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MOSQUITOES AND ASSOCIATED AQUATIC INSECTS IN RICE AGROECOSYSTEMS OF MALAYSIA: SPECIES COMPOSITION, ABUNDANCE, AND CONTROL OF MOSQUITO LARVAE IN RELATION TO RICE FARMING PRACTICES

by

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LIST OF ABBREVIATIONS

a.i	Active ingredient (s)
ANOVA	Analysis of variance
B.sp.	*Bacillus sphaericus*
B.t.i.	*Bacillus thuringiensis israelensis* H-14
ºC	Celsius degrees
Cm	Centimeter (s)
Cx.	*Culex*
df	degree of freedom
DO	Dissolved oxygen
EC	Emulsion Concentration
EPA	Environment Protection Agency
24hr	24 hours (s)
Kg	Kilogram (s)
L or l	Liter
LC	Lethal concentration
m^2	Square meter (s)
m^3	Cubic meter (s)
mg	Milligram (s)
Ml	Milliliter
MMF	Monomolecular film
ppm	Part per million
®	Registered
spp	species
WDG	Water Dispersible Granules
wk	Week

NYAMUK DAN SERANGGA-SERANGGA AKUATIK DI AGROEKOSISTEM PADI DI MALAYSIA: KOMPOSISI SPESIS, KELIMPAHAN DAN KAWALAN LARVA NYAMUK YANG BERHUBUNGKAIT DENGAN AKTIVITI PENANAMAN PADI

ABSTRAK

Kajian dijalankan di kawasan sawah padi di Pulau Pinang. Sebanyak 6 sawah padi, parit dan terusan disampel pada setiap minggu untuk mendapatkan larva nyamuk dan serangga-serangga akuatik. Kajian ini dijalankan selama 17 bulan. Objektif kajian ini adalah untuk mengetahui tentang komposisi spesis, penyebaran dan kelimpahan nyamuk serta serangga-serangga akuatik di habitat sawah padi dan untuk menganalisa hubungan di antara penyebaran spesis-spesis ini dengan fasa-fasa penanaman padi. Nyamuk yang disampel yang berada pada peringkat larva instar ketiga dan keempat adalah lebih tinggi berbanding larva instar pertama dan kedua serta pupa. Spesis nyamuk menunjukkan perbezaan pada penyebaran dan corak kelimpahan di sawah padi. Analisis prinsip komponen menunjukkan hampir semua pembolehubah (71%, 74%, 75%) bervariasi diantara tarikh persampelan dalam komposisi spesis. Walaubagaimanapun, *Culex pseudovishnui* Colles dan *Anopheles jamesii* Theobald dipengaruhi oleh konduktiviti air dan kebanyakannya dijumpai pada fasa muda dan padi beranak. Bilangan *Cx. bitaeniorhynchus* Giles, *Cx. vishnui* Theobald dan *Anopheles sinensis* Wiedemann yang dipengaruhi oleh kedalaman air dan ketinggian pokok padi adalah tinggi semasa fasa padi beranak dan matang. *Culex tritaeniorhynchus* Giles, *Cx. gelidus* Theobald, *Cx. fuscanus* Wiedemann dan *Ficalbia luzonensis* Ludlow dijumpai pada fasa pembajakan dan fasa rang dan dipengaruhi oleh intensiti cahaya dan pH air. Perhubungan diantara taburan dan kelimpahan semua spesis nyamuk adalah signifikan dengan taburan serangga akuatik. Keberkesanan tiga larvasid, Abate®, 96.2%, *Bacillus thuringiensis* var. *israelensis* (serotype 14) VectoBac® technical powder dan *Bacillus sphaericus* Neide (serotype $_{H5a5b}$) Vectolex-WDG®, dinilai didalam makmal terhadap spesis nyamuk yang dikumpul dari lapangan iaitu *Cx.*

bitaeniorhynchus, *Cx. tritaeniorhynchus, Cx. gelidus, Cx. vishnui* dan *An. sinensis.* Tahap kerentanan spesis-spesis ini kepada larvasid-larvasid yang diuji adalah berbeza; *An. sinensis, Cx. tritaeniorhynchus, Cx. gelidus, Cx. bitaeniorhynchus* dan *Cx. vishnui,* dalam susunan tersebut adalah rentan kepada Abate® apabila nilai LC_{50} yang diuji berada dalam lingkungan 0.0000001 ppm sehingga 0.009 ppm dan nilai LC_{90} berada dalam lingkungan 0.0000012 ppm hingga 0.242 ppm. *Culex gelidus, Cx. tritaeniorhynchus, Cx. bitaeniorhynchus*, *Cx. vishnui* dan *An. sinensis* dalam susunan tersebut adalah rentan kepada VectoBac® apabila LC_{50} berada dalam lingkungan 0.000108 ppm sehingga 0.029 ppm dan nilai LC_{90} berada dalam lingkungan 0.000509 ppm hingga 0.142 ppm. Manakala nilai LC_{50} yang berada dalam lingkungan 0.000178 ppm hingga 33.376 ppm dan nilai LC_{90} yang berada dalam ligkungan 0.0356 ppm sehingga 355.94 ppm, *Cx. tritaeniorhynchus, Cx. gelidus, Cx. vishnui, Cx. bitaeniorhynchus*, dan *An. sinensis* dalam susunan tersebut adalah rentan kepada Vectolex®, setelah masing-masing didedahkan selama 24 jam. Manakala apabila nilai LC_{90} berada dalam lingkungan 0.01 ppm sehingga 7.51 ppm, *Cx. tritaeniorhynchus, Cx. gelidus, Cx. vishnui, Cx. bitaeniorhynchus*, dan *An. sinensis* dalam susunan tersebut adalah rentan terhadap Vectolex® selepas masing-masing didedahkan selama 48 jam. Di lapangan, Abate® 500E pada kepekatan 0.28 mg/L mencatatkan 75.3-100% kadar kematian terhadap *Anopheles* spp. dan 75.8-90.5% kadar kematian terhadap *Culex* spp. selepas 3 minggu kawalan dijalankan. VectoBac-WDG® pada kepekatan 0.48 mg/L menyebabkan 55.52-100% kadar kematian terhadap *Anopheles* spp. dan 51.29-100% kadar kematian terhadap *Culex* spp. selepas 5-7 hari kawalan dijalankan. Agnique MMF® pada kepekatan 5L/ha (0.5ml/m^2) pula mencatatkan 84.9-93.7% kadar kematian *Culex* spp. selepas 2 minggu kawalan dijalankan. Ketiga-tiga larvasid pada kepekatan yang sama tidak menunjukkan sebarang kesan yang tidak baik terhadap fauna mikroinvertebrata lazim seperti Corixidae, mayfly naiad, larva damselfly dan Dystiscidae.

