

## Review Paper

# Aquatic insect predators and mosquito control

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**Abstract.** Mosquitoes are serious biting pests and obligate vectors of many vertebrate pathogens. Their immature larval and pupal life stages are a common feature in most tropical and many temperate water bodies and often form a significant proportion of the biomass. Control strategies rely primarily on the use of larvicides and environmental modification to reduce recruitment and adulticides during periods of disease transmission. Larvicides are usually chemical but can involve biological toxins, agents or organisms. The use of insect predators in mosquito control has been exploited in a limited fashion and there is much room for further investigation and implementation. Insects that are recognized as having predatorial capacity with regard to mosquito prey have been identified in the Orders Odonata, Coleoptera, Diptera (primarily aquatic predators), and Hemiptera (primarily surface predators). Although their capacity is affected by certain biological and physical factors, they could play a major role in mosquito control. Furthermore, better understanding for the mosquitoes-predators relationship(s) could probably lead to satisfactory reduction of mosquito-borne diseases by utilizing either these predators in control programs, for instance biological and/or integrated control, or their kairomones as mosquitoes' oviposition repellents. This review covers the predation of different insect species on mosquito larvae, predator-prey-habitat relationships, co-habitation developmental issues, survival and abundance, oviposition avoidance, predatorial capacity and integrated vector control.

## INTRODUCTION

Mosquitoes are important insects not only as nuisance biters but also as vectors of important diseases such as malaria, filaria and dengue particularly in the tropics. The World Health Organization adopted mosquito control as the only method to prevent or control such diseases. Although interest in mosquitos' biological control agents was large at the beginning of the 20<sup>th</sup> century, it is stopped since the discovery of insecticidal properties of the DDT in 1939. Since that time insecticides were extensively used for mosquito control. Due to their deleterious health and environmental impacts, search for environmentally friendly insecticide alternatives has become

increasingly necessary. For this aspect, renewed interest in biological control agents particularly aquatic predaceous insects that inhibit mosquitoes' breeding sites could provide acceptable reductions in mosquito population and it could be included in integrated vector management (IVM) program.

Mosquito's life cycle includes for stages: egg, larva, pupa and adult. The first three stages are aquatic giving high opportunity for the success of predaceous insects for mosquito control. Although information about contribution of aquatic predaceous insects in mosquito eggs predation is very rare, a few references exist on aquatic insects preying upon adult mosquitoes. The predaceous bug *Emesopsi streiti*

(Reduviidae) preying upon adult mosquitoes in bamboo internodes (Kovac & Yang, 1996). Yanovisk (2001) also mentioned that *Microvelia cavicola* and *Paravelia myersi* (Veliidae) fed on adult mosquitoes emerging in tree holes. Fly larvae of *Xenoplatyura beaveri* preying upon emerging adult mosquitoes in *Nepenthes* pitcher plants (Mogi & Chan, 1996). The Dragonflies *Pantala hymenaea* and *Erythemis collocata* attack swarming of *Anopheles freeborni* after sunset (Yuval & Bouskila, 1993). Because of predation of aquatic insects on mosquito larvae and pupae is more observed and significantly affecting mosquitoes emerging, therefore the present review includes only predation against larval stage.

Such predaceous insects are not only preying on nuisance mosquitoes but also preying on mosquito vectors of diseases such as *Anopheles gambiae* (malaria vector), *Aedes aegypti* (dengue vector) and *Culex annulirostris* (encephalitis vector). While Yasuoka & Levins (2007) suggest that conserving aquatic insects associated with mosquito larvae could be effective in controlling mosquito vectors in the study site. Walker & Lynch (2007) stated that targeting malaria vector larvae, particularly in human-made habitats, can significantly reduce malaria transmission.

Although predaceous aquatic insects inhabit a wide variety of aquatic habitats, which would seem to support their usefulness, the selection of biological control agents relies on more important factors. Selection should be generally based on the capacity of a predator to maintain very close interaction with its prey population, capacity to self-replicate/reproduce, climatic compatibility, and potential for unintended and possibly adverse impacts (Waage & Greathead, 1988). Research has confirmed that natural enemies are frequently responsible for significant reductions in mosquito populations and should be indispensable to integrated control which seeks to maintain mosquito vector populations below annoyance and/or disease transmission level (Legner, 1994). Furthermore, introducing and/or augmenting such natural enemies has in some cases

provided satisfactory control (Sebastian *et al.*, 1990; Chandra *et al.*, 2008; Mandal *et al.*, 2008) and sustained release of them over several years may reduce the relative high cost of massive releases.

Since there are some other biological control agents such as bacterium, one of the advantages of the predaceous insects over the other biological control agents is, these insects could reach mosquitoes in some habitats such as tree holes and phytotelmata, water bodies held by plants, in tropics and subtropics that are very difficult to be controlled with other biological control measures.

Some articles have discussed and summarized both aquatic insects and other invertebrates that prey upon mosquitoes. Biology, colonization and potential of *Toxorhynchites* mosquitoes as a biological control agent of vector mosquitoes are fully covered by Collins & Blackwell (2000) while Garcia (1982) discussed the difficulties associated with such methodologies which prevent more widespread utilization of arthropod predators. In addition to *Toxorhynchites* mosquitoes, the predaceous characters of *Culex* (Subgenus *Lutzia*) mosquitoes were reviewed by Pal & Ramalingam (1981). Moreover, Bay (1974) reviewed many aquatic insects that prey upon mosquito larvae and categorized them according to their taxonomic orders. Lacey & Orr (1994) limited their discussion to insect predators that are used as biological control agents in integrated vector control to *Notonecta* and *Toxorhynchites* species whilst Kumar & Hwang (2006) reviewed larvicidal efficiency of amphibian tadpoles, larvivorous fish, cyclopoid copepods in addition to aquatic insects for mosquito biocontrol. Mogi (2007) reviewed insects and invertebrate predators based on adult, egg, larval and pupal mosquito predation beside possibilities of using such predators for mosquito control. Quiroz-Martinez *et al.* (2007) discussed the arthropods (insects, mites and spiders) that prey on mosquito larvae and considerations for the success of these predators in mosquitoes' biological control programs.

The present article not only reviews the predation of different insect groups on mosquito, particularly larvae, but also includes predator-prey associations in different aquatic habitats, the degree to which predators affect mosquito development, survival, abundance and fitness, oviposition avoidance of mosquitoes in response to the presence of aquatic insect predators, factors influencing predatorial capacity, predaceous insects used in integrated vector control and finally difficulties for utilizing predaceous insects for mosquito control. Although the predation of different insects on mosquito larvae and the predators-mosquitoes association in different habitats may be little bit similar to the previously mentioned reviews, the other parts are completely different and presenting new information for utilizing predaceous insects in mosquito biocontrol.

#### **Predaceous insects**

Many aquatic insects in the orders Coleoptera, Diptera, Hemiptera and Odonata are known to prey upon mosquito larvae (principal genera and species of interest are shown in Table 1). Predators may be polyphagous, feeding on a broad range of prey species (generalist predator), oligophagous, with a restricted range of prey; or monophagous, with a very limited range of prey (specialist predators). Most predators of mosquitoes tend to be from the generalist type (Collins & Washino, 1985). Some predators (especially those with chewing mouthparts) eat their prey (Odonata) but others suck the body fluid (hemolymph) of the prey (many beetle larvae and Hemiptera). Although predation may occur during any life stage, most research focused on mosquito larval and pupal stages because egg predation appears to be a minor component of mosquito mortality and predation on the adult stage seems unlikely to provide reliable levels of control in most cases (Collins & Washino, 1985). Also surplus or 'wasteful' killing of uneaten prey is characteristic to the fourth larval instar of various species of the predatory mosquito genus *Toxorhynchites*, it is recently documented in the fourth larval instar of the

predatory midge genus *Corethrella* (Lounibos *et al.*, 2008). Furthermore, surplus or killing activity of *Toxorhynchites* larvae to mosquito pupae is fortunate in the context of control, because pupal production is most highly correlated with subsequent adult densities (Padgett & Focks, 1981) and probability of disease transmission.

According to hunting strategies, predators are classified into neuston that float on the top of the water (Vellidae: Hemiptera), free swimming (some microcrustaceans), climbing stalkers (Zygoptera: Odonata), sprawling ambushers (Anisoptera: Odonata), and cursorial searchers (Dytiscidae and Hydrophilidae: Coleoptera). Predaceous insects are also categorized into surface predators and aquatic predators. The first group comprised insects that forage near or below the water surface to catch their prey and all belong to Order Hemiptera. Predators in the latter group are good swimmers and are able to forage beneath water or/and on subsurface terrain beneath vegetation such as Orders Odonata and Coleoptera and some hemipterans. The following sections present information on the major predator groups and their capacity for mosquito control.

