

Milestone 2: Report C2

"DRWH AND INSECT VECTORS: A LITERATURE REVIEW"

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Milestone 2: Report C2 of the project

Section I: Mosquito - The Major Disease Vector

1.1 Introduction

The health implications of widespread use of DRWH are divided into two aspects , namely;

- a) Concerns regarding water quality (WQ) and possible direct health implications due to contaminants.
- b) Insect vector breeding related to water storage and health implications arising out of it.

A literature review on DRWH- WQ has been presented in milestone report C1. The current report C2 is on the second aspect.

A review of the literature indicates that mosquito is the major insect vector which needs to be considered in the context of DRWH especially in humid tropics. The current review hence deals essentially with mosquito breeding and related diseases. Since designing for mosquito control requires an understanding of breeding habits, life cycle and behavioral patterns of mosquitoes as well as modes of disease transmission, these are discussed below.

1.2 Mosquito borne Diseases

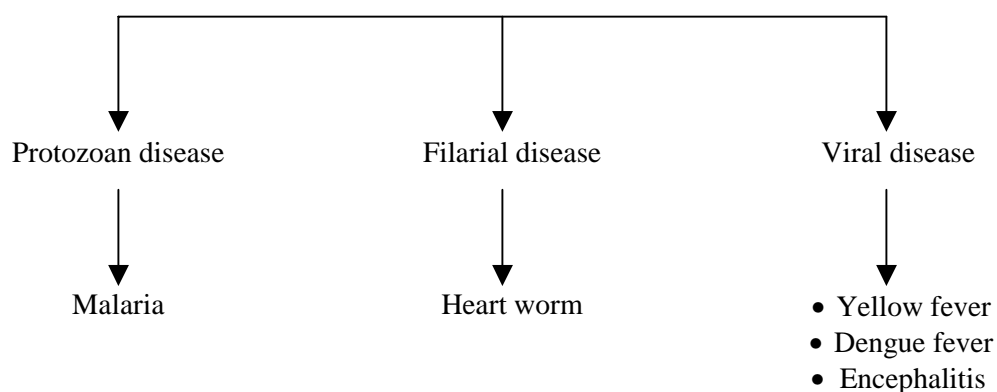
Mosquitoes belong to the Class - Insecta, Order -Diptera and Family - Culicidae. They are thin brown, sized 6.4-12.7 mm, long legged winged insects; adults have three pairs of long, slender legs, and elongate 'beak' or piercing proboscis. Most mosquitoes remain close to the lake, pond or clogged gutter. Rainy season provides plenty of breeding places for them. Breeding of mosquito depends on various factors which include temperature, relative humidity and rainfall pattern. While certain characteristics are common to all mosquitoes, there are differences among different genus and species. Habitat and climate conditions determine the dominance of a particular species in any given area. Larval requirements can also be quite specific to the species. The number of mosquito species under 4 major genus are listed in table 1 along with the number of species which are major disease vectors.

Table 1: Distribution of Mosquito species

Genus	World		India	
	No. of Species	Major Vector	No. of Species	Major Vector
<i>Anopheles</i>	422	60	58	6
<i>Aedes</i>	888	25	111	1
<i>Culex</i>	715	12	57	3
<i>Mansonia</i>	23	7	4	1

Anopheles, *Aedes* and *Culex* are the most common genus and are known to cause various diseases. The major diseases caused by these various species belonging to the sub-family: Anophelinae (*Anopheles*) and Culicine (*Culex*, *Aedes*) are shown in fig. below, and their mode of transmission is discussed briefly.

Diseases caused by mosquitoes



(a) Malaria: caused by protozoan parasites of the genus Plasmodium

- *P. falciparum* - cause most wide spread and dangerous form of malaria
- *P. vivax*
- *P. ovale*
- *P. malariae*

Parasites are transmitted from one person to another person by female *Anopheles* mosquito (males do not transmit the disease as they feed only on plant juices). Parasites develop in the gut of the mosquito and are passed on through the saliva of an infective mosquito, each time it takes a new blood meal. The parasites are carried by the blood stream to the victim's liver, where they invade the cells and multiply. After 9-16 days they return to blood and penetrate the RBC where

they multiply again, progressively breaking down RBC. The infected individual is affected by bouts of fever and anaemia. Anopheline species in selected areas of India are listed below.