MOSQUITOES AND ASSOCIATED AQUATIC INSECTS IN RICE AGROECOSYSTEMS OF MALAYSIA: SPECIES COMPOSITION, ABUNDANCE, AND CONTROL OF MOSQUITO LARVAE IN RELATION TO RICE FARMING PRACTICES

ABSTRACT

The study was conducted in Penang rice fields, Malaysia, where a total of 6 rice fields and associated drainage and irrigation canals were sampled weekly for mosquito immature stages and associated aquatic insects over 17 months period. The aim of the study was to investigate species composition and distribution pattern of mosquito and associated aquatic insects in relation to rice field phases. Mosquito species collected in the 3rd-4th larval instars were higher than 1st-2nd larval instars and pupae. Mosquito species showed difference in their distribution and abundance patterns in the rice fields. Principal components analysis showed the variables explained mostly (71%, 74%, 75%) of the variation among the sampling dates in the species composition. However, *Culex pseudovishnui* Colles and *Anopheles jamesii* Theobald were affected by water conductivity mainly found in the young and tiller phases. *Culex bitaeniorhynchus* Giles, *Cx. vishnui* Theobald and *Anopheles sinensis* Wiedemann were affected by water depth and rice plant height, peaked during the tiller and mature phases. *Culex tritaeniorhynchus* Giles, *Cx. gelidus* Theobald, *Cx. fuscanus* Wiedemann and *Ficalbia luzonensis* Ludlow were affected by light intensity and water pH found during fallow and plough phases. The relationships of all mosquito species distribution and abundance to associated aquatic insects distribution were significantly positive. Three larvicides, Abate®, 96.2%, *Bacillus thuringiensis* var. *israelensis* (serotype 14) VectoBac® technical powder and *Bacillus sphaericus* Neide (serotype $H5_a5_b$) Vecolex-WDG®, were evaluated against field collected mosquito species, *Culex bitaeniorhynchus, Cx. tritaeniorhynchus, Cx. gelidus, Cx. vishnui* and *An. sinensis* in the laboratory. The levels of susceptibility of these species to the test larvicides were different; the LC_{50} values ranged from 0.0000001 ppm to 0.009 ppm and LC_{90} values

ranged from 0.0000012 ppm to 0.242 ppm for *An. sinensis, Cx. tritaeniorhynchus, Cx. gelidus, Cx. bitaeniorhynchus* and *Cx. vishnui*, in the order of susceptible to the Abate®. The LC_{50} values ranged from 0.000108 ppm to 0.029 ppm and LC_{90} values ranged from 0.000509 ppm to 0.142 ppm for *Culex gelidus, Cx. tritaeniorhynchus, Cx. bitaeniorhynchus, Cx. vishnui* and *An. sinensis,* in the order of susceptible to the VectoBac®. While the LC_{50} values ranged from 0.000178 ppm to 33.376 ppm and LC_{90} values ranged from 0.0356 ppm to 355.94 ppm for *Cx. tritaeniorhynchus, Cx. gelidus, Cx. vishnui, Cx. bitaeniorhynchus* and *An. sinensis,* in the orders of susceptible to Vectolex®, 24h exposure, respectively. While LC_{90} values ranged from 0.01 ppm to 7.51 ppm for *Culex tritaeniorhynchus, Cx. gelidus, Cx. vishnui, Cx. bitaeniorhynchus* and *An. sinensis,* in the orders of susceptible to Vectolex®, 48h exposure, respectively. Under field condition, Abate® 500E at 0.28 mg/liter gave (75.3-100%) mortality of *Anopheles* spp. and (75.8-90.5%) mortality of *Culex* spp. 3 wk post-treatment, respectively. VectoBac-WDG® at 0.48 mg/liter yielded (55.52-100%) mortality of *Anopheles* spp. and (51.29-100%) mortality of *Culex* spp. 5-7 days post-treatment, respectively. While Agnique MMF® at 5 l/ha (0.5 ml/m^2) resulted in (84.9-93.7%) mortality of *Anopheles* spp. and (73.4-96.5%) mortality of *Culex* spp. 2 wk post-treatment, respectively. The three larvicides at same rates had no noticeable adverse effects on prevailing macroinvertebrate fauna such as, Corixidae, mayfly naiads, damselfly larvae and Dytiscidae.

CHAPTER 1- INTRODUCTION

1.1. The mosquitoes

1.1.1 Historical background, importance, and distribution

Based on fossil evidence, it is estimated that mosquitoes may have originated in the early tertiary period, some 70 million years ago or even earlier. Mosquitoes, because of their biting nuisance and their role in transmission of deadly human disease organisms are extremely important insects belonging to the Family Culicidae in the Order Diptera. Mosquitoes can colonize a very diverse aquatic habitat types in terms of size and nature, including ponds, swamps, river and stream banks, salt water marshes, polluted water in septic tanks, rock pools, tree holes, discarded domestic containers, discarded tires, plant axils and pitcher plants, rice fields, etc.

Mosquitoes are important vectors of several tropical diseases in humans; including malaria, filariasis, and numerous viral diseases, such as dengue, dengue hemorrhagic fever, yellow fever, and Japanese encephalitis. An estimated two billion people world-wide live in areas where these diseases are endemic (WHO, 1999).

Mosquitoes are distributed throughout the world. Some species exist at altitudes of <14,000 feet; while others can inhabit mines that are 3,760 feet below the sea level. Species range in latitudes northward from the tropics to the Arctic regions and Southward to the ends of the Continents. A wingless species has been reported to exist in Antarctica, while many species do exist in the most remote deserts (Goma, 1966). A few oceanic islands appear to have been free from mosquitoes before the advent of man and modern transport. There are about 3000 species of mosquitoes distributed world-wide. Of these, about 100 species are vectors of human diseases. With *Anopheles* mosquitoes alone, about 380 species occur around the world; some 60 species are sufficiently attracted to humans to act as vector of malaria. A number of

Anopheles species are also vectors of filariasis and viral diseases. About 550 species of *Culex* have been described, most of them from tropical and subtropical regions. Some *Culex* species are important as vectors of bancroftian filariasis and arbovirus diseases, such as Japanese encephalitis. *Aedes* mosquitoes which occur around the world consist of over 950 species. They can cause a serious biting nuisance to people and animals both in tropics and in cooler climates. Most of *Mansonia* mosquitoes are found in marshy areas in tropical countries. Some species are important as vectors of brugia filariasis in south India, Indonesia and Malaysia (Lane and Crosskey, 1993; WHO, 1997).

