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Vector Competence in Mosquitoes: Interaction between Intrinsic & Extrinsic Factors

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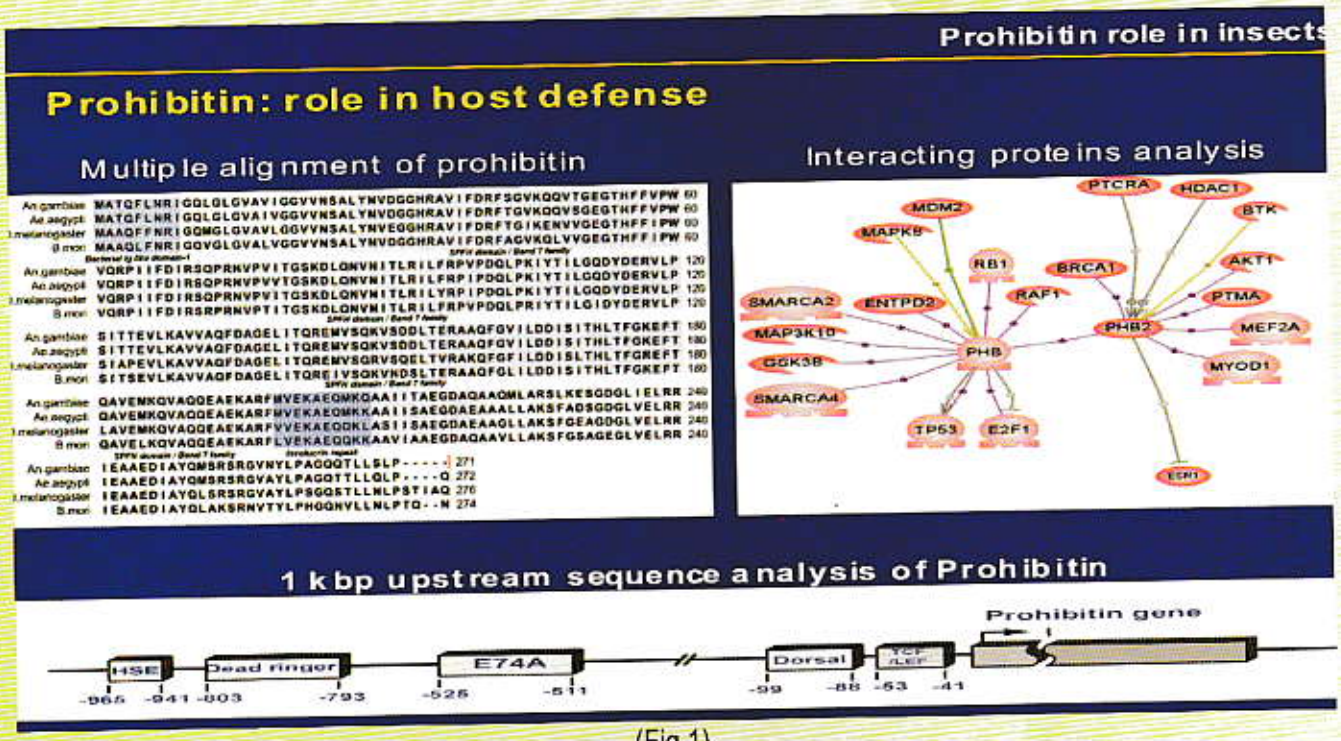
Efforts to control mosquito borne diseases have been impeded, in part, by development of drug resistant parasites, insecticides resistant mosquitoes and a variety of environmental factors. Understanding of vector-pathogen (parasite or virus) interactions will be equally important in developing newer strategies to control vector population as well as to interfere in the vector-pathogen interactions. The molecular information regarding the vector, while the pathogen enters and establishes in it, are not completely understood. Maintenance and transmission of pathogens are depended on the competent mosquito vectors. This vector competence is a multigenic, recessive phenotype, and is influenced by, in addition to various genetics and environmental factors, also susceptibility of vector to infection, permissiveness for propagation of pathogen, duration of incubation and transmission efficiency of the vector. Thus there are both intrinsic and extrinsic factors which determine competence of the vector mosquito.