Major Vector	Zone of Influence
<i>An. culicifacies</i> (rural vector)	Vector of rural malaria in the north, south & central India. All over India except north-east
<i>An. stephensi</i> (urban vector)	All towns except north-east, rural areas of arid/semi-arid zone except in the north
<i>An. minimus</i> (foot hill vector)	North-east states, north West Bengal
<i>An. fluviatilis</i> (foot hill vector)	Foot hills all along the Himalayan range, seepages in irrigation channel
<i>An. dirus</i> (forested areas)	Deep forests in north-east region
<i>An. sondaicus</i> (coastal areas)	Andaman & Nicobar Islands

Anopheline vectors related to various countries in the world are available in the book entitled "The anophelines of India", by T. Ramachandra Rao, ICMR, New Delhi (1981).

b) Lymphatic filariasis

Infection of the human lymphatic system by filarial nematodes (round worms) is vectored by mosquitoes. Humans serve as the reservoir of this disease. Mosquitoes taking a blood meal ingest micro-filaria (larval stage) which are present in the blood. There are many mosquitoes species which act as vectors for the different species of filarial nematodes e.g.

- *Wuchereria bancrofti*: vectors are - *Culex quinquefasciatus*, *Aedes gambiae*, *Aedes polynesiensis* etc.
- *Brugia malayi*: is vectored by *Anopheles barbirostris*, several species of *Aedes* & *Monsonia*.

Heart worm disease: found in dogs and related canines is caused by a filarial nematode (a large thread like round worm), *Dirofilaria immitis*. The adult worms live in the right side of the heart (right ventricle) and adjacent blood vessels (pulmonary arteries) and because of their location are commonly "dog heart worms".

c) Yellow fever

This viral disease is transmitted to man by a specific mosquito, *Aedes aegypti*. One gets yellow fever on being bitten by infected *Aedes* mosquito which injects yellow fever virus through the bite.

d) Dengue

Dengue is caused by an RNA flavivirus exhibiting many stereotypes. *Aedes aegypti* is the main vector of dengue. It is a container breeder adapted to urban life. As people move away from the proximity of natural water and start to sequester water in containers, this mosquito finds a good breeding ground. *A. aegypti* transmits dengue via bite only. A mosquito feeding on a person, who is in the 1st-5th day of the disease symptoms, can vector the disease to another person. The dengue virus does not affect the mosquito in any way.

(e) Encephalitis

- i) Eastern equine encephalitis (EEE): The EEE virus is maintained in enzootic bird- mosquito cycles centered around hardwood. Passerine birds in and around these swamps serve as reservoirs for the virus. Mosquitoes, especially *Culiseta melanura* feed on these infected birds thereby amplifying the amount of virus. (*Aedes sollicitans* and *Coquillitidia perturbans* - mosquitoes are referred to as bridge vectors due to their ability to bridge the gap between birds and mammals and carry the virus out of bird mosquito cycle and introduce it to mammals.
- ii) St. louis encephalitis (SLE): is a mosquito borne viral infection. The virus is known to occur in natural enzootic bird mosquito cycles. The mosquito species responsible for transmitting the infection is the common house mosquito *Culex pipiens*. Occasionally, these mosquitoes will feed on a variety of mammals, including humans, thereby, transmission of the virus can occur through infected mosquitoes that which had previously fed on infected birds. *Culex pipiens* breeds in stagnant water that collects in a variety of places, including catch basins, dis-repaired pools, buckets and other containers, and road side ditches. The virus is thought to enter new areas through migrating birds, because these mosquitoes do not normally fly very far from their breeding sources.
- iii) West Nile encephalitis (WNF): is transmitted through a mosquito borne flavi virus. It is found in Africa, Europe and Asia. Typically, this virus causes clinical disease in only a small percent of humans infected. A number of mosquito species have been shown to transmit WNF, the most notable belonging to the genus *Culex* and *Aedes*.