1.1.2 Life cycle

Mosquito have four distinct stages in their life cycle: egg, larva, pupa, and adult. The females usually mate only once but produce eggs at intervals throughout their life. In order to be able to do so most female mosquitoes require a blood-meal. Males do not suck blood but feed on plant juices. The digestion of a blood-meal and the simultaneous development of eggs take 2-3 days in the tropics but longer in the temperate zones. The gravid females search for suitable places to deposit their eggs, after which another blood-meal is taken and another batch of eggs is laid. Depending on the species, a female lays between 30 and 300 eggs at a time. Many species lay their eggs directly on the surface of water, either singly (i.e. *Anopheles*) or stuck together in floating rafts (i.e. *Culex*). In the tropics the eggs usually hatch within 2-3 days. Some species (i.e. *Aedes*) lay their eggs just above the water line or on wet mud; these eggs hatch only when flooded with water (Clements, 1992; WHO, 1997).

Once hatched, the larvae do not grow continuously but metamorphose in four different instars. The first instar measures about 1.5 mm in length, while the fourth instar is about 8-10 mm. Mosquito larvae feed on yeasts, bacteria and small aquatic organisms. Most mosquito larvae have a siphon located at the tip of the abdomen through which air is taken in and come to the water surface to breath; they dive to the bottom for short periods in order to feed or escape danger. In warm climates the larval period lasts about 4-7 days or longer if there is a shortage of food. The full-grown larvae then metamorphose into a comma shaped pupae. When mature, the pupal skin splits at one end and a fully developed adult mosquito emerges. In the tropics, the pupal period lasts 1-3 days. The entire period from egg to adult takes about 7-13 days under good conditions (Clements, 1992; WHO, 1997).

Female mosquitoes feed on animals and human. Most species show a preference for certain animals or for humans. They are attracted by the body odours, carbon dioxide and heat emitted from the animal or person. The behaviour of mosquitoes determines whether they are important as nuisance insect or vector of disease and governs the selection of control methods. Species that prefer to feed on animals are usually not very effective in transmitting diseases from person to person (Clements, 1992; WHO, 1997).

1.2 Mosquito control

1.2.1 Early control methods

Mosquitoes are deemed either as a source of nuisance or disease-carrying, and their control have been attempted in various ways since the ancient times with the purpose of reducing man-mosquito contact and consequently human suffering. Despite advances in medical science and new drugs, mosquito-borne diseases including malaria, filariasis, dengue and the viral encephalitis remain the most important diseases of humans.

Historically, earlier mosquito control approaches were mainly based on basic source reduction, environmental management and personal protection. Since World War II, disease control methods have relied heavily on broad-spectrum synthetic chemical insecticides to reduce vector populations. The discovery of organic synthetic insecticides in the 1940s and 1950s triggered a shift in mosquito control from the earlier methods to over reliance on chemical insecticides. However, synthetic chemical insecticides are being phased out in many countries due to insecticide resistance in the mosquito population (Yap, 1994). Furthermore, many governments restrict chemical insecticide use due to concerns over their environmental effects on non-target beneficial insects especially on vertebrates through contamination of food and water supplies. As a result the World Health Organization is facilitating the replacement of these chemicals with use of biological control agents, microbial agents in particular and insect growth regulators, both juvenile hormone mimics and chitin synthesis inhibition (Federici *et al*., 2003). At present, mosquito control approaches can be divided into four categories: (1) Source reduction and environmental management, (2) Biological control, (3) Chemical control and (4) Physical barriers and personal protection.

1.2.2 Chemical control

Among the various control approaches, chemical control has been the mainstay for mosquito control since the advent of organic insecticides in the 1940s. In view of the conventional usage and limited development of alternatives, chemical insecticides, including organochlorines, organophosphates, carbamates, and synthetic pyrethroids, will continue to remain as the norm for vector control in the near feature. The trend in chemical insecticide development is to improve efficacy against mosquitoes as well as to reduce the adverse environmental impact including safety to users. Studies on chemical larvicides including newer organic insecticides have been carried out in many countries.

1.2.3 Insect growth regulators

Based on their mode of action, the insect growth regulators (IGRs) can be divided into juvenile hormone mimics (JHs) and chitin synthesis inhibitors (CSIs). Both groups interfere with development processes (metamorphosis) of immature insects. The CSIs act when mosquito larvae are molting and prevent the synthesis of chitin, an indispensable component of the insect cuticle. The JHS interferes with the transformation of immature insect structures into adult structures. Hence, IGRs are specific for arthropod pests. IGRs which include JHs (e.g. methoprene, pyriproxyfen) and CSIs (e.g. diflubenzuron, triflumuron) have been shown to be effective for the control of immature mosquito with the necessary residual effects (Mulla, 1991).

1.2.4. Biological control

Biological control can be defined as the use of biological agents such as, pathogens, parasites and predators for the control of pests. In general, the use of microbial bacterial agents, such as *Bacillus thuringiensis* H-14 (*B.t.i.*), *Bacillus sphaericus* (*B.sph.*) and recently *Pseudomonas fluorescens* are proven effective against various species of mosquitoes. Due to specificity, *B.t.i.* was found to be more effective against *Aedes* and *Anopheles* (Yap *et al*., 1991; Karch *et al*., 1991; Sadanandane *et al*., 2003 and Gunasekaran *et al*., 2004).

Vector control products based on bacteria are designed to control larvae. The most widely used commercial products are VectoBac® and Teknar® which are based on *Bacillus thuringiensis* subsp. *israelensi*s (*B.t.i.*). In addition VectoLex®, a product based on *Bacillus sphaericus* (*B.sph*.), has come to market recently for the control of mosquito vectors of filariasis and viral diseases. The high efficacy that *B.t.i.* showed in laboratory and field trials during the early 1980s led rapidly to its development as a commercial bacterial larvicide for control of mosquito and black fly larvae (Federici *et al.*, 2003).

Biocontrol agents are getting increasingly popular in controlling larval mosquito populations. Among these natural agents are bacteria and larvivorous fish (Asimeng and Mutinga, 1993). Variety of *Bacillus thurigiensis* Berliner and *Bacillus sphaericus* Neide have been widely tested and employed for larval mosquito control in various situation and have proved their effectiveness in rice fields (Sandoski *et al*., 1985).

The successful use of microbial control agent is based upon preparation for the campaign of mosquito control. The prerequisites are: knowledge must be obtained on the larval habitats, which must be carefully mapped, characterized, and also numbered, so they can be identified rapidly during routine operations. Also a precise assessment must be made of the entomological data, such as the composition of and fluctuation in the larval and adult mosquito populations. Moreover, the World Health Organization (WHO) recommended an emphasis on vector control, including biological control and environmental management. Knowledge of the relationships between habitats, environmental factors, and occurrence of mosquito larvae is essential for an efficient application of mosquito control methods (Fischer and Schweigmann, 2004).

Adequate information must be obtained on the climatic factors that influence mosquito densities, such as the occurrence of rainy seasons. The potency and efficacy of the control agent have to be assessed in the laboratory and in various larval habitats. The most appropriate formulation has to be selected, and the sequence of follow-up treatments has to be adapted to the local situation. The spray equipment has to be adapted to the specific characteristics of the product. A proper design of the control strategy must be made based on the results obtained in the preparatory phase (Becker and Rettich, 1994).