#### **Coleopteran predators**

Although aquatic coleopterans are commonly associated with mosquito larvae in different habitats, they have been less explored compared to other insect predators (Chandra *et al.*, 2008). Among coleopterans, families Dytiscidae and Hydrophilidae have received attention as mosquito larvae predators. Adults and larvae of Dytiscidae and Hydrophilidae are common predators in ground pools, permanent and temporary ponds, and artificial mosquito breeding sites and were reported from phytotelmata as well. Although they can reduce mosquitoes densities in some pools (Nilsson & Soderstrom, 1988; Nilsson & Svensson, 1994; Lundkvist *et al.*, 2003), their mosquito control efficacy perhaps is limited by incomplete habitat overlap, alternative prey preference, emigration and cannibalism (Juliano & Lawton, 1990; Lundkvist *et al.*, 2003). Likely, species of the genera *Laccophilus*, *Agabus*

Table 1. Most common and principal genera and species of predaceous insects

Order	Genera and species	Mosquito prey	References	
Coleoptera	<i>Acilius sulcatus</i>	<i>Cx. quinquefasciatus</i>	Chandra <i>et al.</i> , 2008	
	<i>Agabus erichsoni</i>	<i>Ae. communis</i>	Nilsson & Soderstrom, 1988	
	<i>Agabus opacus</i>			
	<i>Colymbetes paykulli</i> , <i>Ilybius ater</i>	<i>Culex</i> mosquitoes	Lundkvist <i>et al.</i> , 2003	
	<i>Ilybius fuliginosus</i>			
	<i>Dytiscus marginicolis</i>	<i>Culiseta incidens</i>	Lee, 1967	
	<i>Lestes congener</i>	<i>Culiseta incidens</i>	Lee, 1967	
	<i>Lacconectus punctipennis</i> <i>Rhantus sikkimensis</i>	<i>Ae. albopictus</i> <i>Cx. quinquefasciatus</i>	Sulaiman & Jeffery, 1986 Aditya <i>et al.</i> , 2006	
Diptera	<i>Anopheles barberi</i>	Tree-hole mosquito larvae	Peterson <i>et al.</i> , 1969	
	<i>Anopheles gambiae</i>	Larvae of same species	Koenraadt & Takken, 2003	
	<i>Bezzia expolita</i>	Mosquito larvae	Hribar & Mullen, 1991	
	<i>Chaoborus crystallinus</i>	Mosquito larvae	Bay, 1974	
	<i>Chaoborus cooki</i>	Mosquito larvae	Borkent, 1980	
	<i>Corethrella appendiculata</i>	<i>Ae. albopictus</i> & <i>Ochlerotatus triseriatus</i>	Kesavaraju & Juliano, 2004 & Griswold & Lounibos, 2006	
	<i>Corethrella brakeleyi</i>	<i>An. quadrimaculatus</i>	McLaughlin, 1990	
	<i>Cx. allostigma</i>	Tree-hole mosquito larvae	Yanovisk, 2001	
	<i>Cx. fuscus</i>	<i>Ae. aegypti</i> , <i>An. stephensi</i> & <i>Cx. quinquefasciatus</i>	Bai <i>et al.</i> , 1982 & Kuldip <i>et al.</i> , 1984	
	<i>Cx. raptor</i>	<i>Cx. quinquefasciatus</i> <i>Cx. fatigans</i>	Ikeshoji, 1966 Prakash & Ponniah, 1978	
	<i>Culicoides cavaticus</i>	<i>Cx. quinquefasciatus</i>	Thangam & Kathiresan, 1996	
	<i>Culicoides guttipennis</i>	<i>Ae. sierrensis</i>	Clark & Fukuda, 1967	
	<i>Culiseta longiareolata</i>	<i>Ae. aegypti</i> <i>Cx. quinquefasciatus</i>	Bay, 1974 Kirkpatrick, 1925 & Al-Saadi & Mohsen, 1988	
	<i>Dolichopus gratus</i>	Mosquito larvae	Laing & Welch, 1963	
	<i>Mochlonyx culiciformis</i>	<i>Ae. communis</i>	Bay, 1974	
	<i>Monohlea maya</i>	Mosquito larvae	Fellipe-Bauer <i>et al.</i> , 2000	
	<i>Ochthera chalybesceens</i>	<i>An. Gambiae</i> s.s.	Minakawa <i>et al.</i> , 2007	
	<i>Tx. amboinesis</i>	<i>Ae. aegypti</i>	Focks <i>et al.</i> , 1985	
	<i>Tx. brevipalpis</i>	<i>Ae. aegypti</i>	Gerberg & Visser, 1978	
	<i>Tx. brevipalpis conradti</i>	<i>Ae. africanus</i>	Sempala, 1983	
	<i>Tx. kaimosi</i>	<i>Ae. africanus</i>	Sempala, 1983	
	<i>Tx. rutilus rutilus</i>	<i>Ae. aegypti</i> <i>Ae. aegypti</i> & <i>Cx. quinquefasciatus</i>	Padgett & Focks, 1981 Focks <i>et al.</i> , 1982	
	<i>Tx. splendens</i>	<i>Ae. albopictus</i> & <i>Ochlerotatus triseriatus</i> <i>Cx. quinquefasciatus</i> <i>Ae. aegypti</i>	Griswold & Lounibos, 2006 Aditya <i>et al.</i> , 2006 Amalraj & Das, 1998, Wattal <i>et al.</i> , 1996 & Wongsiri & Andre, 1984 Yasuda & Hagimori, 1997	
	<i>Tx. towadensis</i>	mosquito larvae		
	Hemiptera	<i>Abedus indentatus</i>	mosquito larvae	Washino, 1969
		<i>Anisops</i> sp.	<i>Cx. annulirostris</i>	Shalan, 2005 & Shalan <i>et al.</i> , 2007
		<i>Belostoma flumineum</i>	Mosquito larvae	Washino, 1969
		<i>Buenoa scimitar</i>	<i>Cx. quinquefasciatus</i>	Rodriguez-Castro <i>et al.</i> , 2006
		<i>Corisella</i> sp.	Mosquito larvae	Washino, 1969
		<i>Diplonychus indicus</i>	<i>Ae. aegypti</i> & <i>Cx. fatigans</i>	Venkatesan & Sivaraman, 1984
		<i>Diplonychus</i> sp.	<i>Cx. annulirostris</i>	Shalan, 2005 & Shalan <i>et al.</i> , 2007
<i>Enithares indica</i>		<i>An. stephensi</i> , <i>An. stephensi</i> & <i>Cx. quinquefasciatus</i>	Wattal <i>et al.</i> , 1996	

	<i>Laccotrephes</i> sp. <i>Notonecta glauca</i> <i>Notonecta hoffmani</i>	<i>Ae. vittatus</i> <i>Cx. pipiens</i> mosquito larvae	Service, 1965 Beketov & Liess, 2007 Scott & Murdoch, 1983 & Murdoch <i>et al.</i> , 1984
	<i>Notonecta shootrii</i> <i>Notonecta undulate</i> <i>Notonecta unifasciata</i> <i>Siagra hoggarica</i> <i>Sphaerodema annulatum</i> <i>Sphaerodema rusticum</i>	<i>Culiseta incidens</i> Mosquito larvae <i>Culex</i> larvae <i>Cx. quinquefasciatus</i> <i>Armigeres subalbatus</i>	Lee, 1967 Ellis & Borden, 1970 Bay, 1967 Alahmed <i>et al.</i> , 2009 Aditya <i>et al.</i> , 2005
Odonata	<i>Aeshna flavifrons</i> , <i>Coenagrion kashmirum</i> , <i>Ischnura forcipata</i> , <i>Rhinocypha ignipennis</i> and <i>Sympetrum durum</i> <i>Brachytron pratense</i> <i>Crocothemis servilia</i> <i>Enallagma civile</i> <i>Libellula</i> sp.  <i>Orthemis ferruginea</i> <i>Tramea lacerate</i> & <i>Tramea torosa</i> <i>Trithemis annulata scortecii</i>	<i>Cx. quinquefasciatus</i>  <i>An. subpictus</i> <i>Ae. aegypti</i> <i>Cx. tarsalis</i> <i>Ae. aegypti</i> (larvae and pupae) Mosquito larvae <i>Culiseta incidens</i>  <i>An. pharoensis</i>	Mandal <i>et al.</i> , 2008  Chatterjee <i>et al.</i> , 2007 Sebastian <i>et al.</i> , 1990 Miura & Takahashi, 1988 Bay, 1974 & Sebastian <i>et al.</i> , 1980 Cordoba & Lee, 1995 Lee, 1967  EL Rayah, 1975