The extrinsic factors govern, to a large extent, the propagation of pathogen inside the vector. They may also influence vector density itself. Availability of nutrition, humidity and temperature are the most conspicuous among them.The intrinsic factors are the inherited traits which influence host preferences as well as ability of the mosquito to get infected, say by arboviruses. There exist internal barriers in the vector at different stages and tissue systems which regulate diseases transmission. At molecular level, the barriers are affected through specific enzymes, immune peptides, specific receptors and gut microbes.

Adaptive stress tolerance (Hormesis) at different stress conditions has been a well established factor known among living systems. Exposure to sublethal levels of temperature or pesticide concentrations not only confers tolerance to higher temperatures and/or higher pesticide concentrations but also make mosquitoes adaptableto higher virus/parasite load. Severe summers in most parts of the world are followed by severe epidemics, implying that the vector competence is enhanced under such conditions. We had demonstrated that stress inducible gene expression is associated with increased vector competence. However, these results were restricted to known stress proteins and their involvement in cellular as well as virus associated metabolic processes.

***Serratia odorifera* modulates vector susceptibility to DENV-2:** We recently identified, among the diversity of micro flora in the midgut of mosquitoes (anopheles as well as Aedes species), a Gram negative bacterium,

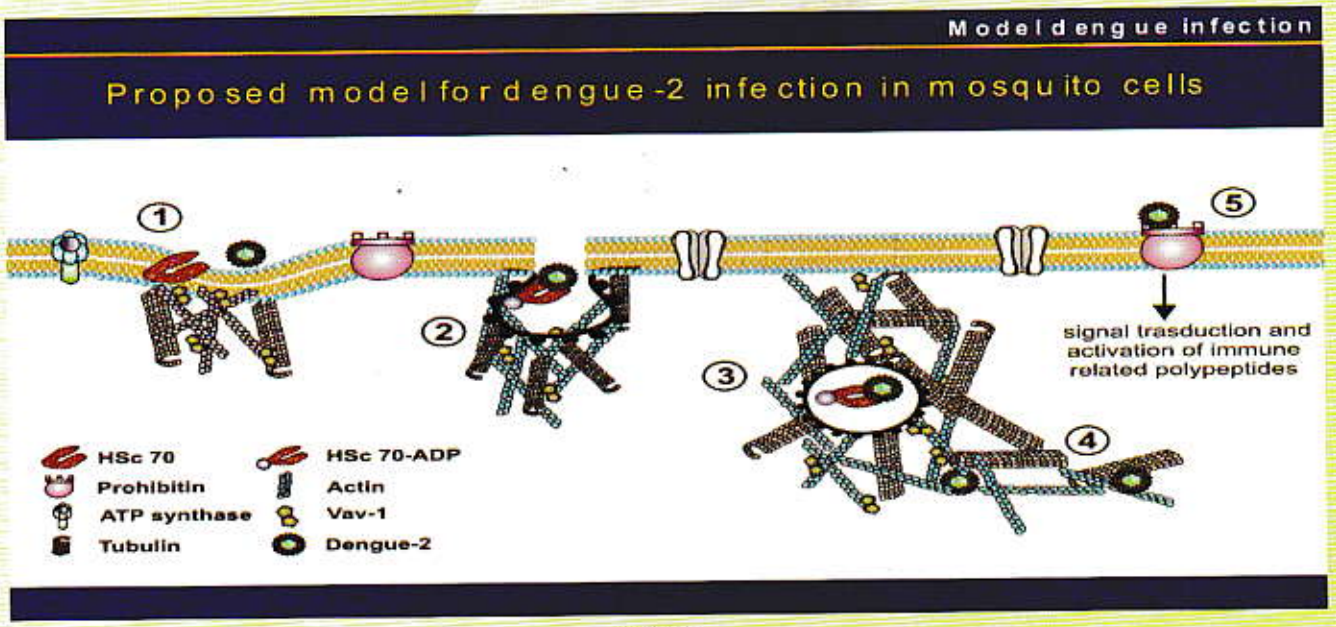


(Fig.1).