1.3 Characteristics of Mosquito species

For prevention and control of mosquitoes, it is important to know their general morphology/ physiology and other general features and variations among different species.

Life cycle

Adult female (only) mosquitoes seek a blood meal and produce a batch of eggs/individual eggs. Some mosquitoes lay eggs on the sides of tree holes or discarded containers, or in depressions in the ground that can hold water. The eggs can lay dormant for several years and hatch when they are flooded by rainfall. Several flooding and drying cycles are usually required to hatch all the eggs laid by a particular mosquito. Other mosquitoes lay eggs directly on the surface of water.

The life cycle from the stages of eggs to larvae to adults is depicted in Fig. 1. The eggs are often attached to one another to form a raft. The individual eggs or rafts float on water. They hatch in 24-28 hours releasing larvae that are commonly called 'wigglers'. Larvae can be often seen wriggling up and down from the surface of water. The larval stage is always aquatic and they shuttle from the sub-surface to obtain oxygen through a snorkel-like breathing apparatus. Generally, the larvae feed on micro-organisms and organic material in water. In about 7-10 days after the eggs hatch, larvae (which go through 4 instars or developmental stages) change to the pupal or 'tumbler' stage in preparation for adult life. The pupal stage does not feed but unlike most insect pupae it is extremely active. The adult emerges from the pupal case using air pressure and assumes a terrestrial existence. Male mosquitoes mate with females one to two days after they emerge. A few days after emerging from water, female mosquitoes begin to seek an animal to feed on. Males do not bite, but feed on plant juices. Mosquitoes can travel a mile or more from their breeding spot to find a meal.

The differences in the different stages of life cycle between various species of mosquitoes is important for DRWH design. All mosquitoes go through the stages of egg-larvae-pupae-adult. The egg and pupae are in water, but once the head develops, the adult goes in the air. However notable differences are seen among various genus. These details and differences are summarised in table 2 and further schematically presented in figures 2 and 3.

Conclusions

From the above review it is clearly seen that all mosquitoes have one key requisite; they need water to complete their life cycle. Since DRWH involves water collection and storage, there is a potential for mosquito breeding. All the available literature pertaining to the aspects of mosquito breeding and control with reference to DRWH are compiled in the subsequent section-II: 'DRWH design and insect breeding'.

Under the project, steps have been taken to examine closely the various measures which are to be taken for the prevention of mosquito breeding and control in DRWH system. These are presented in section-III.

Table 2: Characteristics of major mosquito species

PARAMETERS	<i>ANOPHELES</i>	<i>AEDES</i>	<i>CULEX</i>
Sub-family:	Anophelinae	Culicine	Culicine
Life span	7-10 days (male) 4-5 weeks (female)	Average of 20-30 days Females deposit ~ 4 batches of eggs	-
Breeding site	River margins, river bed pools, canal, seepage from water from canal/dam, rainwater (burrow/pits) Low lying grounds, hoof marks & wheel ruts, rice fields, wells, ponds, brackish water pools	Containers, discarded tins, empty pots, broken bottles, coconut shells, artificial collection (of waters) e.g. coolers, tyres (discarded)	Domestic & peridomestic sources such as cesspools, open ditches, sewage and waste water
EGGS	Laid singly (has lateral floats)	Laid singly, no floats, breed in clean/unpolluted water (man made/artificial containers)	Clusters/rafts of 100-250 eggs, no floats
Viability	1-2 days at normal temperature. May be extended upto 1 week in winters	If dried under natural conditions, are viable for 6 months or longer	~ 2-3 days
LARVAE	(yellowish, green/ grey) remain parallel to surface of water, no siphon (breathing tube)	Remain vertically hanging to water surface, has a siphon (breathing tube)	Remain vertically hanging to water surface, has a siphon
No. of Instar & size			
I Instar	< 1 mm	-	-
II Instar	1-2 mm	-	-
III Instar	4-5 mm	5 - 10 mm	-
IV Instar	5-7.5 mm	~ 10 mm	-
To what water depths it can reach	~ upto 1 m	Few meters	
Emergence	7-10 days normally	7-10 days	-