and *Rhantus* have been also reported as potential agents of biological control of mosquitoes (Lee, 1967; Nilsson & Soderstrom 1988; Aditya *et al.*, 2006). A recent field study (Chandra *et al.*, 2008) showed that *Acilius sulcatus* (Family: Dytiscidae) larvae have significant impact on mosquito larvae (*Culex quinquefasciatus*, *Culex bitaeniorhynchus*, *Culex tritaeniorhynchus*, *Culex vishnui*, *Culex gelidus*, *Anopheles subpictus*, *Anopheles vagus*, *Anopheles aconitus*, *Anopheles barbitrostris*, *Anopheles annularis* and *Armigeres subalbatus*) that prevail in cement tanks in Sainthia in the district of Birbhum, West Bengal, India. A significant decrease in larval density of different mosquito species after 30 days from the introduction of *A. sulcatus* larvae was noted, while with the withdrawal, a significant increase in larval density was noted indicating the efficacy of *A. sulcatus* in regulating mosquito immatures. In the control tanks, mean larval density did not differ throughout the study period.

#### Dipteran predators

The most common and famous dipteran mosquito predator is *Toxorhynchites* mosquito that has been introduced as a

biological control agent of container-breeding mosquitoes in many different ecological habitats. A preliminary field trial on the Caribbean island of St. Maarten demonstrated the feasibility of using the predaceous mosquito larva, *Toxorhynchites brevipalpis* to control *Ae. aegypti* larvae (Gerberg & Visser, 1978). Sixteen days after the introduction of *Tx. brevipalpis* eggs into *Ae. aegypti* breeding containers, the 21 houses sampled were negative for *Ae. aegypti* and *Cx. quinquefasciatus* and the house index (the percentage of examined houses that are positive for *Ae. aegypti* larvae) dropped to zero. Focks *et al.* (1982) used *Toxorhynchites rutilus rutilus* to control *Ae. aegypti* and *Cx. quinquefasciatus* mosquitoes in residential blocks within a substandard urban area of New Orleans, Louisiana. Mosquitoes emergence from automobile tires, buckets and paint cans, treated with 1 or 2 first instar larvae of *Tx. r. rutilus* decreased by 65 and 72% respectively, while overall control for both treatment levels was 74%. Weekly releases of another *Toxorhynchites* larval predator, *Toxorhynchites amboinensis*, into a 16-block neighborhood with substandard housing in New Orleans, Louisiana, during 1982 reduced *Ae. aegypti* densities by 45%

when compared with similar but untreated areas (Focks *et al.*, 1985). Increasing the number of adults released per week from 100 to 300 females per block did not improve the degree of control achieved, indicated that lower release numbers may be adequate to achieve this level of control, whilst releasing 100 female predators per block resulted in a 40% reduction in *Cx. quinquefasciatus*. Collins & Blackwell (2000) reported that other attempts to control vector mosquitoes using *Toxorhynchites* spp. mosquitoes have been made in many regions of the world including the Caribbeans, Asia and Africa. In one example, the larval density of *Ae. aegypti* were reduced by more than 90% after the release of *Toxorhynchites splendens* in water tanks in suburban Bangkok, Thailand (Wongsiri & Andre, 1984). These results suggest that it may be possible to develop a practical method to control *Ae. aegypti* mosquitoes in urban areas using *Tx. amboinensis*.

The application of *Toxorhynchites* mosquitoes to control *Ae. aegypti* larvae in developing countries has two additional benefits. Firstly, they have an unusual life cycle in that they are not capable of blood feeding and therefore are not pests or vectors. Secondly, these mosquitoes could be reared locally instead of importing insecticides. Although the aforementioned data are examples of successful mosquito suppression with *Toxorhynchites* mosquitoes, Annis *et al.* (1989 & 1990) reported that this predator is unsuccessful in field application in Indonesia. Repeated release of *Toxorhynchites* first instar larvae in waterlogged places had no effect on mosquito population in Indonesia due to their inability to withstand periods of starvation and to their accidental removal from containers during the act of water consumption. The same maybe true for other aedines since a study conducted in Zika forest, Uganda, on the breeding interactions between *Aedes africanus* and two mosquito predators, *Toxorhynchites brevipapis conradti* and *Toxorhynchites kaimosi*, revealed a significant reduction in the numbers of *Ae. africanus* larvae and pupae

in the tree holes that were also inhabited by predator larvae (Sempala, 1983).

Likewise, other mosquito larvae, particularly *Culex* (Ikeshoji, 1966; Panicker *et al.*, 1982; Thangam & Kathiresan, 1996; Mariappan *et al.*, 1997; Yanovisk, 2001), *Culiseta* (Kirkpatrick, 1925; Al-Saadi & Mohsen, 1988), certain *Anopheles* larvae (Peterson *et al.*, 1969), *Aedes* (Ramalingam & Ramakrishnan, 1971; Mogi & Chan, 1996), the *Ochlerotatus* subgenus *Mucidus* (Mattingly, 1961), the *Psorophora* subgenus *Psorophora* (Carpenter & LaCasse, 1955; Campos *et al.*, 2004) and *Topomyia* (Ramalingam, 1983; Miyagi & Toma, 1989) are known to prey upon mosquito larvae. Ikeshoji (1966) used larvae of *Cx. fuscans* to control *Cx. quinquefasciatus* larvae in small ditches in simulated field conditions. When daily 63 egg rafts of *Cx. quinquefasciatus* were released into the ditches for a period of 3 weeks and 25 first instar larvae of *Cx. fuscans* were introduced daily starting from the fifth day, an average of 156 larvae of *Cx. quinquefasciatus* per day survived to pupate (indicating about 99.98 reduction in pupation). Furthermore, when 2000 larvae of *Cx. quinquefasciatus* were introduced at one end of a ditch 20 cm wide and 100 larvae of *Cx. fuscans* were introduced at the other end, most of the predaceous larvae had swum about 6 m to reach the prey population within 3½ h of their release indicating how much this predator could find its prey. Under laboratory conditions, *An. barberi* larvae were shown to prey upon early instars of various tree hole mosquito larvae. More interestingly, recent study has shown that predation occur within and between larvae of members of the malaria vector *An. gambiae* complex and may affect their adult population densities (Koenraad & Takken, 2003).

Other Dipteran insects particularly ceratopogonid (Hribar & Mullen, 1991; Fellipe-Bauer *et al.*, 2000), chaoborid (McLaughlin, 1990), chironomid (Naeem, 1988), corethrellid (Kesavaraju & Juliano, 2004; Griswold & Lounibos, 2006), culicoid (Clark & Fukuda, 1967; Bay, 1974),

dolichopodid (Laing & Welch, 1963), tipulid (Yanoviak, 2001) and other brachyceran (Kitching, 1990) larvae were recorded as mosquito larvae predators (Table 1).

### **Hemipteran predators**

Belostomatidae, Nepidae and Notonectidae are the most important families of predaceous Hemipteran bugs. The backswimmers (Family: Notonectidae) are the most common bugs preying upon mosquito larvae, important factor in reducing immature mosquito population and considered promising in mosquito control.

The role of hemipteran predators in controlling mosquito larvae has been recognized since 1939 in New Zealand, when stock troughs with *Anisops assimilis* were found to be free of mosquitoes whereas puddles in depressions surrounding the troughs contained mosquitoes (Kumar & Hwang, 2006). Bay (1967) found that almost 100% of mosquito emergence was prevented in field-situated, screened, 100 gallon fibreglass tubs with one square meter of water surface and *Notonecta unifasciata* compared to more than 12000 adult mosquitoes emerged from the control tubs. In another field experiments, in stock tanks (troughs holding drinking water for cattle and horses) in Santa Barbara County, California, *Notonecta hoffmani* were also shown to strongly influence mosquito larvae populations (Murdoch *et al.*, 1984).

The striking effects of those predaceous bugs, *Notonecta* and *Anisops*, are probably due to the physical simplicity of these troughs, tanks and tubes, and particularly to the lack of prey refuges. For instance, emergent vegetation in ponds and other water bodies provide partial protection for mosquito immatures. This effect was experimentally investigated and confirmed by Shaalan (2005) and Shaalan *et al.* (2007) whereas predation potential of *Anisops* and *Diplonychus* bugs was significantly reduced by the presence of vegetation.