1.3. Role of rice ecosystems in abundance of mosquito populations

Rice is the most important cereal crop in the developing world and some parts of the developed world. Rice is the staple food for over half of the world's population and is the number one cultivated crop in Asia. Rice cultivation is thought to be the oldest form of intensive agriculture by man. For example, in Madagascer, at the end of the 18th century, Marina Kings developed land irrigation and rice cultivation using manpower from the coasts until the rice has become a monoculture covering most of the arable land of the highlands (Laventure *et al*., 1996).

To date, some 140 million hectares of land are devoted to growing rice, 90 % of this is both cultivated and consumed in Asia. Rice feeds humans more than any other crop; it is the staple food of over 60 % of the world's population (Consultative Group on International Agricultural Research *et al*., 1998; IRRI, 2002; Riceweb, 2002).

Rice is an annual grass belonging to the genus *Oryza* that has two main species each with a great number of varieties, *Oryza sativa* in Asia and *Oryza glaberrima* in West Africa. Some 120.000 varieties of rice are known worldwide and research continues in developing and promoting new rice varieties. Field duration of rice crop is usually between 90 and 120 days, but new varieties may mature earlier (Keiser *et al*., 2002; Bambaradeniya and Amerasinghe, 2003).

Increases in productivity have been achieved through growing high-yielding varieties (HYV). Most rice cultivators require fields to be flooded for varying periods. In fact, HYVs are especially sensitive to water shortage and drought, and generally need more water, at least during part of their development than the less productive strains. Flooding does not only achieve better growth and crop yields but also reduces the soil toxicity and controls weeds.

Cultivated rice has one of the two main systems: upland (dry rice) or lowland (wet rice); upland rice does not require flooding for its growth and can be cultivated like other cereal crops even in mountainous areas. On the contrary, lowland rice requires constant irrigation and is maintained in 10-50 cm of water (Ijumbo and Lindsay, 2001). However, rice is most commonly cultivated as a lowland crop. Water affects the physical character of the plants as well as the nutrient and physiochemical characteristics of the soil.

Flooding also benefits the crops by providing nutrients, fresh clay particles and organic matter that enrich the soil with nitrogen and blue–green algae which fix atmospheric nitrogen. This increased the availability of phosphorus, iron and manganese compounds and helps control weed growth (Ijumbo and Lindsay, 2001; Keiser *et al.,* 2002).

This widely practiced system of growing rice in mostly stagnant water provides the habitat for mosquitoes that serves as vectors of malaria, filariasis and various arboviruses as well as for the snail that serves as intermediate hosts of schistosomiasis. Over 40 viruses have been identified in studies on rice fields agrosystems, but by far, the most important is Japanese encephalitis. The association of rice cultivation with ill health has been known for centuries, for example, in the 1300s the cultivation of rice in Valencia in Spain was forbidden because of health reasons. The term rice malaria was coined in the 1938s to describe the association of rice cultivation in Europe with malaria. It is also believed that the malaria epidemics in the 1600s in the Carolinas in the United States resulted from the introduction of rice agriculture (Service, 1989).

1.4 Objectives

The main objectives of this study are as follows:

1. To intensively survey rice field habitats for qualitative and quantitative determinations of mosquitoes, including species distribution and determination of population age composition and succession in relation to growth stages of rice.

2. To establish seasonal population density patterns of mosquito species and co-existing aquatic insects including predators, and to estimate variation rate of both populations attributed to the rice culture practices, such as insecticide and herbicide applications against rice pest insects, weeds and fertilizers used in the rice fields. This may provide essential information on developing an effective integrated control, and give thorough information to agricultural authorities on how to modify and improve the rice culture management to maximize control effect against the vector mosquitoes and to minimize its side effect on the non-target organisms particularly their predators.

3. To include samples from the rice field habitats showing distribution and abundance of mosquito species and associated aquatic insects which are common to the rice field agroecosystem. Station and habitat similarities and differences will be described and correlations of mosquito and associated aquatic insect numbers with environmental and biological parameters will be shown.

4. Laboratory and field evaluations to determine efficacy of *Bacillus thuringiensis* H-14, *Bacillus sphaericus,* temephos (Abate®) and Agnique® MMF as larvicides against mosquito species; and impact of this larvicides on non-target organisms in the field.

CHAPTER II- LITERATURE REVIEW

2.1 ECOLOGY OF MOSQUITO SPECIES AND ASSOCIATED AQUATIC INSECTS IN THE RICE FIELDS AGROECOSYSTEM

2.1.1 MOSQUITO SPECIES AND ASSOCIATED AQUATIC INSECTS BREEDING IN RICE FIELDS AND ASSOCIATED CANALS

Increased rice production inevitably results in the expansion of mosquito larval habitats. About a quarter of the 60–odd *Anopheles* species listed as important vectors of human malaria breed in rice fields. Furthermore, rice fields also produce important mosquito vectors of human filariasis and viral diseases (Mogi, 1984). The relative abundance of mosquitoes breeding in these rice fields varies extensively by season and spatially. This variation has been attributed to the different rice cultural practices (Chambers *et al*., 1981; Stark *et al*., 1984; Chandler *et al*., 1991; Abu Hassan, 1994). Although the principal vector of Japanese encephalitis (JE), *Culex tritaeniorhynchus* and other culicine mosquitoes that can transmit this disease (i.e., *Culex bitaeniorhynchus, Culex epidesmus, Culex fuscocephala, Culex gelidus, Culex pseudovishnui, Culex sitiens, Culex vishnui* and *Culex whitmorei*) (Gajanana *et al*., 1997; Sehgal and Dutta, 2003), are able to breed in ground water habitats, such as sunlit pools, roadside ditches, tidal marshes of low salinity or man-made containers, their preferred major larval habitats are rice fields (Mogi, 1984). Rice agrosystem perfectly fits the ecological requirement of vectors and in fact malaria and JE are important vector–borne diseases associated with rice production in developing countries (Sehgal and Dutta, 2003).

In Africa (Lindsay *et al*., 1991), rice fields provide a wide range of mosquito breeding habitats, specifically suitable for pioneer species, such as members of the *Anopheles gambiae* complex, the main vectors of malaria. In Madagascar, Robert *et al*. (2002) observed that rice fields are efficient breeding places for malaria vector *An.*

gambiae. The larvae were found in rice fields that do not have emergent vegetation and thus exposed to the sun.

In Texas and Arkansas, USA, (Kuntz *et al*., 1982; Peloqunin and Olson, 1985) rice fields and associated pastures represent a significant source of mosquito breeding sites for the dark rice field mosquito, *Psorophora columbiae,* and the malaria mosquito, *An. quadrimaculatus*, and several other species in Arkansas. Olson and Meek (1980) found that levees in fields planted with rice as well as the fields maintained in a fallow state represent an important source of oviposition sites for *Ps. columbiae*. The adjacent irrigated rice fields were the only major source of mosquito, and mosquito densities were related to the water regimens in the rice fields (Snow, 1979). In southwestern Louisiana, USA (Mclaughlin and Vidrine, 1987), rice land irrigation provides additional breeding habitat for the mosquito species inhabiting permanent water, as well as those breeding in temporary pool, such as *Ps. columbiae*.