PARAMETERS	<i>ANOPHELES</i>	<i>AEDES</i>	<i>CULEX</i>
Sub-family:	Anophelinae	Culicine	Culicine
ADULT Size	~ 0.5 cm	0.4-0.5 cm	~ 0.5-0.9 cm
Color	Grey, rest at 45o angle to surface	Black with white patches on the legs, rests parallel to surface	Dark grey, rests parallel to surface
Sitting posture Resting habits	<ul style="list-style-type: none"> • Body parallel to resting surface. • In cattle sheds, under bushes & in tree holes, an indicator of outdoor resting 	-	-
Eating habits i) (male) ii) (female)	Nector/fruit juices Haematophagus	Nector/plant juices Haematophagus	Nector/plant juices Haematophagus
Biting habits/time	Both indoor/outdoor Through out night but peak biting occurs between 19 and 4 hrs. Predominantly zoophilic	Mostly indoor than out doors Diurnal, day biters with 2 peaks of biting (1st at the dawn after the sunrise and 2nd at dusk before sun set)	Both indoor/outdoor From dawn to dusk
Flight range	1-3 km	50 -200 m	1-2 km
ADULT DENSITY (needed to maintain transmission)	1 infective bite	No estimate of adult density is known for transmission possibly one infective bite	Usually very high density is required Needs several infective bites for transmission
Diseases caused	Malaria Filaria	<ul style="list-style-type: none"> • Yellow fever • Dengue • DHF • Filaria • Chikungunya fever 	<ul style="list-style-type: none"> • Ban croftian filarias • Japanese encephalitis • West Nile fever • Viral arthritis • Epidemic/polyarthritis