Although the costs of colonization and mass production, coupled with the logistics of distribution, handling and timing of release at the appropriate breeding site, impede the use of notonectids in mosquito

control (Legner, 1994), results of a recent study for mass rearing and egg release of the predatory backswimmer *Buenoa scimitar* for the biological control of *Cx. quinquefasciatus* were impressive (Rodriguez-Castro *et al.*, 2006). Production of backswimmer eggs were observed for 263 days and eggs that were released in artificial containers continued to produce new individuals until adult stage. These backswimmers produced a significant reduction in mosquito larval density in 5 sampling dates out of 7.

### **Odonatan predators**

Odonata larvae are voracious and important predators of mosquito larvae in freshwater ecosystems. They detect their preys by compound eyes and mechanical receptors and capture them with their labium.

The dragonfly larvae of *Trithemis annulata scortecii* were intense and active predators when used to control mosquito larvae, especially *Anopheles pharoensis*, in irrigation channels in Gezira Province, Sudan (EL Rayah, 1975). Bay (1974) reported that dragonfly larvae are known to prey heavily on bottom feeder mosquitoes like *Aedes* larvae. Sebastian *et al.* (1980) found that complete elimination of all *Ae. aegypti* larvae and pupae were achieved between day 4 and 9 depending on the density of aquatic stages of mosquitoes present per container when dragonfly larva, *Labellula sp.*, was used. The larval stages were found to last 2-3 months in the containers. This long life coupled with high predation rate is likely to make dragonfly larvae highly successful predators and could be used in biological control of *Aedes* mosquitoes. Again, Sebastian *et al.* (1990) conducted a pilot field study, involving periodic augmentative release of predaceous larvae of a dragonfly, *Crocothemis servilia*, to suppress *Ae. aegypti* during the rainy season in Yangon, Myanmar. Four laboratory-reared, three-week-old *C. servilia* larvae were placed in each major source of *Ae. aegypti* larvae immediately after the 3<sup>rd</sup> collection and then monthly for 3 successive months. The larval population of *Ae. aegypti* reduced to very low levels in 2 to 3 weeks and suppressed it

progressively until the trial ended. The adult mosquito population was greatly reduced after about 6 weeks and progressively diminished thereafter until the trial ended. Chatterjee *et al.* (2007) found that significant decrease in *An. subpictus* larval density in dipper samples was observed 15 days after the introduction of *Brachytron pratense* dragonfly larvae in concrete tanks under field conditions in India. Similarly, the larvae of 5 odonate species *Aeshna flavifrons*, *Coenagrion kashmirum*, *Ischnura forcipata*, *Rhinocypha ignipennis* and *Sympetrum durum* in semifield conditions in West Bengal, India, significantly lowered the mosquito larval density in dipper samples after 15 days from the introduction, followed by a significant increase of larval mosquito density after 15 days from the withdrawal of the larvae (Mandal *et al.*, 2008). These results (Sebastian *et al.*, 1980, 1990; Chatterjee *et al.*, 2007; Mandal *et al.*, 2008) are suggestive of the use of odonate larvae as potential biological agent in regulating the larval population of mosquito vectors.

Unlike the strong mosquito predation capacity of dragonfly larvae, damselfly larvae may feed less on mosquito larvae. Breene *et al.* (1990) found no mosquito larvae in the gut of the larvae of the damselfly *Enallagma civile*. Larvae gut contents analysis revealed that they preyed upon chironomid larvae and other aquatic invertebrates rather than mosquito larvae although they were observed in the pond where the larvae were collected.

Although odonate larvae have been investigated less compared to other predaceous aquatic insects, their long life cycle, predation capacity and sharing of habitats with mosquito immatures are advantageous for their being a potential biological control agents.

#### **Predator - Mosquito association by habitat type**

Several ecological studies of predator-prey associations involving mosquito larvae in different aquatic habitats have been documented. The following sections are reviewing this association beside factors influencing it in different habitats.

#### **Temporary water bodies (Habitats) associations:**

Many predaceous insects were found associated with both nuisance and mosquito vectors in temporary habitats such as man-made ponds, snow melt pools, rain pools, flood water pools and other different pools. McDonald & Buchanan (1981) found that mosquitoes colonized the man-made ponds within one day of formation followed by predaceous Coleoptera, Hemiptera and then Odonata. A significant inverse relationship was noted between mosquitoes and predators densities in 3 out of 4 trials.

Predators distributed in melted pools have been investigated by few scientists. Larson & House (1991) studied the arthropod fauna of small, acidic pools in a domed, ombrotrophic bog over an ice-free season. Taxa varied in abundance between pools of various classes and two principle communities were identified. Oligochaetes (segmented worms with few setae), beetles and mosquitoes dominated small, astatic pools and odonates, chironomids and several other taxa predominated in large, stable, vegetated pools. Within the large pools, odonate larvae were the dominant predators. In a similar study, Nilsson & Svensson (1994) compared assemblages of dytiscid water beetles and immature mosquitoes in two boreal snowmelt pools that differed chiefly in temperature owing to difference in shading and duration. The total abundance of dytiscids (including larvae) was similar in the two pools, whereas species richness was more than twice as high in the warmer, less ephemeral pools. The mosquito fauna of both pools were strongly dominated by *Aedes communis*, whose initial numbers were similar in the two pools, however, first-instar larvae suffered much higher mortality in the warmer pools.

A large number of different predator fauna have been associated with *Anopheles* larvae in different aquatic bodies. Lozano *et al.* (1997), found that the most abundant and diverse predators associated with *Anopheles albimanus* larvae in various hydrological types in southern Mexico, were aquatic Coleoptera (20 genera) followed by Hemiptera and Odonata (each with 16



genera). All the predators were significantly more abundant in temporary lagoons. Coleopterans and Hemipterans varied significantly among all locations however no significant difference was found in the abundance of odonates. Insect predators were correlated with occurrence of *Anopheles* immature stages in water bodies in south Punjab, Pakistan (Herrel *et al.*, 2001). Six *Anopheles* species and 9 insect predators were collected. Out of the 6 *Anopheles* species 4 (*An. subpictus*, *Anopheles culicifacies*, *Anopheles stephensi* and *Anopheles pulcherrimus*) were highly correlated with presence of predators. Mogi *et al.* (1995, 1999) studied the mosquito larvae and larvivorous predator communities on lands deforested for rice field development in dry and wet area in central Sulawesi, Indonesia. Collected predators were from 3 insect orders (Odonata, Hemiptera and Coleoptera). In the dry area, Anisoptera larvae, notonectids and dytiscids were the dominant predators while in the wet area; dytiscids, zygopterans and anisopterans were the dominant predators. Surface predators all belonged to order Hemiptera and they were less abundant than aquatic predators. Munga *et al.* (2007) identified seven families (Hydrophilidae, Dytiscidae, Corixidae, Nepidae, Notonectidae, Belostomatidae, and Corduliidae) of larval mosquito predators from the larval habitats (drainage ditches, cow hoofprints and disused goldmines) of the malaria vector *An. gambiae* s.l. in natural habitats in Western Kenya Highlands. Predator density in disused goldmines was significantly higher than that of other habitat types. Invertebrate predators were found to associate larvae of the malaria vector *An. albimanus* in 78.6% of the body types harbouring immature mosquitoes in a low-lying area of Haiti (Caillouët *et al.*, 2008). Larval *An. albimanus* and associated predators were found in permanent and semi-permanent groundwater habitats including (in order of greatest abundance) hoof/footprints, ditches, rice fields, and ground pools. Predators were dominated by order Coleoptera (Hydrophilidae and Dytiscidae) followed by orders Hemiptera (Belostomatidae, Corixidae, Notonectidae

and Gerridae), Odonata (Libellulidae, Aeshnidae and Coenagrionidae), Ephemeroptera (Baetidae) and Diptera (Syrphidae), respectively.