In Malaysia (Yap and Ho, 1977) found that *Anopheles campestris*, *Cx. tritaeniorhynchus* and several other species of *Anopheles* and *Culex* commonly occure in rice fields. Amerasinghe and Indrajith (1994) found that usually the larval populations are low after transplanting of the rice seedlings, reaching a peak a few weeks later and declining thereafter as the plants reach a height of 60 cm. This population decline is usually due to the physical obstruction caused by the plant growth that provided extensive cover. Another factor could be the establishment of predators in the fields. The level of mosquito breeding diminishes when the fields are drained before harvesting; and if same water remains, mosquito breeding continues at a low level in the residual pools and rain–flooded fields (Abu Hassan, 1994). When the fields become completely overgrown with vegetation, and flooding occurs from time to time, only *Anopheles* and *Culex nigropunctatus* were found to breed. Abu Hassan *et al*. (1998) found 29 mosquito species biting human and bovid hosts, and 11 of those breed in rice fields in Muda area of Malaysia. *Anopheles sinensis* (Jaal and Macdonald, 1993) is a common and widely–distributed rice field species in northwest coastal Malaysia, occuring in exposed irrigation canals, grassy pools and ditches. Among seasonal habitats, rice fields provide a major breeding source for *Cx. tritaeniorhynchus, Cx. pseudovishnui*, *Anopheles peditaeniatus* and *Anopheles vagus*. *Culex fuscocephala* was also found in this habitat in high abundance where decaying vegetation was used as a source of organic fertilizer (Amerasinghe and Indrajith, 1994).

In Indonesia (Service, 1989), dominant rice field anopheline mosquitoes are *An. aconitus* and *An. barbirostris*, and an increased prevalence of malaria even leading to epidemics has been attributed to double rice cropping in a year. Similarly, in parts of China, two population peaks of adult mosquitoes occur in areas supporting two rice crops and a single peak in areas where only one crop a year is grown (Service, 1989).

In India, Kant *et al*. (1996, 1998) observed 14 anopheline and 15 culicines species in rice fields of Gujrat, with *An. subpictus* and *Cx. vishnui* as the two dominant species. These authors have also reported association of mosquito species with prevailing algae. Singh *et al*. (1989) stated that *An. culicifacies* populations, initially in large numbers, gradually decline with increasing height of rice plants, eventually becoming scarce and replaced by other anopheline species which also breed in irrigation channels almost throughout the rice growing season. Extensive breeding of *An. culicifacies* was found in the rice field channels and in rice fields (Kant and Pandey, 1999).

In Iran, the distribution of the Iranian culicinae studied by Zaim (1987) revealed that 33 species of Culicinae belonging to four genera were found. Species such as *Culex modestus, Culex pusillus, Cx. bitaeniorhynchus, Cx. pseudovishnui, Cx. quinquefasciatus* and *Cx. tritaeniorhynchus* were the most common inhabiting the rice fields. While Takagi *et al*. (1997) observed that among the 8 *Culex* species collected in 3 areas of northern Thailand, *Cx. tritaeniorhynchus, Cx. vishnui* and *Cx. gelidus* were predominant.

In Sri Lanka, Amerasinghe and Munasingha (1988) and (Amerasinghe and Ariyasena(1991), recorded 49 species during the 12-month phase of human settlement and infrastructure construction, and 42 species during the succeeding 12-month-period under irrigated rice culture. Development resulted in the elimination of some pre-existing breeding habitats. The overall changes from uninhabited forest to settled irrigated rice field sharply increased the prevalence of *An. annularis, An. peditaeniatus, Aedeomyia catastica, Mimomyia hybrida, Mansonia uniformis* and *Cx. tritaeniorhynchus*. Amerasinghe (1993) stated that 26 species of mosquitoes were found in rice fields of the dry zone in the Eastern province of Sri Lanka, while Bambaradeniya (2000a) recorded 14 species in rice fields in the central zone.

In Japan, Toma and Miyagi (1992) studied the effect of rice plant canopy on the density of mosquito larvae and other insects. They observed the species predominating in open water habitats in cultivated areas, paddy fields and other flatland areas including swamps, ponds, and ground pools were *An. sinensis*, *Cx. bitaeniorhynchus, Cx. pseudovishnui* and *Cx. tritaeniorhynchus*.

In the Philippines, *Cx. tritaeniorhynchus* was shown to have 2 or 3 population peaks each year depending on the weather conditions, with 2 or 3 rice crops grown each year (Schultz and Hayes, 1993 and Takagi *et al*., 1995 & 1996). Population abundance of *Cx. gelidus* was directly correlated with total monthly rainfall.

Lacey and Lacey (1990) found that invertebrate predators, i.e., Coleoptera, Hemiptera or Odonata are known to substantially reduce mosquito larval populations in rice fields. However, these predators are highly sensitive to temperature, presence of vertebrates, growth of rice and chemical pollutants. These workers also did a comprehensive review of the mosquitoes in rice fields, covering aspects of their ecology, medical importance and control, and listed 137 species of mosquitoes that breed in rice fields worldwide. They provided details of the prevalent species breeding in different habitats within the rice ecosystem as well as within the rice fields during different stages of the rice cultivation cycle.

In India the presence of notonectids was negatively associated with larval abundance of *Cx. pseudovishnui*, *Cx. tritaeniorhynchus* and *Cx. vishnui* (Sunish and

Reuben, 2002). Studies on the biological control of mosquito larvae revealed that natural populations of larvivorous cyclopoid copepod species is useful for controlling *Anopheles*, *Culex* and *Aedes* larvae (Marten *et al.,* 1989; Marten, 1990; Kay *et al*., 1992; Marten *et al.,* 1994; dos Santos *et al.*, 1997; Rawlins *et al*., 1997; Marten *et al*., 2000a, 2000b; Dieng *et al*., 2002 and Zoppide de Rao *et al*., 2002).

According to Bambaradeniya and Amerasinghe (2003), mosquitoes are the most widely studied aquatic insects associated with the rice fields since this ecosystem constitutes the favored breeding sites of several species. Only a few researchers have studied aquatic insects other than mosquitoes in rice fields. Yano *et al*. (1983) recorded 117 species of aquatic coleopterans in 14 families in rice fields worldwide. A study in the Muda rice area of Malaysia showed several other orders: Diptera (Family: Chironomidae and Culicidae), Coleoptera (Family: Hydrophilidae and Dytiscidae), Hemiptera (Family: Corixidae, Pleidae, Nepidae, Belostomatidae), Odonata (Family: Libellulidae, Coenagrionidae) and Ephemeroptera (Family: Baetidae) comprised the aquatic insect fauna. The dominant aquatic insects were from the families: Chironomidae, Dytiscidae, Corixidae and Belostomatidae and the aquatic representations of the Coleoptera, Hemiptera and Odonata were all predatory insects (Rozilah and Ali, 1998).

