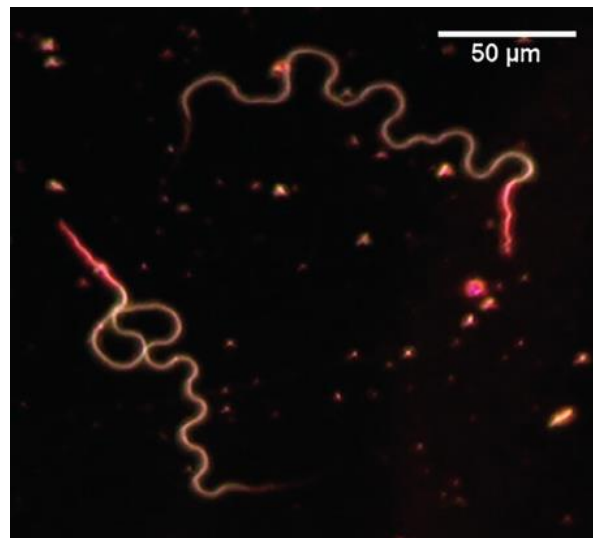


Figure 3. *Aedes aegypti* sperm. Rigid heads are stained with ethidium bromide. (Scale bar, 50 μm).

- (2) Isolation and characterization of a maleness gene, named Yob - a master regulator of the sex determination process in the African malaria mosquito, *Anopheles gambiae*, have raised a new hope to control vector population by wholly transforming population into that of males.
- (3) In insects, sex is commonly determined by a primary genetic signal that during the first hours of life activates a short cascade of genes, whose sex-specific products ultimately control whether an individual will develop as male or female. The molecular mechanisms underlying these developmental processes are surprisingly extremely variable, and in particular the primary sex-determining genes drastically differ in their nature between different groups of insects. Similar to humans, many insects possess a pair of sex chromosomes, with females carrying identical XX chromosomes and males XY chromosomes, the Y chromosome harbouring a dominant male-determining gene. The molecular identity of such maleness genes has remained enigmatic. The Yob gene represents only the second known case in insects which is encoded on the Y chromosome, and that activity of Yob was limited to males and was necessary to generate male-specific products of the sex determination pathway genes. The Yob transcripts are highly detrimental to females. When injected into mixed-sex early embryos of *Anopheles gambiae* and *Anopheles arabiensis*, Yob killed females before they hatch from eggs, but leaves male development unaffected.
- (4) A female mosquito needs two things to reproduce: human/animal blood and a visit from a male mosquito to impregnate her. The blood provides the nutrient-rich meal she will use to feed her developing eggs. The male provides not only sperm, but also a complex mix of proteins and other molecules, secreted from the so-called male accessory glands (MAGs) and deposited along with the sperm during mating. The secretions form a gelatinous "mating plug," which blocks access to the uterus. Both the blood meal and copulation provide necessary signals for the female to begin development of eggs in her ovary. While much is known about the events triggered by feeding, relatively little is known about how mating stimulates oogenesis. Now taking a deep peep into the sex life of the mosquito, scientists seem to have discovered

a male steroid hormone, named 20-hydroxy-ecdysone (20-E), delivered by the male mosquito to the female (*An. gambiae*) during sex. This hormone turns on a switch – a “mating signal” – to tell her to divert resources to producing an egg. Blocking the activation of this switch could be a new way of limiting mosquito populations and therefore the spread of malaria. The 20E hormone interacts with a protein in the female mosquito’s reproductive tract that stimulates egg production. The interaction of male hormone and female protein boosts the accumulation of fat in the female’s ovaries, which leads to eggs being produced more quickly and in higher numbers. On the other hand, virgin females rarely develop eggs (Robinson, 2013).