Fischer *et al.* (2000) described the seasonal variations of insect community of the rain pools during a 1-year period. A total of 45 insect taxa were identified: 18 Coleoptera, 15 Diptera, 9 Heteroptera, 1 Ephemeroptera and 2 Odonata. Culicid mosquitoes represented 76 % of the pooled abundance of insects. The maximum richness of entomofauna was at the end of the summer (32), in coincidence with maximum rainfall and temperature whilst the minimum faunal richness (2) was recorded during the spring drought. Similarly, Fischer & Schweigmann (2008) found six mosquito species and 23 predatory insect taxa in temporary rain pools during the summer and fall season in Buenos Aires city. Both mosquito immatures and predators were disproportionately more abundant in pools with high flooded surface, depth, and duration. In another study, Campos *et al.* (2004) found that 41 predaceous insect taxa associated with the floodwater mosquito *Ochlerotatus albifasciatus* from spring to fall. Coleoptera and Diptera were dominant and diverse while Ephemeroptera and Odonata were scarce in numbers and species. Six lentic aquatic habitats: (1) cemented temporary pools (cemented walls); (2) cemented open water storage tanks (mainly for rain water storage); (3) house hold water storage tanks (large plastic containers to buckets); (4) stagnant stream side pools; (5) temporary roadside ditches; and (6) clogged sewage drains were found to be hosting mosquito immatures and predators in Darjeeling Himalaya, India (Aditya *et al.*, 2006). *Toxorhynchites splendens*, dytiscids (Coleoptera) and odonates were associated with mosquito immatures in both temporary pools and cement tanks whilst gerrids (Hemiptera) were associated with mosquito immatures in temporary pools, stream pools and sewage drains. The population of *Tx. splendens* immatures was positively correlated with the population mosquito immatures ( $r = 0.071$ ).

### **Tree-hole associations:**

A variety of invertebrates utilize tree holes as breeding sites. Because they are primary breeding sites for many disease vectors, particularly mosquitoes and biting midges, tree holes are an economically important habitat (Yanovisk, 2001). Although *Toxorhynchites* mosquitoes are well known as tree-hole mosquito predators, several other predaceous insects are important tree-hole occupants. This article primarily focuses on predators other than *Toxorhynchites* mosquitoes since they were recently reviewed by Collins & Blackwell (2000), however, they will be briefly mentioned.

Commonly recorded predaceous insects in tree holes are dragonflies, damselflies and a new genus of water bug in the family Veliidae. Tree holes are not the primary habitat for odonates. Out of approximately 6000 species, only 47 have been reported from this habitat, at least 64% being Zygoptera (Corbet, 1983). Anisopteran and zygopteran larvae inhabiting tree holes were recorded in forests of different geographical regions. Orr (1994) reported that *Pericnemis triangularis* (Coenagrionidae), *Indaeschna grubaueri* (Aeshnidae) and *Lyriothemis cleis* (Libellulidae) were breeding in phytotelmata in the understorey of lowland mixed dipterocarp rainforest in Borneo. Corbet & McCrae (1981) collected 2 large nymphs of *Hadrothemis scabrifrons* from a water containing cavity in a tree root in lowland rainforest near the Kenya coast. Larvae of the anisopteran odonate *Hadrothemis camarensis* (Kirby) were found in water-containing tree holes, in Kakamega forest, western Kenya (Copeland *et al.*, 1996). Larvae were collected during 4 consecutive years of sampling in 46% of tree holes, and in 26% of tree-hole samples. Larvae were more likely to be found in tree holes during wetter months. Distribution of larvae among tree holes was clumped. Larvae occurred more often in tree holes of larger surface area and gape size. These attributes correlated positively with median water volume (0.15 - 42 L) and height above the forest floor (up to 22.45 m). Larvae of

chironomidae and culicidae predominated numerically among prey of odonate larvae, with smaller larvae preying more on the former and larger ones on the latter. Two other insect predators were encountered in tree holes: *Toxorhynchites* sp. and a new genus of the water bugs of family Veliidae (order: Hemiptera). Veliids were found in 11.2% and *Toxorhynchites* sp. in 41% of treehole samples for which their presence or absence was noted. Neither taxon was associated negatively or positively with the occurrence of odonates. Louton *et al.* (1996) surveyed the aquatic macrofauna of water-filled internodes of *Guadua* bamboo in a lowland tropical forest in Peru. They found a community of 29 species dominated by Diptera and Odonata. The predaceous insects comprised 4 damselflies (*Mecistogaster jocaste*, *Mecistogaster linearis*, unknown *Mecistogaster* species and *Microstigma rotundatum*) and Dipterous larvae including family Ceratopogonidae (subfamily: Ceratopogoninae) and family Culicidae (*Toxorhynchites* sp. and the facultative predators *Sabethes* spp. A and B, and *Trichoprosopon pallidiventer* and *Trichoprosopon* sp.). Besides the predaceous mosquito, *Toxorhynchites theobaldi*, larvae of 5 common species of Odonata (*Gynacantha membranalis*, *Triacanthagyna dentata*, *M. linearis*, *Mecistogaster ornata* and *Megaloprepus coerulatus*) were collected from water-filled tree holes in a lowland forest in Panama (Fincke, 1999). Another study for the macrofauna of water-filled tree holes on Barro Colorado Island, Panama revealed the presence of 54 macroinvertebrate taxa (Yanovisk, 2001). Most of the species were in the insect order Diptera and out of the total fauna, 36% (20 species) were mosquito predators in the insect orders Hemiptera (2 species), Coleoptera (2 species), Odonata (6 species) and Diptera (10 species). Interestingly, Yanovisk (2001) reported that *Cx. allostigma* and *Sigmatomera amazonica* prey on mosquitoes in water-filled tree holes on Barro Colorado Island, Panama.

### **Rice field associations:**

The following studies showing that predator complex is a major source of mortality for immature stages of mosquitoes in rice fields and strongly supports the hypothesis that natural enemies should be an important component in rice field mosquito control program.

Except for damselfly larvae, predaceous insects were significantly more abundant in rain-fed fields than in irrigated fields of northern Sulawesi, Indonesia (Mogi *et al.*, 1995). Various factors could be involved such as the scarcity of submerged plants that provide oviposition substrates, perching sites and refuges for some aquatic predators, and emergent and floating vegetation which obstruct oviposition by some predators. Larvivorous fish may reduce the abundance of insect predators and significant detrital interaction also may exist among insect predators. Furthermore, insecticides and other chemicals for rice production probably are used more frequently in irrigated fields than in rain-fed fields. According to plant age and maturity, damselfly larvae were more abundant in mature and harvested fields, whereas dragonfly larvae, Notonectidae, Vellidae, Hydrophyllidae and Dytiscidae were often abundant in ploughed and young fields. Another study investigating the colonization of rice fields by mosquitoes and larvivorous predators in asynchronous rice cultivation areas in the Philippines was conducted by Mogi & Miyagi (1990). The samples were taken from rice fields at 6 different phases of maturity (fallow, ploughed, nursery, newly transplanted, after tillering, mature). Dytiscidae, Anisoptera and Zygoptera were the primary aquatic predators in fallow or mature fields while Hydrophilidae and Notonectidae had no clear succession patterns. Nepidae were collected only from mature fields. Among surface predators, Vellidae was most abundant in fallow fields (in one study site) and in planted fields (in the other study site) and other predators were rare. These results indicated that the abundance of aquatic predators decreased at the onset of ploughing and then recovered slowly as rice plants grew. In case of surface

predators, the pattern is similar but less conspicuous.

Notonectids, dytiscids and larvae of Anisoptera and Zygoptera were among biotic factors influencing the abundance of Japanese encephalitis vectors in rice fields in India (Sunish & Reuben, 2002). Notonectid populations decreased with rice plant growth and were the most abundant insect predators. Dytiscids dominated the early weeks of the cultivation cycle but Anisoptera and Zygopteran larvae were also abundant early in the cycle. Multiple regression analysis showed that notonectids (both nymphs and adults) were negatively associated with larval abundance. While the impact of Zygoptera was observed only during short and long-term crop seasons, dytiscids showed a significant mortality factor for mosquito larvae once during the summer season. In a latter study, Sunish *et al.* (2006) mentioned that predatory notonectids, anisopterans and dytiscids significantly influenced the survival of immatures of *Cx. vishnui* complex, a Japanese encephalitis vector, in rice fields in Southern India.