Serratia odorifera which is transtadially maintained in the life cycle of vector mosquitoes. We observed that presence of this bacterium in the midgut significantly enhanced susceptibility (from 30% to about 65%) of the mosquito to Chikugunya (CHIK) as well as Dengue (DENV-2) viruses. A 40kDa (P40) extracellular glycoprotein synthesized by *S. odorifera* was identified to be interacting with the midgut brush border. Expression of this protein was enhanced by increased external temperature. The total proteomic analysis also revealed that P40 selectively binds to the mosquito protein, identified as Prohibitin. Both CHIK and DENV-2 could also interact with P40, possibly through their E protein. P40, however, did not interact with other viral receptors in the midgut. Prohibitin on the other hand is a non receptor protein interacting with these viruses. Prohibitin has been thus identified as an important intrinsic refractory molecule in the vectors. Interestingly, even the plasmodium would interact with Prohibitin suggesting Prohibitin as a universal refractory molecule in mosquitoes. Prohibitin is also a part of innate immunity controlling pathway and could substitute rel factor. Thus Prohibitin would be visualized to play a significant role in confining resistance to viral/parasite infection in mosquitoes (Fig.1).

dentification and characterisation of DENV-2 binding proteins from Aedes: The roles of cell surface proteins as well as other cellular proteins in DENV-2 attachment, entry and assembly are not yet fully understood. Employing Virus Overlay Protein Binding Assay (VOPBA), we identified DENV-2 binding polypeptides from midgut brush border membrane fraction of *Aedes aegypti* adult and larvae , and also from the *in vitro* cultured cell lines, viz., C6/36(*Ae. albopictus*) and A7 (*Ae. aegypti*). Identities of Vav-1, hsc70,orisis, actin, tubulin and prohibitin were confirmed by MALDI anlysis and also using monospecific antibodies (Fig.2).

***Drosophila melanogaster* as a model test organism for arboviruses:** Most of the receptor molecules for DENV-2 were found to be normal house keeping polypeptides in mosquitoes, which are also found in other



(Fig.2).

insects. Consequently, other dipterans, such as Drosophila could also permit DENV-2 infection and propagation. Indeed, *D. melanogaster* midgut as well as cell line showed presence of these polypeptides which interacted with DENV-2. On injecting sub-lethal dose of DENV-2, Drosophila exhibited 61% susceptibility 7 days determined by detection of virus in the brain tissue. This virus could reinfect productively *Aedes aegypti* as well as Drosophila with normal efficiency. Drosophila cell line S2 also was found to be as susceptible as C6/36 cells.

Gene expression profiles of Drosophila on 24 h and 48 h post infection were studied using *D. malanogaster* cDNA microarrays. Analysis of up regulated and down regulated expression of a number of genes (524 after 24h, and 656 after 48h postDENV-2 infection) revealed that Drosophila could be used as a model host system for molecular and physiological analysis of DENV-2 virus infection, and possibly in case of other related arboviruses.

Use of Drosophila microarrays could be extended for gene expression analysis to other arboviral host insects whose whole genome sequences are not yet available. To test such an application,molecular composition of gene expression pattern in *An stephensi* infected by Plasmodium was examined using Drosophila cDNA microarrays. Ant- plasmodial response mounted in *An stephensi* was found to be similar to that in *An. gambae* in this study.

A Tribute to Professor Chris Curtis

Prof Sajal Bhattacharya, Dept. of Zoology, Asutosh College, Kolkata
& Chairman NAVBD (Eastern Chapter)
and
Miss Swaha Bhattacharya
Dept. of Biotechnology, St.Xavier's College, Kolkata

On 13th May 2008, the world lost one of its most leading medical entomologists, Professor Chris Curtis who belonged to the London School of Hygiene and Tropical Medicine. Chris Curtis will always be remembered as a pioneer in the development of low-technology methods for mosquito control. He introduced Insecticide-Treated Nets (ITNs) as a dominant technology measure to effect the prevention of transmission of malaria by mosquito vectors. He led a team of researchers that demonstrated how the 'mass effect' of ITNs can reduce the ability of the local mosquito population to transmit malaria, thus providing protection to everyone in the area. As a humanitarian, he had taken a number of initiatives for government-aided mass bed-net distribution in Africa and due to this noble endeavour of his, thousands of lives were saved from the vice-like grip of malaria. Professor Curtis had made the service of practical public health his mission in life. During his PhD, which was a study of basic genetic mechanisms in fungi, he tried looking for useful applications of his research work andcame up with the idea of using chromosomal translocations as a method of genetic control. He moved on to develop translocations in tsetse flies, which are especially vulnerable to this form of control because of their low fertility. He came to India to work on the genetic control of mosquitoes in a World Health Organization research project in Delhi unit, India. He joined the London School of Hygiene and Tropical Medicine in 1976 after false rumors about biological weapons caused the Delhi unit to close (http://www.lshtm.ac.uk/news/curtis/obituary.html). Back in London, his focus was on various forms of "appropriate technology for mosquito control", which would remain the principal area of his research for the remaining part of his life. One of these technological measures was a simple idea to suffocate Culex mosquitoes that breed in places like pit latrines by using loose beads of expandable polystyrene.