Lee (1998), in his studies in Korea at Bulkyo, Bosong-gun, and Chollanamdo rice fields stated that the total number of aquatic insect taxa in these fields were 25 species belonging to 22 families in 10 orders.

Che Salmah (1996) in Malaysia found that aquatic insects, such as, Hydrophilidae, Dytiscidae, Nepidae, Vellidae, Gyrinidae, Dragonflies and mosquitoes were abundant in rice fields. Some of them, such as, Gyrinidae and Dytiscidae were early colonizer but few others can be found throughout the season and they laid eggs as soon as the rice fields were inundated with water.

Victor and Reuben (1999), in South India studied the breeding pattern of immature mosquitoes and the successional changes in the abundance of aquatic insects, stating a total of 14 families consisting of 17 subfamilies of aquatic insects belong to five different orders; Coleoptera, Diptera, Ephemeroptera, Hemipterta, Odonata (Anisoptera and Zygoptera). The study also revealed that Notonectidae, Libellulidae, and Vellidae acted as predators of immature mosquito in rice fields. Water from irrigation canals was considered to be a major source of colonization by the predators of mosquito larvae and could limit breeding seasons and prolong developmental periods of mosquitoes (Mogi, 1993).

In Indonesia, aquatic habitats of mosquitoes and larvivorous predators in deforested lands in central Sulawesi were studied by Mogi *et al* (1999), observing Anisoptera and Zygoptera nymphs, Dytiscidae and Notonectidae as dominant among aquatic predators.

In New Zealand, Lester and Pike (2003) stated that invertebrate predators can directly and indirectly influence mosquito population dynamics. For example, the backswimmer, *Notonecta* spp. (Hemiptera: Notonectidae) is highly predaceous, consuming large numbers of mosquito larvae and thereby influencing aquatic communities. In addition, the presence of backswimmers in a body of water can significantly reduce oviposition by adult mosquitoes. Other commonly observed larval predators of mosquitoes included the diving beetle and damselfly larvae.

In Laos and Thailand, Heckman (1974 and 1979, Bambaradeniya and Amerasinghe, 2003) carried out comprehensive studies on rice field organisms. He found that the insects were the dominant group of aquatic invertebrates observed during surveys.

2.1.2 THE EFFECT OF CULTURAL PRACTICES ON ABUNDANCE OF MOSQUITO SPECIES AND ASSOCIATED AQUATIC INSECTS IN RICE FIELDS

Cultural practices that have been shown to be important in determining mosquito populations are: land rotation, water management, application of chemicals to control insect pests of rice, and application of chemicals to control plant pests in rice fields (Chambers *et al*., 1981). However, increasing urban agriculture is thought to play a major role in increasing malaria in urban areas as it increases breeding sites for immature *Anopheles* spp. It also raises the economic status of the population allowing improved malaria protection (Afrane *et al*., 2004).

In Malaysia, the rice fields in small plots with vegetation cut close to the ground are left lying under water, when degrade, create an environment conducive to enhance breeding of JE vector, *Cx. tritaeniorhynchus* (Heathcote, 1970). The same author reported, heavy oviposition lead to large populations of the mosquito, up to 40 pupae per m^2.

In Egypt (Morsy *et al*., 1995), rice fields were infested with *Culex antennalus* and *Anopheles pharoensis*. These two species prefer clear, shallow, stagnant water with thick growth of vegetation. The breeding waters for these mosquito species are usually alkaline.

Japanese encephalitis vectors abundance is closely related to agro-climatic features (Peiris *et al*., 1993 and Phukan *et al*., 2004), most notably temperature and monthly rainfall (Solomon *et al*., 2000 and Bi *et al*., 2003). In addition, potential JE vectors were rarely found at altitudes above 1200 m. However, the most important causative factor of JE vectors is the management of paddy water; the peak periods of mosquito abundance are associated with cycles in local agricultural practices.

In Thailand, the highest numbers of larvae and pupae of JE vectors were collected when the rice fields were ploughed with water in the fields (Keiser *et al*., 2005). The vector population decreased after transplanting when the fields were flooded and stayed low until harvesting (Somboon *et al*., 1989).

An ecological study carried out by Lee (1998) to compare organically and conventionally farmed rice fields in Korea during the rice growing periods revealed abundance of two vector mosquitoes, *An. sinensis* and *Cx. tritaeniorhynchus*, which were lower in the organically farmed rice fields compared to the conventionally farmed rice fields. The application of urea, a nitrogenous fertilizer in rice fields, significantly increased the grain yield and the population densities of mosquito larvae and pupae.

In Indian rice fields, synthetic nitrogenous fertilizers were found to be responsible for a significant increase in anopheline and culicine larval populations (Victor and Reuben, 2000). Fields treated with inorganic fertilizers (N.P.K.) had significantly increased population densities of immature mosquitoes than fields treated with organic manure, i.e. farmyard manure and green manure.

In Kenya, peaks of *An. arabiensis* larvae were found to coincide with both rice transplanting and application of ammonium sulphate fertilizer (Mutero *et al*., 2000). Sunish and Reuben (2001) found that the height of the rice plants, water temperature, dissolved oxygen, ammonia nitrogen and nitrate nitrogen strongly influence the abundance of immature mosquitoes in India. Application of synthetic nitrogenous fertilizers to the rice fields was followed by a rise in concentration of ammonia nitrogen and a subsequent increase in nitrate nitrogen level in the rice field water, which caused an increase in the density of mosquito larvae. Predominant species observed were of the *vishnui* subgroup, *Cx. tritaeniorhynchus*, *Cx. pseudovishnui* and *Cx. vishnui* (Sunish and Reuben, 2002). More recently, results of a study by Mutero *et al.* (2004a) on the effect of ammonium sulphate fertilizer on mosquito larval populations in rice fields showed a significant overall increase in the larval populations of *Anopheles arabiensis* and culicine mosquitoes after ponds were treated with the fertilizer. Significantly more fourth instar larvae of *An. arabiensis* were collected in fertilized plots than in control plots. They also found that the first application had the most impact compared with the second and third applications. The studies suggest that ammonium sulphate fertilizer reduces turbidity of water in rice fields, thereby making them visually more attractive for egg-laying by *An. arabiensis* and culicine mosquitoes.