Andis & Meek (1985) studied mortality and survival patterns for immature of *Psorophora columbiae* in the laboratory setting and in rice fields in Louisiana, USA. Predators consumed at least 24% (younger age classes) of the larvae in each field, a maximum of 56% (older age classes) and were the most significant mortality factor for immature *Ps. columbiae* in rice fields. Total mortality of the mosquito larvae was high with only 2.6% surviving to the pupal stage. It can be inferred that predation may be restricted to older age classes and reduce larval survival, which finally lead to a reduction in the adult *Ps. columbiae* population density. Likewise, insect predation was the most important mortality factor for mosquito larvae and pupae in Philippine rice fields (Mogi *et al.*, 1984). Survival from hatching to emergence was 50 - 88.8% in predator - free cages set in the rice fields, whereas survival of natural populations exposed to predators was 0.0-1.8% for *Culex* and 1.1-4.7% for *Anopheles* in the same rice fields. In Thai rice fields,

mortality of immature anopheline mosquitoes attributed to aquatic predators was variable (19-54%) and correlated positively with the predators abundance. Surface predators were a non-significant minor mortality factor (0-10%) (Mogi *et al.*, 1986). Diabaté *et al.* (2008) found that emergence success of *An. gambiae* in Rice fields and puddles experiments in Burkina Faso was significantly affected by predaceous insects. The backswimmers *Anisops* sp. and *Anithares* sp. (Hemiptera: Notonectidae), the water boatman *Micronecta* sp. (Hemiptera: Corixidae), the dragonfly *Tramea* sp. (Odonata: Libellulidae) and the beetles *Berosus* sp. and *Laccophilus* sp. (Coleoptera: Hydrophilidae and Dytiscidae) were associated to *An. gambiae* larvae. The number of predators was higher in rice fields than in puddles and the backswimmers were the most abundant predators in both rice field and puddles with a mean collection of 45.7 and 21.8 predators/m<sup>2</sup>, respectively.

#### **Recent techniques for detecting predator – mosquito associations:**

In addition to classical surveys, recent techniques could be used to detect natural predators associated with mosquito larvae. By using Precipitin tests performed on the gut contents of possible predators collected from different areas and habitats in Kenya, Service (1973) identified Coleoptera and Diptera as insect predators. In a later study, Service (1977) used the same technique to compare mosquito predator fauna in rice fields, pools and ponds in Kenya. Forty two predator species were identified, the most important of which were Coleoptera larvae, Hemiptera and predaceous adult Diptera. Rice fields harbored more predator fauna than temporary pools and small ponds. A DNA-assay was utilized to confirm predation among larvae of the *An. gambiae* complex (Koenraadt & Takken, 2003). Furthermore, a range of molecular techniques and applications that allow prey to be identified, often to the species and even stage level, were reviewed by Symondson (2002). These techniques include enzyme electrophoresis,

a range of immunological approaches utilizing monoclonal and polyclonal antibodies to detect protein epitopes, and the polymerase chain reaction (PCR)-based methods for detecting prey DNA. The PCR-based techniques are displacing all other methods since they have been shown to be highly effective and more reliable.

#### **Predators' influence on mosquito oviposition, development, survival, abundance and fitness**

Research findings indicating that the development, survival and abundance of larval mosquito populations in the field are limited by predaceous insects which are primarily responsible for mortality in immature stages of mosquitoes. This effect has been reported in many different aquatic habitats and is responsible for restraining the density of such prey populations below the critical threshold where transmission of diseases could not occur (Das *et al.*, 2006).

#### **Predators' influence on mosquito oviposition:**

Animals take risk of predation into account when making decisions about how to behave in particular situations. Chemosensory cues are important and are used to detect the presence of predators or even their presence in the immediate past, and may also provide information on predator activity level and diet (Kats & Dill, 1998). In their review article for chemical detection of natural enemies by arthropods, Dicke & Grostal (2001) reported that of all chemical information gathered by animals, cues about predation risk usually has important and immediate consequences for the future fitness of animals and they result in various responses strategies towards such predators including avoidance. For all mosquito species, location and selection of an oviposition site is essential life-cycle behavior and involves visual, olfactory and tactile responses (Bentley & Day, 1989). Oviposition is an important component of most mosquito borne diseases because pathogen acquisition by mosquito vectors usually requires taking of at least one blood meal and disease transmission usually

requires the completion of at least one oviposition cycle before pathogen transfer can occur with subsequent blood meal.

It was believed for a long period that predator-prey interaction was largely attributed to predation mechanisms until Chesson (1984) showed that the effect of notonectid bugs on mosquito larvae is mainly due to selective oviposition by gravid mosquitoes. He manipulated the density of aquatic predaceous bugs (*N. hoffmani* and *N. kirbyi*) in stock troughs to assess the predator's effect on mosquito larvae. Over a three-month sampling period very few large mosquito larvae or pupae were collected from the side of the trough where notonectids were located, whereas large densities were collected from the side free from notonectids. To ensure that this was not an artifact of the side of the trough chosen for notonectid addition or removal, the predators were moved from one side to the other and same results were obtained. It was thought that these experimental results could be explained by selective mosquito oviposition. This hypothesis was supported by laboratory experiments in which female mosquitoes laid the fewest egg rafts in tubes containing the predaceous notonectids. Moreover, laboratory and field experiments also demonstrated that notonectids may disrupt mosquito egg rafts, but no evidence of a reduction in subsequent hatching success was obtained. This means that the predator does not feed on the mosquito egg rafts and confirms the selective oviposition hypothesis. Other predators such as dragonfly larvae, however, consume egg rafts. Likely, later studies have investigated ovipositional responses of the mosquito *Culiseta longiareolata* to some insect predators.

Stav *et al.* (1999) reported that the predaceous dragonfly larvae of *Anax imperator* produced 52% reduction in *Cs. longiareolata* oviposition in outdoor artificial pools. The reduced number of *Cs. longiareolata* egg rafts found in the presence of *A. imperator* was largely due to oviposition habitat selection by *Cs. longiareolata* females. Larvae of *Cs. longiareolata* were highly vulnerable to

predation compared to *Cx. laticinctus* and were also the only dipteran species that avoided *Anisops* pools when ovipositing (Eitam *et al.*, 2002). Stav *et al.* (2000) found that the egg rafts of the mosquito *Cs. longiareolata* deposited in the free *Anax* treatment were fewer than deposited in caged *Anax* and control treatments. There was no statistically significant difference in the number of egg rafts between control and caged *Anax* pools which means that, while *Culiseta* females oviposit fewer egg rafts in the presence of *Anax*, they did not respond to predation risk from the caged *Anax*. In general, this individual response could have population-level consequences. For instance, it may increase the equilibrium size of the *Cs. longiareolata* population relative to the population in which oviposition is discriminative with respect to *Notonecta maculata* (Spencer *et al.*, 2002). Furthermore, females of the malaria mosquito vector *An. gambiae* laid significantly fewer eggs in rainwater conditioned with the predatory backswimmer *Notoecta* sp. than in unconditioned rainwater, indicating that predators influence selection of oviposition site by this malaria mosquito vector (Munga *et al.*, 2006). More interestingly, females of the malaria mosquito vector *An. gambiae* s. l. tend to avoid oviposition sites containing older instar larvae of the *An. gambiae* (McCrae, 1984). The reason was discovered later on to be avoidance of offspring predation by older instar larvae (Koenraadt & Takken, 2003).

These previously mentioned studies showed that notonectid bugs and dragonfly larva *A. imperator* affect oviposition habitat selection in some mosquito species at a stable density of these predators. However, the relationship between predator density and mosquito oviposition response was not studied until Eitam & Blaustein (2004) tested the oviposition response of 2 mosquito species, *Cs. longiareolata* and *Cx. laticinctus*, to a range of the predator *N. maculata* in artificial pools. Both mosquito species oviposited less in predator pools but the response was not related to the predator density, whereas vulnerability of *Culiseta*

immatures to predation was density dependent. So, although mosquitoes can detect the predator at any density, they may be unable to discriminate predator density. The vulnerability of *Culiseta* to predation could thus be due to mitigating effects of the biotic community inside the pools. Similarly, effects of pool depth combined with risk of predation on oviposition habitat selection by *Cs. longiareolata* were studied recently (Arav & Blaustein 2006). Results indicated that although *N. maculata* affected oviposition pattern of this mosquito, pool depth did not affect oviposition habitat selection for this mosquito.

All these studies have not assessed the mode of detection of predators until Blaustein *et al.* (2004) demonstrated and confirmed that the cue for oviposition avoidance of *Cs. longiareolata* to *N. maculata* was a predator-released chemical (kairomone): *Notonecta* water (without *Notonecta* replenishment) repelled oviposition for 8 days. Consequently, this mode of detection is an advantage for predators and it is very important from the mosquito control point of view whereas such kairomones could be produced commercially for mosquito control. Furthermore, oviposition habitat selection in *Cs. longiareolata* is an adaptive response to the trade-off between the risk of predation and negative density-dependent effects (Spencer *et al.*, 2002) whilst findings of Kiflawi *et al.* (2003) suggest that it is driven by a mixed strategy, played by all females, whereas all females follow a single, simple behavioral 'decision rule' that is responsible for the lack of complete predator avoidance. Mosquitoes may detect predators cues either from the air, when the cues possess sufficient volatility, or by a gustatory mechanism involving direct contact with the water, when the cues possess low volatility (Clements, 1992). Silberbush & Blaustein (2008) tested whether *Cs. longiareolata* can detect the chemical cues from *N. maculata*, without touching the water. *Cs. longiareolata* oviposited significantly more in the central pools surrounded by channels containing control water than in pools surrounded by *Notonecta* conditioned water

channel (56 of 81 egg rafts (69%) were oviposited in the control pools) indicating that gravid *Cs. longiareolata* females detected predators cues from the air which means that predator-released cues (kairomones) are air-borne cues.