A close colleague of Professor Curtis, Dr. Jo Lines stated that Chris Curtis was responsible for introducing numerous techniques mostly methods of evaluating interventions that are now considered standard. Lines also said that Curtis was a highly regarded theoreticalscientist who produced simple models of genetic control in insects and the evolution of resistance to insecticides.

It is ironical to note that Chris Curtis' death anniversary which is 13th May 2008 coincides with the birth date,13th May 1857 of Sir Ronald Ross, who unraveled the mystery of malaria transmission mechanism. Both are great stalwarts in this field of dealing with the menace called mosquito. In fact, the first author considered it to be a great privilege of being able to work with him closely in LSHTM. In spite of being a globally renowned scientist, he has always appeared as an incredibly humble and generous human being. During the first author's stay in London, and even after that, he has always come to look up to Professor Curtis as a guide and mentor whose knowledge and experience can be relied upon at all times. His has been a life well-lived and apart from being a world-class scientist, Chris Curtis will be fondly and lovingly remembered for his humility and utter goodness of spirit by the thousands of people whose lives he touched.

NEWS/EVENTS

1. Constitution of New Executive Committee

The New Executive Committee of the Academy has been constituted in 5th December 2008 after the election through secret ballot held in the month of November 2008.The following are the current office bearers.

1.	President	:	Prof D N Deobagkar
2.	Secretary General	:	Dr M R Ranjit
3.	Joint Secretary	:	Dr B N Nagpal
4.	Treasurer	:	Dr R K Hazra
5.	Executive Member	:	Dr Sajal Bhattachrya
6.	Executive Member	:	Dr V K Dua
7.	Executive Member	:	Dr Neena Valecha
8.	Executive Member	:	Dr Ashwani Kumar
9.	Executive Member	:	Dr S K Ghosh
10.	Executive Member	:	Dr Jagbir Singh

2. Establishment of Regional Chapters of the Academy

Four regional chapters of the Academy have been established. These are in Bangalore (South), Haridwar (North), Kolkata (Eastern & North Eastern) and Goa (West). The Chairman for Southern chapter will be Dr S K Ghosh, for Northern chapter Dr V K Dua, for Eastern & North Eastern chapter Dr S Bhattacharya and for Western chapter Dr Ashwani Kumar.

3. Organization of Seminars/Symposiums

(i) X International Symposium on Vectors and Vector Borne Diseases was organized jointly by National Academy of Vector Borne Diseases, National Institute of Malaria Research and Goa University, Goa from 4th-6th November 2009. It was inaugurated by the Chief Minister of Goa Mr Digambar Kamat. At least 250 Scientists and Research Scholars from India and abroad belonging to Research Institutions, Universities, Colleges, Vector Borne Disease Control Programme Managers and Scientific Staff working at the center, states and urban areas attended the Symposium. The theme of the Symposium was Vector Borne Diseases: Health, Environment and Socioeconomic implications.

(ii) A daylong National Seminar on "Challenges of Mosquito Borne Diseases" was organized jointly by Eastern and Northeastern Chapter of National Academy of Vector Borne Diseases and Ashutosh College, Kolkata (under Calcutta University) and Academic Excellence and Access (FAEA) on 13th Feb 2009.At least 120 Scientists, Acdemicians and Research Students, Post Graduate Students from West Bengal and Orissa attended the Seminar.

4. Awards for the Year 2009

1. **NAVBD Award related to Environmental aspects** : Dr. B.N. Nagpal
2. **Bayer Environmental Science Award.** : Dr. Neena Valecha,
3. **Godrej Sara Lee Award for Personal Protection from Mosquitoes** :Dr. V.K. Dua
4. **Vestergaard Frandsen Award for Vector Control**:Dr. Makawitage Devika Bernadiae Perera
5. **Bio-tech International Award for Excellence in promotion of Bio-Control of Vectors:** Dr. S.K. Sharma

5. Best Poster Presentation

Senior Group

1. **K. Pai,** University of Pune, Pune
   N.S. Korgaonkar, NVBDCP, DHS, Goa
3. **D. Sukumaran,** DRDE, Gwalior
4. **R. Gupta,** CDRI, Lucknow.