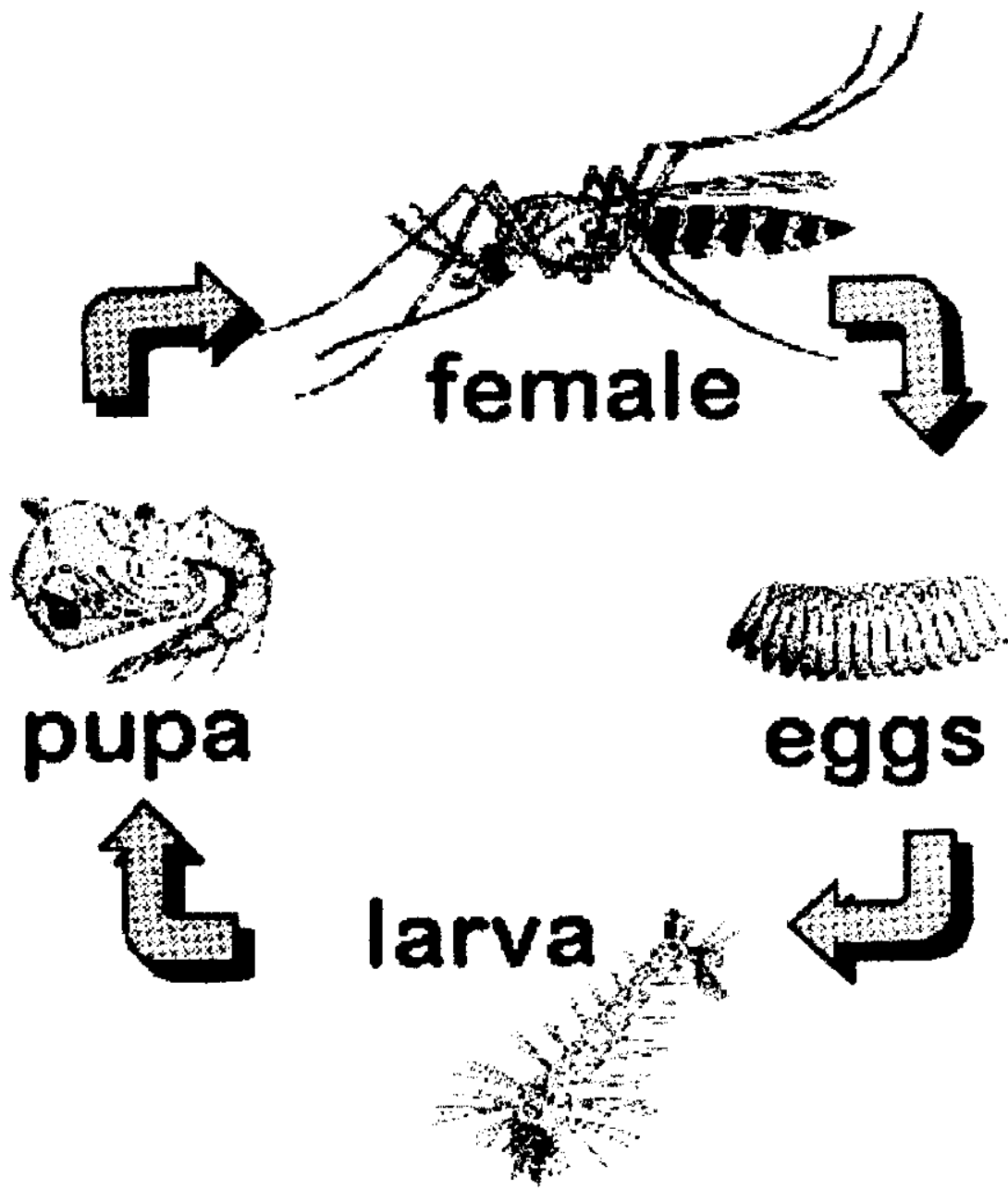



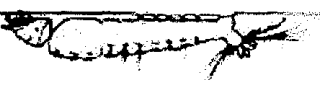
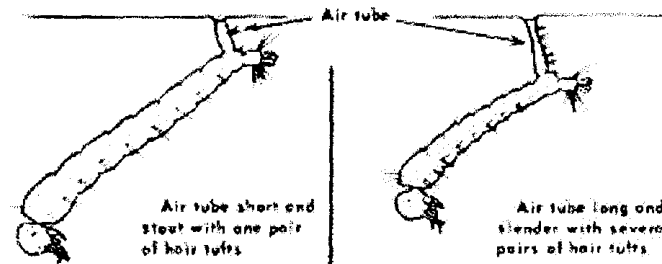

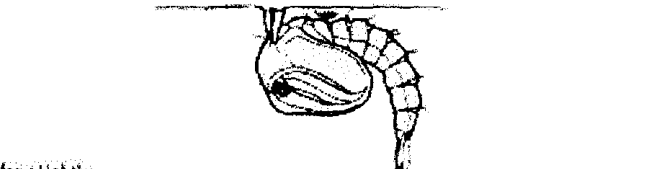
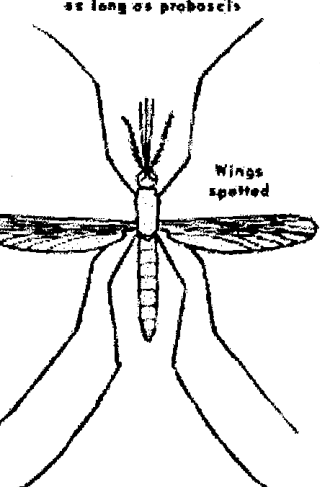
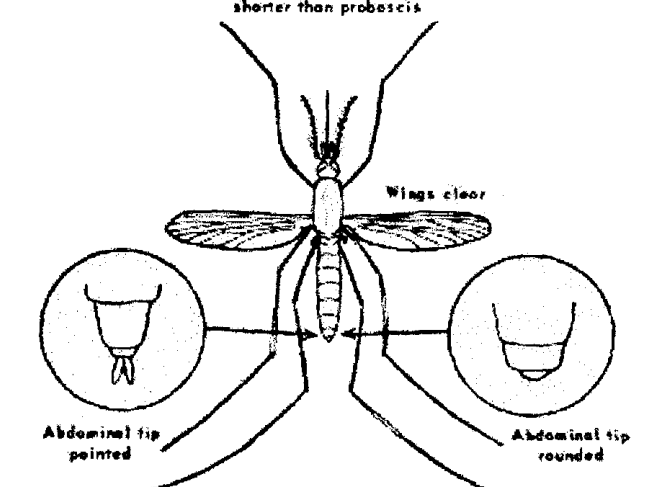
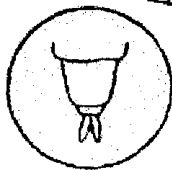