In Taiwan, studies on ten rice fields indicated that the size of the mosquito population was mainly related to flooding and drying practices and the application of insecticides against insect pests of rice (Cates, 1968). The larval habitat of mosquitoes was eliminated during the drought period and larval populations of *Cx. vishnui, Cx*. *tritaeniorhynchus, Cx. bitaeniorhynchus*, and *An. sinensis* disappeared. The decline in the mosquito population also coincided with the first application of insecticide against rice pests. Mclaughlin *et al*. (1987) observed that manipulation of arable land for rice field production has created another environment for anophelines.

Species composition and breeding pattern of mosquitoes in relation to rice cultivation in north peninsular Malaysia studied by Rashid *et al*. (1995) showed that *Cx. tritaeniorhynchus* was the dominant mosquito species in the agro-ecosystem followed by *Cx. gelidus, An. peditaeniatus*, *An. sinensis, Cx. bitaeniorhynchus* and *Cx. pseudovishnui*. Main breeding habitat was the rice field but the irrigation canals, drainage ditches and patches of water pools also served as additional breeding habitats. Mosquito breeding was studied in relation to rice farming schedule and water filling schedule. *Culex* spp. and *Anopheles* spp. started to appear when the rice fields were inundated with water and ploughed. Breeding continued until the water was drained off when the rice is ready to be harvested. *Culex* spp. were dominant after the rice was harvested. Hoof prints of cattle and tire tracks made by harvesting equipment in recently harvested rice fields were considered to be an important source of oviposition sites for populations of *Ps. columbiae* breeding in Texas ricelands (Meek and Olson, 1977).

In Kenya, mosquito fauna of rice fields were divided into two groups (Chandler and Highton, 1975). The first consisted of species which breed through the rice cycle and were little affected by changes in water depth and rice height. The second group appears to be limited to certain stages within the rice cycle. Anophelines occured largely during the early rice growth, while *Culex* spp. appeared later in the cycle.

Gahan and Wilson (1969) found *Psorophora* spp. to be predominant, while the rice fields are being alternately flooded and dried, but when they remain flooded continuously, they disappear. *Anopheles quadrimaculatus* then becomes prevalent until the rice fields are drained. Flood application and any subsequent water level fluctuation create breeding conditions favourable to floodwater species, such as *Ps. columbiae*, and the maintenance of water in the fields is favourable to standing water breeders like *An. quadrimaculatus* (Stark and Meisch, 1984; Sandoski *et al*., 1987).

The relationship of insect predators and phytoplankton with the abundance of *Cx. tritaeniorhynchus*, *Cx. vishnui* and *Cx. pseudovishnui* mosquito larvae and pupae in rice fields was investigated by Sunish and Reuben (2002) during three rice growing seasons. Notonectidae were the most abundant insect predators, whereas diatoms dominated among phytoplankton. However, blue-green algae increased paddy yield without enhancing mosquito production (Victor and Reuben, 2000).

2.1.3 THE SUCCESSION OF MOSQUITO SPECIES IN RICE FIELDS

The aquatic community of organisms in rice fields is a dynamic system related closely to rice plant growth, rice cultivation practices, and seasonal climatic changes. Each mosquito species often has a preference for a particular phase in rice field development, which may result in an orderly succession of species as the rice plants develop and mature. The abundance of aquatic macro invertebrates, including predators, also changes during the growth of a single rice crop (Mogi and Miyagi, 1990).

Somboon *et al*. (1989) found that *Cx. tritaeniorhynchus, Cx. gelidus* and *Cx. fuscocephala* were the main vectors of JE in Amphoe Muang, Chiang Mai, Northern Thailand. The JE vectors showed a sharp rise in the population in July when most of rice fields were ploughed and a marked decline in mosquito population densities occurred after transplanting in August when the fields were flooded. The average number of larvae and pupae per m² in rice fields was highest in July when the fields were ploughed, but the densities decline between transplanting and harvesting.

Studies on ecological succession and association of anophelines were made in the paddy fields of India. Sharma and Prasad (1991) and Kant *et al*. (1992) observed breeding of three anophelines: *An. culicifacies, An. subpictus* and *An. nigerrimus*. Breeding of *An. culicifacies* and *An. subpictus* occurred in the early stage of rice cultivation and stopped after rice transplantation. *Anopheles nigerrimus* breeding started nearly 30 days after rice transplantation, just after *An. culcifacies* and *An. subpictus* stopped breeding, and continued until harvesting. An inverse correlation between larval density of both *An. culicifacies* and *An. subpictus* and the height of the plants were observed. *Anopheles culicifacies* was found in abundance in newly transplanted rice fields and during early months of rice cultivation with a peak prevalence in the non–monsoon (Rabi) season (Kant and Pandey, 1999).

In Egypt, larval abundance of *Cx. antennalus, An. pharoensis* and *Cx. perexiguus* varied monthly according to the stage of rice growth (Kenawy *et al*., 1998; el Shazily *et al*., 1998). The plant height and irrigation practices were considered to be major factors affecting the abundance of these species. In Kenya, mosquito larvae were most abundant in nursery paddies in which the plants were well spaced out and were not more than 25 cm heigh (Surtees, 1970).

A study of Koraput district of Orissa state paddy fields with a perceptible water flow top–hill area supported heavy breeding of the principal vector *An. fluviatilis* at all stages of paddy growth (Sadanandane *et al*., 1991). *Anopheles culicifacies* and *An. annularis* breeding became scarce when the paddy plants reached a height of 80 cm. The maximum production was during early stage of paddy growth or after harvesting.

Snow (1983) observed succession on different species of mosquitoes in Gambia during irrigation of the rice fields through one cycle of irrigated rice cultivation.

Few mosquitoes existed before the irrigation but reached its peak four weeks after full-scale irrigation and then declined in abundance. Others were common around the middle of the rice–growing cycle 6–13 weeks after the start of full irrigation and showed more extended peaks of abundance. *Anopheles ziemanni* reached its peak as the rice crop neared maturity.

Forattini *et al.* (1994) in their study on breeding of *An. albitarsis* in association with rice growth in irrigated paddy fields, observed that breeding occurred in each stage up to five weeks after transplantation. Population densities of *Cx. tritaeniorhynchus, Cx. gelidus* and *An*. *sinensis* follow rice-cropping patterns at certain stages of rice production. Manipulation of seedling beds reduced mosquito propagation early in the season (Lu, 1987; Tsuda *et al*., 1998).

Marquetti *et al*. (1991) observed three groups of mosquitoes with respect to its growing pattern in association with the rice growing cycle in Cuba. The first group, which were previously breeding in the natural swamp areas, increased in number as a result of irrigation of rice fields, but show little relation between their numbers and the cycle of rice growth. The second group was the primarry colonizers of the rice fields. The third group is typical in the intermediate stage of the rice–growing cycle.