- (5) Approximately half of the human population is at risk of dengue. Additional mosquito borne pathogens, e.g., chikungunya and Zika, are spreading globally. *Aedes* mosquitoes are important mosquito vectors for these infections. In the absence of approved vaccines, therapeutic or prophylaxis, vector control remains the only means to combat multiple mosquito-borne pathogens. Auto-dissemination strategies have attracted attention as a method to reduce mosquito populations and benefit from mosquito behavior, in which a female mosquito visits multiple breeding sites. As practiced currently, ‘dissemination stations’ are attractive to adult females, which enter the station, become contaminated with a juvenile hormone analogue (JHA), exit and then contaminate breeding sites with levels of JHA that are lethal to immature mosquitoes. The auto-dissemination method is particularly attractive for those species that breed within small, cryptic sites, which serve as refugia from existing insecticidal measures. This approach envisages release of JHA-contaminated adult male mosquitoes, which do not bite or transmit pathogens. The males deliver JHA to breeding sites, either directly or indirectly, i.e., via the cross-contamination of females, which subsequently transfer JHA to breeding sites. The examined autocidal method can be used preemptively, e.g., in areas with low densities of indigenous mosquitoes and in advance of the natural population increase. Unlike auto-dissemination approaches that rely upon the indigenous population, an approach based on artificially-reared males can be more intensive change, because the number of males released is limited only by the logistics of male rearing and release as methods for mass production of mosquitoes are developed already. The auto-dissemination approach has been shown effective at treating cryptic refugia that remain unaffected by existing mosquito control methods. This approach relies on adult mosquito behaviour to spread larvicide to breeding sites at levels that are lethal to immature mosquitoes. The novel approach combines elements of autocidal and auto-dissemination strategies by releasing artificially reared, male mosquitoes that are contaminated with an insecticide.
6. And, finally, sterilized male mosquitoes using genetical techniques is moving a great pace for developing control strategies. Here following gene transfer and/or gene editing technologies females normally mate with sterile males, producing fewer eggs that are either never fertilized or the eggs and developing early instars are not viable, and cease to advance further, and perish before reaching final instar in aquatic life. In this technology even if there is a 30% reduction in the population it will still have a pretty substantial impact (Mains James et al., 2015).

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- Ha, Young-Ran, So-Ra Oh, Eun-Seok Seo, Bo-Heum Kim, Dong-Kyu Lee, Sang-Joon Lee, 2015. Detection of Heparin in the Salivary Gland and Midgut of *Aedes togoi*. *Korean J Parasitol* 52(2): 183-188.
- Mains James, W., Corey L. Brelsfoard and Stephen L Dobson, 2015. Male mosquitoes as vehicles for insecticide. *PLoS Negl Trop Dis* 9(1):e0003406. doi: 10.1371/journal.pntd.0003406.
- Robinson, R. 2013. His Hormone, Her Oogenesis: How Male Malaria Mosquitoes Trigger Female Egg Development. *PLoS Biol* 11(10): e1001694. doi:10.1371/journal.pbio.1001694.

SOMA's 13th CONFERENCE – A FORTNIGHTLY WEBINAR SERIES

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Prof. Dr B.K. Tyagi

- (1) Dr Patrick Manson and discovery of filariasis – mosquito connection: the birth of medical entomology. 13th Natl. Conf. Med. Arthropod., SOMA's fortnightly webinar; Vol 1(1); 20 Aug., 2020, 11 am.
- (2) Dr Ronald Ross: An untold story of the first Indian Nobel Laureate and a great Malariologist: Lessons Learned From Dr Ross's Life. SOMA's fortnightly webinar; Vol 1(2); 16 Sep, 2020; 11 am.

Prof. Dr B. Reddy Naik

- (1) Dr Ronald Ross: An untold story of the first Indian Nobel Laureate and a great Malariologist: Discovery of Mosquito-malaria connection. SOMA's fortnightly webinar; Vol 1(2); 16 Sep, 2020; 11 am.

WEBINARS PRESENTED

B.K. Tyagi

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1. Prof. Dr B.K. Tyagi

- (i) Migratory locusts in changing environment, with special reference to the Thar Desert (India): Their biology, adaptations, migration, agricultural damage and

control. International Webinar on World Environment Day with the theme: "Biodiversity Protection", jointly organized by Voice of Indian Concern for the Environment/Voice of International Community for the Environment- VOICE; 5th June; 11 am.

- (ii) Insects and other arthropods of Biomedical Importance. UGC STRIDE Scheme sponsored 21-days Refresher Course on RESEARCH METHODOLOGY through online; jtly. orgd. Aizwal, Mizoram, India, K.S. Rangaswamy College of Technology, Trichengode, TN, India and Nature Science Foundation, Coimbatore, TN, India; 13th June, 2020; 11 am.
- (iii) The complexity of mosquito-borne diseases & why have we been unable to control them? Webinar organized by TIGS, Bengaluru; 17th June, 2020
- (iv) Distribution, Prevalence, Surveillance and Control of major Dengue vectors: *Aedes aegypti* and *Ae. albopictus*. Webinar for North Bengal University, Siliguri, WB; 5th Aug., 2020; Hrs 11.00 am-12.30 pm.
- (v) Anthropogenic agro-forestry environment and conflagration of deadly epidemics of malaria and dengue. *Webinar on International Virtual Internship Program on Agro-environment (IVIPAEM) 2020; NOV. 6-13, 2020.*
- (vi) MOSQUITO DIVERSITY: The Ugly and The Beautiful - a discussion on taxonomy, chorogeography, ecology and behaviour. ZSI/NRC's NWS-FDI-Webinar-02; 23rd Sep., 2020; 10 am.
- (vii) Origin and Evolution of Species: theories and hypotheses. Online Refresher Course in "Life Sciences" with a focus on "Vision NEP-20: Integrated Life Sciences – Challenges and Opportunities"(VILSCOP), orgd. Zoology Depatt, Osmania Univ., Hyderabad, Telangana, India; 5th October, 2020; 10 am.