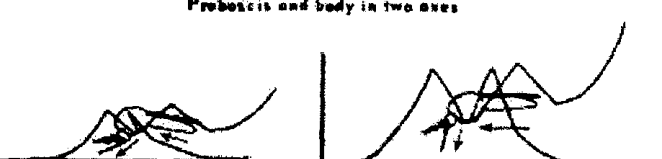


Fig. 1: Life cycle of a mosquito

Fig. 2

Principal Characters for Identifying Mosquitoes of General Importance

ANOPHELES	AEDES	CULEX
EGGS		
 <p style="font-size: small;">Laid singly Has floats</p>	 <p style="font-size: small;">Laid singly No floats</p>	 <p style="font-size: small;">Laid in rafts No floats</p>
LARVAE		
 <p style="font-size: small;">Rest parallel to water surface No air tube Head rotated 180° when feeding</p>	<p style="font-size: small;">Rest at an angle</p>  <p style="font-size: small;">Air tube</p> <div style="display: flex; justify-content: space-around;"> <div style="text-align: center; padding: 5px;"> <p style="font-size: x-small;">Air tube short and stout with one pair of hair tufts</p> </div> <div style="text-align: center; padding: 5px;"> <p style="font-size: x-small;">Air tube long and slender with several pairs of hair tufts</p> </div> </div>	
PUPAE		
	 <p style="font-size: small;">Pupae differ slightly</p>	
ADULTS		
<p style="font-size: small;">Maxillary palps as long as proboscis</p>  <p style="font-size: small;">Wings spotted</p>	<p style="font-size: small;">Maxillary palp shorter than proboscis</p>  <p style="font-size: small;">Wings clear</p> <div style="display: flex; justify-content: space-around; margin-top: 10px;"> <div style="text-align: center; padding: 5px;">  <p style="font-size: x-small;">Abdominal tip pointed</p> </div> <div style="text-align: center; padding: 5px;">  <p style="font-size: x-small;">Abdominal tip rounded</p> </div> </div>	
<p style="font-size: small;">Proboscis and body in one axis</p> 	<p style="font-size: small;">Proboscis and body in two axes</p> 	

Section -II: DRWH and Mosquito Breeding

It is known that any water source is a potential ground for breeding of mosquito and hence adequate precaution for mosquito control must be taken in designing DRWH. Available reports on this aspect are chronologically presented below.

- * One of the first references available is from engineering construction, malaria control bulletin. Though not in the context of DRWH, the authors describe a mosquito proof domestic cistern, with protection of the opening from entry of mosquitoes. [(Health Bulletin No. 32 (Malaria Incidental to Engineering Construction) (see Appendix-1)].
- * Waller (1989) in a study on RWCS in Nova Scotia, Canada, and Bermuda, asserts, that if the cistern surface is not covered, open storage may provide a breeding place for mosquitoes, which may act as vectors for diseases including haemorrhagic fever. Exposure to sunlight may promote algal growth. He recommends disinfection of cistern water using UV light including occasional addition of chlorine. Filtration devices have been used in some systems, but these should be properly constructed, designed and maintained.
- * In a discussion on how mosquito breeding can be prevented, Noriyuki Fukano (1994) has suggested installation of insect -proof mosquito nets or other insect proof devices in the vent routes (drain pipes, overflow pipes etc.). If tank water is continuously consumed and replenished, no batch of water would stay in storage for a long time. Therefore, even if mosquito enters into the tank and spawns eggs, there is virtually no likelihood of mosquito proliferation. However this is not always the case in rain water storage. Also mosquitoes breed easily in rain water drainage pits. As rain water from roofs and the ground surface drain into sewers many larvae accumulate. To change conventional rain water drainage pits to ones made of permeable matter is a good solution to avoid mosquito problems, even if some cost is involved.
- * Commenting on prevention of mosquito breeding, Appan (1994) recommended that a mosquito proof device has to be installed between the filter and the rain water storage tank. He presented data on the use of simple and inexpensive roof water collection systems in some South East Asian countries. Collected samples in most locations were positive for Total and Faecal Coliform. It is proposed that collected roof water be boiled, disinfected with household bleach or subjected to radiations from sunligh. In a certain RWH systems in Indonesia ,fishes are being reared to prevent mosquito breeding in tanks .