Junior Group

1. **Sutopa B. Dwivedi,** Institute of Bioinformatics, Bangalore
2. **S. Swain,** RMRC, Bhubaneswar
3. **M.S. Paingankar,** Dept. of Zoology, University of Pune, Pune
4. **T.C. Yahathugoda,** University of Ruhuna, Galle, Sri Lanka.

5. Institution of Awards

The National Academy of Vectors and Vector Borne Diseases has instituted several awards in recognition of the outstanding research contributions made by the scientists in the field of vectors and vector borne diseases.

(i) Vestergaard Frandsen Award for Vector Control

"Vestergaard Frandsen Award" consisting of a cash award of Rs 200,000(two lakh) is to be given to active researchers who are nationals of any SAARC member country and have contributed significantly in the fields of understanding the mechanism of insecticide resistance, vector bionomics and control.

(ii) Bayer Environmental Science Award

"Bayer Environmental Science" has initiated the above award consisting of Rs 100,000 (one lakh) in cash to be given to a well established scientist having working experience in India and who has made an outstanding contribution in any field of vector borne diseases preferably in the vector/disease management aspect.

(iii) Godrej Saralee Award

M/S Godrej Saralee has declared an award named "Godrej Saralee Award" which will be presented during annual International/National Symposium organized by the Academy for excellence in research on personal protection from Mosquitoes. The Award includes a cash prize of Rs. 2 lakh and a citation.

(iv) National Academy of Vector Borne Diseases Awards

There are Two awards (a medal and a citation) for the outstanding contributions in the field of Vector Borne Diseases, one for work related to Environmental aspects and other on Molecular Biology.

(v) Best Poster Presentation Awards

During Annual meeting/Symposium of the Academy Best Poster Presentation awards are given in two categories.
(1) Senior scientists (35 years and above): Three awards
(2) Junior scientists (below 35 years): Three awards

Announcement

The National Academy of Vector Borne Diseases will hold its next Annual Conference (XI) at the end of this year (2010) or early next year (2011). The interested host can propose his/her willingness to President of the Academy by 15th April 2010 so as to decide the venue.

Address

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Inauguration of X International Symposium on Vectors and Vector Borne Diseases Nov 4-6, 2009, Goa

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Bayer Environmental Science Award to Dr. Neena Valecha

Godrej Sara Lee Award for Personal Protection from Mosquitoes to Dr. V.K. Dua

Bio-tech International Award for Excellence in promotion of Bio-Control of Vectors to Dr. S.K. Sharma

NAVBD Award related to Environmental Aspects to Dr. B.N. Nagpal

A moment during the Valedictory function of the X International Symposium

National Seminar Organised by Eastern & North Eastern State Chapter of NAVBD

NATIONAL ACADEMY OF VECTOR BORNE DISEASES

(Regn. No. 19767/199 of 1994 – 95 under societies Registration Act XXI of 1860)
(Regional Medical Research Centre, Bhubaneswar- 751023, India)

MEMBERSHIP FORM

1. Name :

2. Designation :

3. Date of Birth :

4. Academic Qualification :
(Only Post Graduate Degrees)

5. Address : Present :

Phone..... FAX..... Email.....

Permanent :

Phone..... FAX..... Email.....

6. Experience (years) Research :

Teaching :

Management :

7. Type of Membership & Fees:

Membership	Period	Amount (Rupees)	For scientists abroad (US $)
Life Member	Life time	1000	50(SAARC countries) 100 (Others)
Institutional member	Life time	20,000	1000(SAARC countries) 2000 (Others)
Patron (by donating)	Life time	more than 20,000	more than US$2000

Members up to 31.03.1995 were treated as founder members. The membership fee (Bank Draft drawn in favour of **Treasurer, National Academy of Vector Borne Diseases, Bhubaneswar**) and the filled up form may be sent to Dr. R. K. Hazra, Treasurer, National Academy of Vector Borne Diseases, Regional Medical Research Centre, Bhubaneswar- 751023, Orissa, India. (Mobile: 91-9861173867, E mail- rupenkh@yahoo.co.in)

*Sl. No. 2, 3, 4 & 6 are not applicable for institutions.

Place:

Date: Signature