2. Dr Rina Tilak

- (i) Scrub typhus-an emerging challenge. AHCF webinar 28 Nov 2020.
- (ii) Tick Borne Diseases. Refresher Course in "Life Sciences" with a focus on "Vision NEP-20: Integrated Life Sciences – Challenges and Opportunities" (VILSCOP), UGC-HRDC, Osmania University, Hyderabad- 5th -19th October, 2020.

3. Prof. Dr Neera Kapoor

- (i) Prof. Neera Kapoor presented a Paper on "Online Curriculum to combat novel diseases" in International Virtual Conference of Open and Distance Education held on 2-4 July, 2020 organized by Open University of Hong Kong.
- (ii) Prof. Neera Kapoor delivered 3 lectures on Molecular Entomology in Disease Control in virtual workshop for college teachers organized by Uttrakhand Open University (UKOU), Haldwani held on 8-12 July,2020.

HONOURS, AWARDS AND DISTINCTIONS

B.K. Tyagi
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- (1) Dr B.K. Tyagi was invited to act the Chief Guest for the one-week long International Virtual Internship Programme on Agro-Environmental Management (IVIPAEM) 2020 commencing Nov. 6, 2020.

- (2) Dr. Varun Tyagi "NPDF" Defence Research Laboratory (DRDO), Tezpur Assam and Fellow of Society of Medical Arthropodology (SOMA) received a certificate for his winning in different lab level competition from the Director DRL (DRDO) and the Vice Chancellor of Tezpur University, Tezpur on the occasion of the 59th DRL Lab Raising Day and closing ceremony of *Hindi Pakhwada*.



PROMOTIONS AND APPOINTMENTS

B.K. Tyagi

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- (i) Prof. Dr B. Reddya Naik, appointed as the Chairman of the Board of Zoology Studies, Osmania University, Hyderabad, Telangana State, India w.e.f. 2nd Sep., 2020.
- (ii) Dr Roop Kumari, Joint Director, NVBDCP, Delhi has taken VRS. However, she will continue to work as a Consultant with the WHO SEARO, Delhi.
- (iii) Dr Rajeev Tyagi, PhD, has joined as Ramalingaswami Fellow and Faculty, Division of Cell Biology and Immunology, Biomedical Parasitology and Nano-immunology Lab., CSIR-Institute of Microbial Technology (IMTECH), Sec-39A, Chandigarh-160036, India [Tel: 91-172-6665278 (Off), 91-172-6665279 (Lab); Email: rajeevtyagi@imtech.res.in , rajeev.gru@gmail.com]
- (iv) Dr Prabhakar Mishra has joined as Assistant Professor, Dept. of Biotechnology, School of Applied Sciences, REVA University, Bangalore-64, India; Email: prabhakar.science99@gmail.com
- (v) Dr. Ayushman Ghosh has joined as Scientist 'C', Department of Biotechnology (GOI), New Delhi – 110003; Contact No: 8971308496, 9163150963

PUBLICATIONS

B.K. Tyagi

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(1) Vijay Veer and Varun Tyagi,

2020. *Anopheles (Cellia) baimaii*

(sibling species of the Dirus

complex): the invincible vector

of forest malaria in north east

India. In: Vector Biology and

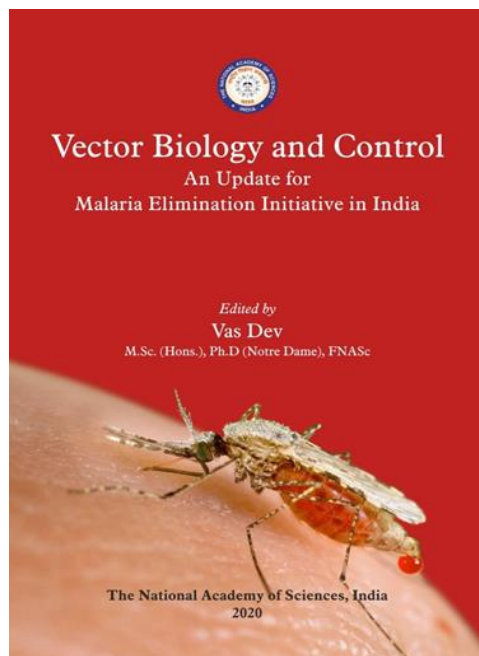
Control an update for

malaria elimination initiative in

India. Ed. Vas Dev. National

Academy of Science, India, pp.