- * According to Daulat et al. (1997) sometimes the water in the container becomes contaminated by toads, frogs, mosquitoes, cockroaches and rats. He suggested that the lid of the containers should be kept closed at all times or polyethylene sheet may be used to close the mouth of the container to prevent breeding of mosquito and other insects. Periodic cleaning is needed.
- * Bambrah & Haq (1997) pointed out that a tight cover ensures dark storage conditions preventing growth of algae and breeding of mosquito. Open containers or storage ponds are generally unsuitable for storing drinking water.
- * Hartung in his study at Rwanda (1999) suggests that the tank should be tightly closed to prevent mosquito breeding in RWH tanks. He is of the opinion that water in the tanks should not be exposed to direct sunlight in order to avoid algal growth.
- * Gould's (1999) in the chapter on 'Rainwater Quality Issues' has highlighted 'Mosquito breeding and control. The relevant details are presented in Appendix 2.
- * Malaria Research Centre is an apex body in India working on the prevention and control of mosquito and malaria. Details on how mosquito can be prevented by physical, chemical and biological methods are summarized in table 2. Some of these methods may prove useful in designing DRWH.

Table 2 General measures for Mosquito Control *

CONTROL MEASURES	ANOPHELES	AEDES	CULEX
Physical	Adults: House screening and mosquito nets (1.5 mm size or 25/26 mesh holes). Recommended doses for impregnation are: Permethrin: 0.2-0.5 gm/sq. m Deltamethrin: 0.025 gm/sq. m Cyfluthrin: 0.05 gm/sq. m Larvae: oiling and environmental management	Environmental management Container management • Elimination of breeding sites • Preventive measures for breeding in containers	Environmental sanitation Mosquito proofing
Chemical	Larvicides: abate (temophos) & chloropyriphos Adulticides: pyrethrins, Spraying and residual insecticides, repellants are also used for personal protection	Chemical larviciding: 1% temephos (Abate) granules @ 0.1 g/l Focal sprays (Pyrethrins-0.1 %) Insecticide treated curtains/nets Thermal fogging ULV	Chemical larviciding: Temephos, Fenthos, Chlorophyriphos etc. ULV thermal fogging
Biological	• Larvivorous fishes (<i>Gambusia affinis</i> , <i>Lebister reticulatus</i>) • Invertebrate predators Microbial insecticides: Bti (<i>Bacillus thuringiensis</i>) & B.S (<i>Bacillus sphareicus</i>)	• Larvivorous fishes • Bti (<i>Bacillus thuringiensis</i>) • Invertebrate predators such as mesocyclops	• Larvivorous fishes (<i>Poecilia reticulata</i>) and <i>Oreochromis mossambicus</i> (Peters) control breeding in cow dung pits. • Bti (<i>Bacillus thuringiensis</i>) & B.S (<i>Bacillus sphareicus</i>)

* These are general control measures. Not all of these could be considered safe for DRWH

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Mosquito breeding and control

The three main groups of mosquitoes and their associated diseases are *Aedes* associated with dengue and yellow fever, *Anopheles* with malaria and *Culex* with filariasis and extreme nuisance. Larvae from members of all three groups may be found in tanks containing stored rainwater, especially in tropical regions. It is the *Aedes* group, and particularly *Aedes aegypti*, the vector responsible for transmission of dengue and yellow fever, which is most commonly found in water storage containers both large and small in and around the house. While evidence suggests that covering containers can significantly reduce the prevalence of mosquito larvae in the water, it is difficult to prevent the problem completely.