The predators cues not only affecting mosquitoes' oviposition but also alter their life cycle traits (Beketov & Liess, 2007). Results of their experiments showed that chemical cues from the predator *N. glauca* feed with prey's (*Cx. pipiens*) conspecifics caused a decrease survival, delayed immatures development and reduction in body size of emerged mosquitoes while chemical cues from predators fed with *Daphnia magna* (a crustacean invertebrate animal) produced only delayed development. The effect of the cues on larval development and body size of imagoes were significantly stronger for females than for males which is very important for mosquitoes suppressing particularly diseases vectors.

In summary, selection of oviposition site by female mosquitoes depends more on the presence of predators and less on predator density. Furthermore, predator density, as indicated by the concentration of their kairomones, could affect the oviposition deterrent potential and would be an important consideration in utilizing either predators or their kairomones for biological control of mosquitoes.

#### **Predators' influence on mosquitoes development, survival, abundance:**

Influences of predaceous aquatic insects on the development, survival and abundance of important *Aedes*, *Anopheles* and *Culex* mosquito vectors are briefly summarized in Table 2. Unlike *Culex*, information about role of predators on development, survival and abundance of *Aedes* and *Anopheles* mosquitoes is limited.

In some cases, variations in predation effects are due to difference in predator species breeding with the mosquito species. For instance, role of predators on the development and survival of immature stages of *Cx. annulirostris* in different regions in Australia was variable. In Victoria, McDonald & Buchanan (1981) mentioned

that the survival rate of *Cx. annulirostris* from egg hatch to eclosion was 11% and predation by associated Coleoptera, Hemiptera and Odonata was estimated to be largely responsible for the low survival. In

the Brisbane area of southeast Queensland, Mottram & Kettle (1997) found that predator densities in three surveyed sites were significantly different, being lowest in the temporary pools (0.32 %) and highest in the

Table 2. Influences of predators on mosquito development, survival and abundance

Mosquitoes	Predation influence	Reference
<i>Anopheles albimanus</i>	Mosquito production was negatively associated with predators	Marten <i>et al.</i> , 1996
<i>An. arabiensis</i>	Populations of predators (notably odonates) were one of the conditions associated with the abundance of <i>An. arabiensis</i> larvae in market-garden wells.	Robert <i>et al.</i> , 1998
<i>An. gambiae</i> s. l.	Predators, parasites and pathogens have been identified as major causes of larval mortality up to 98%.  Field experiment indicated that emergence success was over 3 fold higher in predator free cages than in cages with predators (164.8 adults/cage and 49.6 adults/cage respectively).	Service, 1973 & 1977  Diabaté <i>et al.</i> , 2008
<i>An. gambiae</i>	Survival increased when first stage larvae introduced into semi permanent pools, before and after removing the natural fauna from 3% (presence of fauna), to 58% when the fauna had been removed. When a number of first stage larvae was introduced daily, survival to pupation increased from nil to 7-20% when the other fauna had been effected.	Christie, 1958
<i>Aedes aegypti</i> & <i>Ae. triseriatus</i>	When exposed to <i>Tx. rutilus</i> (hatch to adult) <i>Ae. aegypti</i> usually failed to produce adults whilst <i>Ae. triseriatus</i> always produced adults.	Grill & Juliano, 1996
<i>Ae. scapulari</i>	Immature stages were found in water reservoirs where aquatic insects are not observed but no mosquito larvae were found when predators were found.  Mortality ranged from 68 to 96 % and was the most important cause of death and was the key-factor best accounting for the population fluctuations of this mosquito species.	Garcia <i>et al.</i> , 1996  Casanova & Do Prado, 2002
<i>Culex annulirostris</i>	Survival rate from egg hatching to eclosion was 11%.  Predators killed 69.1%, 68.7% and 43.2% of immatures in the flooded grassland, semi-permanent pool and temporary pool respectively.  Predators dominated by dytiscids and dragonfly naiads, reduced larval survival by 58%.	McDonald & Buchanan, 1981  Mottram & Kettle, 1997  Rae, 1990
<i>Cx. pipiens</i>	<i>Anax imperator</i> caused statistically significant reduction (32.4%) in the number of <i>Cx. pipiens</i> larvae surviving to the pupal stage.	Stav <i>et al.</i> , 2005
<i>Cx. quinquefasciatus</i>	Immature stages were found in water reservoirs where aquatic insects are not observed but no mosquito larvae were found when predators were found.  <i>Sphaerodema annulatum</i> significantly reduced the rate of pupation (6 – 35) and adult emergence (0.4 – 28.8 per day) under laboratory conditions.	Garcia <i>et al.</i> , 1996  Aditya <i>et al.</i> , 2004

<i>Cx. tarsalis</i>	In insecticide treated pond, larval population densities fluctuated between 0 and 15/dip, while in the untreated pond, where different predaceous insects are found, population densities remained low and never approached a 1/dip level.	Miura <i>et al.</i> , 1978
	Predation mortality ranged from 3.7 to 84.5% and was the most important cause of death at 5 of 6 study sites.	Reisen <i>et al.</i> , 1989
	Predation by coleopteran larvae significantly affected larval population.	Walton <i>et al.</i> , 1990
<i>Cx. tritaeniorhynchus</i>	Mortality from egg hatching to adult emergence was 95.2, 95.7 and 93.9% in rice fields, borrow-pits and groud-pools respectively.	Apiwathnasorn <i>et al.</i> , 1990
	Predators complex were very important factor for the larval population in fallow rice fields while adult emergence rate was very low in the presence of the predators, the average being 0.02 and the higher the predator density the lower the emergence rate.	Mogi <i>et al.</i> , 1980
	Effect of predation on the survivorship ranged from 0.017% during the monsoon to 0.725% during the postmonsoon season.	Reisen & Siddiqui, 1979
<i>Culiseta longiareolata</i>	<i>Anax imperator</i> reduced <i>Cs. longiareolata</i> larvae survival to the pupation (78%).	Stav <i>et al.</i> , 2005

flooded grassland (1.76 %). The mortality calculations suggested that predators killed 69.1%, 68.7% and 43.2% of immature *Cx. annulirostris* in the flooded grassland, semi-permanent pool and temporary pool, respectively. For the same mosquito species at the Ross River dam in Townsville, North Queensland, Australia, the invertebrate predators, dominated by dytiscids and dragonfly larvae, reduced larval survival by 58% (Rae, 1990).

In other cases this variation is attributed to both mosquito species and different predators. The most obvious example is the field experiment that has been conducted by Lundkvist *et al.* (2003) in artificial ponds over two successive years to determine how population levels of mosquito larvae are affected by predaceous diving beetles (Dytiscidae). Mosquitoes that colonized the ponds were predominantly species of the genus *Culex*. In 2000, most of the dytiscids that had colonized the ponds were small (*Hydroporus* spp.) and had no impact on the size of larval mosquito populations. Conversely, in 2001, larger beetles (*Ilybius*, *Rhantus* and *Agabus* spp.) were more common and mosquito larvae were significantly fewer in ponds with high numbers of dytiscids.

A recent study conducted by Das *et al.* (2006) mentioned that breakdown of predator populations was responsible for the sudden increases in vector populations above the threshold for disease transmission during heavy rainy periods. In rice fields, notonectid predators exhibited a significant positive correlation with *Cx. vishnui* larvae. Important predators recorded in shallow pools were notonectids, damselfly larvae, *Diplonychus indicus* and hydrophilids. Dragonfly larvae and gerrids were recorded in cement tanks. The conclusion was that rice fields are stable ecosystems where regular interaction occurs between mosquito larvae and their natural enemies and a sudden increase in the mosquito population is uncommon. Contrarily, in transient habitats (shallow water pools and cement tanks) no such stability is present and they become more important as breeding habitats in terms of seasonality and number.