43-48.



- (2) **Lakshmi, G, Neera Kapoor and Shilpi Garg,** 2020. Awareness on critical prevention practices to combat the drug resistance in *Plasmodium falciparum* and *Plasmodium vivax* malaria from Rajasthan, India. *International Journal of Mosquito Research* 7(4): 83-90.
- (3) **Singh Shweta, Neera Kapoor and R. K. Jauhari,** 2020. Biological Control of Pre-Adult mosquitoes: Inter-Mosquito trematode larval antagonism in the vicinity of Doon Valley. *Int. J. Zool. Res.(IJZR)*, 10(1):35-42.
- (4) **Lakshmi, G., Shilpi Garg and Neera Kapoor,** "BOOK REVIEW: PIERCING POWER IS MORE THAN THE HUMAN POWER ", *International Journal of Creative Research Thoughts (IJCRT)*, ISSN:2320-2882, Volume.8, Issue 4, pp.2406-2407 <http://doi.one/10.1729/Journal.23491>. Available at :<http://www.ijcrt.org/papers/IJCRT2004332.pdf>

NEWS

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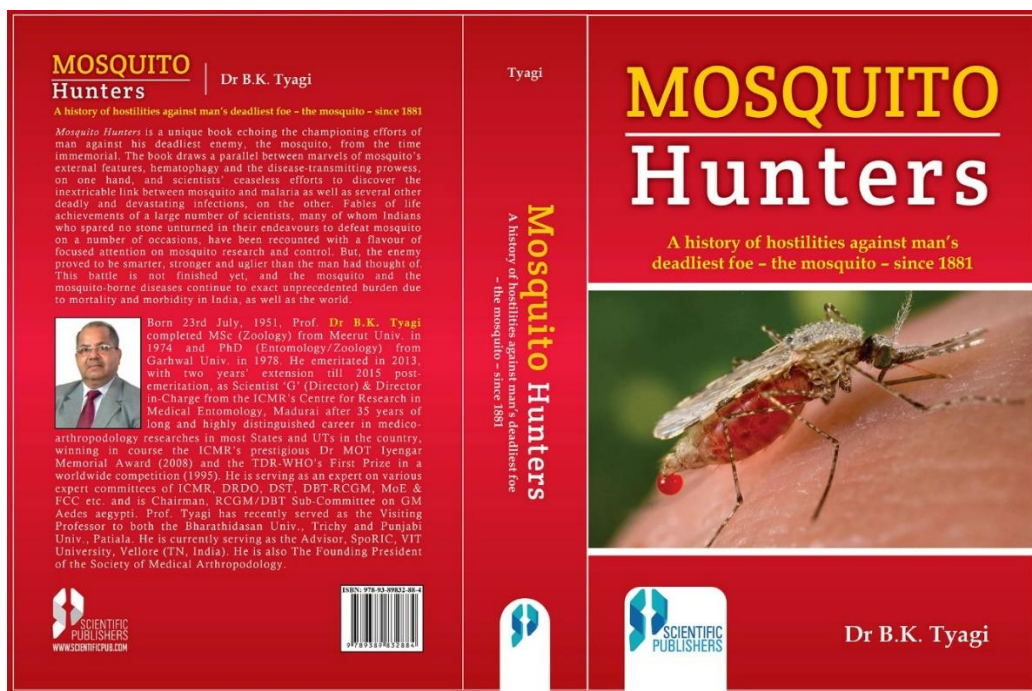
- (1) The SOMA will be shortly organizing a webinar in lieu of the planned 1st International/13th National Conference of Medical arthropodology during Nov. 2020, in view of the ongoing onslaught by the COVID-19. The Conference will now take place on Aug. 19-20, 2021; a circular will follow shortly.

BOOKS PUBLISHED

B.K. Tyagi

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- (1) Prof. Dr B.K. Tyagi, 2021. Mosquito hunters: A history of hostilities against man's deadliest foe- the mosquito – since 1881. Scientific Publisher (India). 452 pp.



The Editorial Board wishes to thank all contributors, especially Prof. Dr B. Reddy Naik, Dr Rina Tilak, Dr S. Shiva, Dr D.S. Suman and Dr Varun Tyagi for their timely help in shaping this issue of the *SOMA Newsletter*.

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