2.1.4 MOSQUITO SPECIES AS VECTOR OF DISEASES

All crops that require standing water for irrigation purposes usually support mosquito populations. Development of the rice fields very often lead to an increase in malaria, filariasis or arthropod–borne virus infections in humans. Irrigation is a key strategy to enhance crop production system, but it often results in negative health outcomes and is consequential to the increased frequency and transmission dynamics of water-associated infectious disease (i.e., schistosomiasis) or water-related vector-borne disease (i.e., malaria) (Surtees, 1970; Brandling-Bennett *et al*., 1981; Li *et al*. 1981; Welch and Olson, 1987; Carnevale *et al*., 1999; Herrel *et al*., 2001; Girardin *et* *al*., 2004). Rice fields generally constitute an important source of vector mosquitoes. The use of irrigation to flood agricultural land during rice cultivation has over the years been associated with an increase in the number of disease vectors and in certain cases a corresponding increased health burden due to malaria and other vector and water-borne diseases (Lacey and Lacey, 1990; Dossou-Yovo *et al*., 1998; Ijumba and Lindsay, 2001). Among the common mosquito–borne diseases of humans, malaria and filariasis have been the most common for centuries. This is true in many parts of the tropics and subtropics. Malaria was strongly associated with water logging, with poor maintenance of irrigation systems and with rice cultivation (Yaghoobi-Ershadi *et al*., 2001; Hamad *et al*., 2002; Killeen *et al*., 2002; Boelee, 2003; Chimbari *et al*., 2004; Dolo *et al*., 2004; Klinkenberg *et al*., 2004; Mutero *et al*., 2004; Sissoko *et al*., 2004).

In Malaysia, field and laboratory observations on *An. sinensis* in relation to transmission of brurgia filariasis was observed in one of 39 *An. sinensis* infected with subperiodic *B. malayi* (Chiang *et al*., 1986). This species was the most prevalent in the area and displayed 2 peaks of abundance in a year. *Anopheles sinensis* is prominent in many Asian malaria transmission situations. However, several species of *Anopheles* including *An. campestris* are the main vectors of malaria in West Malaysia. *Culex tritaeniorhynchus, Cx. sitiens* and *Cx. gelidus* are the main vectors of Japanese B encephalitis (WHO, 1969 in Yap and Ho, 1977; Vythilingam *et al*., 1994). In Sarawak, Heathcote (1970) isolated JE virus almost exclusively from *Cx. gelidus,* a pig-biting mosquito, and the rice–field breeder *Cx. tritaeniorhynchus*, a species that bites man as well as pigs.

Irrigation, especially in rice cultivation has a strong association with malaria vectors as many anophelines lay their eggs in the flooded rice fields, in the irrigation and drainage canals (Klinkenberg *et al.,* 2003). Much research on this subject matter has been done in Asia. Mogi *et al*. (1995) considered rice fields as important larval habitats for mosquito vectors of human diseases in tropical Asia. This increase in potential habitats for immature mosquitoes also may accompany changes in habitat quality for mosquito reproduction; mosquito immature populations increase quickly in flat rice fields.

The recent expansion of rice–growing areas has facilitated the increased production of mosquitoes in tropical countries. In Thailand, several *Anopheles* species breeding in rice fields are considered actual or potential vectors of human malaria and filaria. On the other hand, JE occurs sporadically throughout the year, with potential to become epidemic in the rainy season between May and August in Northern Thailand. The disease appears in the areas where the animal farms, i.e., pigs and cattle are near houses, and humans dwell beside the rice fields where the vectors breed (Mogi *et al*., 1986; Poneprasert, 1989).

Japanese encephalitis virus was first isolated in Java, Indonesia from pooled female *Cx. tritaenIorhynchus;* whereas, in the Philippines, rice fields are the main breeding habitat for potential vectors of Japanese B encephalitis, such as, *Cx. tritaeniorhynchus*, *Cx. vishnui, Cx. bitaeniorhynchus, Cx. fuscocephals* and *An. annularis* (Hoedojo *et al*., 1980; Mogi *et al*., 1980a,b & 1984; Olson *et al*., 1985; Lilian and Leagas, 1989 and Schultz and Hayes, 1993).

In Sri Lanka, the species of medical importance in the Mahaweli project are *An. jamesii, An. culicifacies, Cx. gelidus, Cx. pseudovishnui, Cx. tritaeniorhynchus* and *Cx. vishnui* (Amerasinghe and Indrajith, 1994). These species used breeding habitats associated with irrigation development, i.e., canals, reservoirs, ponds, rice fields as well as natural habitats. The ecology of *Culex* spp. in rice fields had been studied by Amerasinghe and Ariyasena (1991), and Amerasinghe (1995). They found that a high density of JE vector species are in rice fields. The impact of these man-made breeding sites is much more important than that of natural breeding places. For example, a significant increase in the abundance of *Cx. tritaeniorhynchus,* and increased human-vector contact have been noted following completion of the large rice-irrigation scheme in the Mahaweli project. *Culex tritaeniorhynchus,* the vector of Japanese encephalitis and *An. sinensis,* the vector of both malaria and land-filariasis breed mainly in the rice fields (Bahang *et al*. 1984). Their vector efficiency totally relies on high population densities of human being (Ree *et al*., 1982). In another study carried out in Madagascar (Laventure *et al*., 1996), 95% of *An. funestus,* the main malaria vector, was breeding in the rice fields just before harvest and in the fallow lands. In China, Lu (1987) and Cowper (1988) observed *An. sinensis,* vector of malaria and *Cx. tritaeniorhynchus,* vector of JE, are predominantly rice fields breeders.

Rift valley fever (RVF) virus was diagnosed as the cause of infection in humans and livestock in Jizan region, Saudi Arabia. Both *Cx. tritaeniorhynchus* and *An. arabiensis* were susceptible to RVF virus and transmitted among hamsters (Jupp *et al*., 2002).

In India, mosquitoes of the *Cx. vishnui* subgroup are the most common vectors of JE and *An. culicifacies,* a principal malaria vector, predominantly breed in rice fields (Kant and Pandey, 1999; Sunish and Reuben, 2002). *Anopheles pharoensis* are known to be malaria vectors in Egypt, and *An. gambiae* the most common vector of malaria in sub–Saharan Africa. These species breed in rice fields (El Said *et al*., 1986). A large number of studies have referred to *Cx. tritaeniorhynchus* as the principal vector of JE which breeds mainly in the rice fields (Bendell, 1970; Simpson *et al*., 1970; Self *et al*., 1993; Mogi et al., 1980a,b; Meshra *et al*., 1982; Mishra *et al*., 1984; Thisyakorn and Nimmannitya, 1985; Gingrich *et al*., 1987; Peiris *et al*., 1987; Phanthumachinda, 1989; Somboon *et al*., 1989; Sucharit *et al*., 1989; Suroso, 1989; Vythilingam *et al*., 1993 and1995; Kanojia *et al*., 2003; Arunachalam *et al*., 2004; Phukan *et al*., 2004 and Keiser *et al*., 2005).