In a survey of 150 households in three villages in Khon Kaen Province, Thailand, *Aedes* mosquito larvae were present at all households. They were found in 95 per cent of small indoor clay storage containers, in 32 per cent of all rainwater jars and 4 per cent of rainwater tanks (Chareonsook et al., 1985). Hewison and Tunyavanich (1990) reported that villagers complained of increased mosquito infestation in north-east Thailand following the widespread introduction of ferrocement water jars. While this may have been due to the increased availability and presence of water in the village, it also seemed to be much worse where containers were not covered. To reduce the problem significantly it was recommended that all containers, large and small, and both inside and outside the house, must be covered and screened with some form of mesh. In Queensland, Australia, a survey of 1349 premises revealed that rainwater tanks provided important breeding sites for immature *Aedes aegypti* mosquito larvae (Tun-Lin et al., 1995). A study conducted in two villages in southeast Nigeria, where all households use rainwater stored in earthen ware pots around the house, revealed the widespread presence of *Aedes aegypti* mosquito larvae. In this case, the vector was associated with the transmission of yellow fever, and between 53 and 76 containers per 100 households were found to be infested during the wet season (Bang et al., 1981).

Various approaches to mosquito control can be used. The addition of small amounts of domestic kerosene (5ml per 1000 litres) works well, but can give the water an unpleasant taste and may not be suitable for tanks lined with plastic. [Note: Commercial or industrial kerosene should *not* be used]. Various forms of biological control, such as keeping fish and dragonfly larvae in the tanks to consume mosquito larvae, have also been tried with some success (Skinner, 1990; Corbet, 1986). While biological control may be a useful and effective tool at specific locations over limited time periods, the best guarantee against preventing mosquitoes from laying eggs in rainwater jars, tanks or other water storage containers is to make sure they are inaccessible. To exclude mosquitoes, containers must be tightly covered and any openings properly screened with fine nylon or metallic mesh. Nevertheless, regular inspection is also essential to alert users to potential problems and should prompt immediate action when necessary. Leaving rainwater tanks, jars and in-house water storage containers unscreened or uncovered in areas where malaria, dengue or yellow fever are endemic is to court danger.

Section III: Designing of Mosquito Control in DRWH

Based on the literature review, it is seen that mosquito control measures have to be applied at all stages of the mosquito life cycle, with suitable ovicidal, larvicidal and adulticidal interventions. (see Figure 4). The preventive measures with regard to DRWH may be divided into three groups:

i) Prevention of mosquito breeding in the surroundings.

- Appropriate physical, chemical and biological means of control can be used.
- Plants which repel mosquitoes can be grown around the DRWH site.
- If there are depressions in the surrounding B.S., BTI, larvivorous fishes, aquatic plants, plant extracts (oil layer), kerosene oil etc. can be used. Some of the well tested chemicals may also be used.

ii) Prevention of mosquito breeding in the DRWH system.

- Avoid all the factors which result in attracting mosquitoes.
- Tightly closed lids may be provided to the water storage system, so that there are no openings for the entry of mosquitoes.
- Screen may be used (with hole size less than 1 mm) to bar entry of mosquito larvae.
- Filter should be disinfected (eg. with household bleach).
- No stagnating water should be allowed around the DRWH site.
- Gutter leading to the storage should have a free flow of water.

iii) If inspite of the above, eggs or larvae have entered DRWH, various ovicidal and larvicidal measures have to be considered.

- The measures practiced for killing bacteria may result in dying of mosquito eggs and larvae. These include high temperature, boiling and use of botanicals.
- Algal bloom both promote or discourage mosquito larvae depending on certain conditions.
- Measures for protection from mosquito biting should be undertaken.

Suggestions for further work

Some of the above ideas on mosquito control are being evaluated on an experimental scale under the project. Following environmental friendly mosquito control measures for preventing the entry of eggs, larvae and adult mosquitoes are being integrated in DRWH design:

- Prevention of breeding in the surroundings using certain aquatic and terrestrial plants which deter mosquitoes. In this context use of certain plants and extracts used traditionally in mosquito control would be examined.
- Screening of the entry points by mesh / nylon of appropriate mesh size (size of holes < 1mm).
- Filter and filter treated with bleaching powder, at the entry point for water storage.
- Use of food grade plastic sheet/beads and botanical extracts for topping the water and prevent development of mosquito larvae.