Predators inhabiting water-filled tree holes are known to decrease the prevalence of mosquito larvae. Predation by predaceous midge larvae, *Pentaneura* sp., produced low densities of mosquito larvae found in the water field bracts of *Heliconia imbricate* (Naeem, 1988). This predation affected 2 mosquito species, *Wyeomyia pseudopecten*,



a resident species, and *Trichoprosopon digitatum*, a non-resident species. Predation kept resident mosquito densities low while completely excluded the non-resident mosquito from the bracts. Larvae of 4 common species of odonata, a mosquito and a tadpole were the major predators collected from tree holes in the lowland moist forest of Barro Colorado Island, Panama, and mosquito larvae were their common prey (Fincke *et al.*, 1997). Tree holes colonized naturally by predators and prey had lower densities of mosquitoes if odonates were present than if they were absent. While controlling for the quantity and species of predator, hole volume and nutrient input were tested by using artificial tree holes placed in the field. In large and small holes with low nutrient input (number of mosquito larvae), odonates suppressed both the number of mosquitoes present and the number that survived to pupation. Increasing nutrient input (and consequently, mosquito abundance) to abnormally high levels damped the effect of predation when odonates were relatively small. However, the predators grew faster with higher nutrients, and large larvae in all three genera reduced the number of mosquitoes surviving to pupation, even though the abundance of mosquito larvae remained high. The presence of a 4<sup>th</sup> instar *Tx. rutilus* significantly reduced the abundance of late stage *Ae. triseriatus* mosquitoes (Lounibos *et al.*, 1997). The pupal stage of this prey was more negatively affected by *Tx. rutilus* (Bradshaw & Holzappel, 1983) than other tree-hole mosquitoes in Southern North America. Extinctions of aquatic stages of *Ae. triseriatus* within tree holes were common, but in most holes were not significantly associated with the presence of *Tx. rutilus*, indicating that predation does not routinely drive mosquito prey locally extinct in this ecosystem.

#### **Predators' influence on mosquitoes fitness:**

The influence of predators on mosquito fitness was first reported by Lounibos *et al.* (1993) when they investigated the influences of food type and predation on fitness of the

treehole mosquito *Ae. triseriatus*. Results indicated that the presence of *Tx. rutilus* significantly affected the fitness of *Ae. triseriatus* to a greater degree than food type. Survivorship of immature stages in cohorts with predator access was very low while the mean of P<sub>50</sub> (time to 50% pupation) was significantly greater than cohorts with detritus. The female size in cohorts with the predaceous mosquito, particularly wing size, was unexpectedly smaller than other cohorts with or without food access. It was suggested to be due to the fact that the presence of *Tx. rutilus* may reduce movement of larval *Ae. triseriatus*, thereby decreasing food intake and size at metamorphosis. Additionally, Dicke & Grostal (2001), in their review for chemical detection of natural enemies by arthropods, reported that predation risk usually has important and immediate consequences on prey fitness. Lundkvist *et al.* (2003) found a negative correlation between the number of diving beetles in artificial ponds and the mean body length of mosquito larvae that has serious consequences on the fitness of emerging females.

More interestingly, recent study (Diabaté *et al.*, 2008) indicated that predaceous insects influencing the divergent selection amongst the molecular forms of the malaria vector *An. gambiae*. Predation increased the developmental success of larvae of M form over the S form in both puddles and rice fields. Higher density of predators belonging to Notonectidae (*Anisops* sp. and *Anithares* sp.) and Dytiscidae (*Laccophilus* sp.) families increased the relative success of the M form whilst higher density of Libellulidae (*Tramea* sp. ) and Hydrophilidae (*Berosus* sp.) specimens appeared to decrease the relative success of the M form, but their effects were not significant.

#### **Factors' influencing capacity of predaceous insects**

Factors influencing predation potential of predaceous insects are shown in Table 3. These factors are classified into biological and physical. Unfortunately, the literatures have shown that investigations for factors influencing predation capacity of aquatic

beetles are limited compared to the other predaceous insects (Diptera, Hemiptera and Odonata).

Increased predation of one mosquito prey species over another by a predaceous insect does not always mean a real preference since it could be due to differences in their means of evaluating predation risk (Sih, 1986). Sih (1986) reported that the behavior of *Ae. aegypti* larvae towards the predator *N. undulata* was a response to disturbance per se, whilst the *Cx. pipiens* response was mediated by chemical cues that may have involved a combination of notonectid digestive enzymes and partially digested mosquito materials, associated with the actual predation act. So, because *Cx. pipiens* has an evolutionary history of contact with notonectids, it suffered a lower predation rate from *Notonecta* than did the *Ae. aegypti* that lacking this evolutionary behavior. Consequently, the reduced predation rate could be explained as *Cx. pipiens* showed both stronger and more precise antipredator responses than *Ae. aegypti*. Further to this, the findings of Husbands (1978) imply that prey behavior could influence its persistence in the mosquito larvae-notonectid system. He found that notonectid predators quickly destroyed *Aedes nigromaculis* compared to *Culex tarsalis* due to the former showing little reduction in their movement or shift in their habitat use but, in contrast, the latter shifting to feed quickly among emergent vegetation. Grill & Juliano (1996) also suggested that in some systems, prey behavior patterns are more related to vulnerability to predation. In further confirmation of this hypothesis, *Ae. albopictus* did not respond to cues produced by *Tx. rutilus* and was more vulnerable to predation than *O. triseriatus* (Kesavaraju & Juliano 2004).

Collins & Resh (1985) stated other factors influencing the capacity of damselflies at Coyote Hills Marsh, Fremont, CA. The damselfly microdistribution, age-specific feeding habits, phenology, and the architecture of the habitat that supports the larvae were anticipated to reduce the predaceous capacity of *Enallagma civile*, *E.*

*carunculatum* and *Ischnura cervula* against *An. occidentalis*.

Lee (1967) found that mosquito larvae are consumed more than pupae by predators and assumed that this was due to the inclination of pupae to exhibit rapid tumbling action when startled. Contrarily, both bugs of family belostomatidae (Order: Hemiptera) and *Toxorhynchites* mosquito larvae have an advantage over the other aquatic predaceous insects that restrict their prey selection to the larval instar only. This is worthy of note, in particular for mosquito vectors of diseases, since pupal reduction directly reduces mosquito emergence and subsequent disease transmission.

### ***Predaceous insects and integrated mosquitoes control***

The concept of integrated control is a fairly specific one, which historically has meant the use of a combination of chemical and biological agents in as compatible a manner as possible (Axtell, 1979). Sometimes cultural and/or physical control methods have been included. The role of biological control agents, especially arthropod predators, in integrated vector control (IVC) was reviewed by Lacey & Orr (1994). They mentioned that selection of candidate biological control agents for integrated vector control would depend on a variety of factors including efficacy, cost consideration, environmental impact and compatibility with other interventions.

The microbial insecticides *Bacillus thuringiensis* and *Bacillus sphaericus* were combined with predaceous insects more often than other control measures in integrated mosquito control. The toxin of *B. t.* serotype H14 was applied to control *Cx. tarsalis* mosquitoes in pesticide-sensitive habitats (Mulligan & Schaefer, 1981). Complete control of *Cx. tarsalis* at a wildlife area was obtained with *B. t.* H14 at 0.8 kg/ha and predation by naturally-occurring aquatic beetle larvae extended the control of *Cx. tarsalis* through 22 days after treatment. *B. t.* H14 was innocuous to the selected non-target fauna. Similarly, application of *B. t.* H14 at 1.1 kg/ha reduced *Cx. tarsalis* numbers by 93% at a duck club without affecting

Table 3. Factors influencing predation potential of aquatic predaceous insects

Factor	Predator	Mosquito prey	Influence	Reference
Prey species	<i>Notonecta undulata</i> (Hemiptera)	mosquito larvae & other insect preys	Mosquito larvae preferred over the other preys	Ellis & Borden, 1970
	<i>Cx. (Lutzia) fuscamus</i> (Diptera)	<i>Aedes, Anopheles &amp; Culex</i> <i>Ae. aegypti, An. stephensi</i> & <i>Cx. quinquefasciatus</i>	No apparent preference by <i>Cx. fuscamus</i> Predation was generally highest against <i>An. stephensi</i> followed by <i>Cx. quinquefasciatus</i> and <i>Ae. aegypti</i> .	Bai <i>et al.</i> , 1982 Kuldip <i>et al.</i> , 1984
Biological	<i>Emiliares indica</i> (Hemiptera)	<i>An. stephensi</i> & <i>Cx. quinquefasciatus</i>	Feeding rates were higher for first instar larvae than for pupae	Wattal <i>et al.</i> , 1996
	<i>Labellula</i> sp. (Naiad) (Odonata)	<i>Ae. aegypti</i> (larvae and pupae)	Consumed both stages with preference towards larvae, particularly smallest ones, over pupae	Sebastian <i>et al.</i> , 1980
	<i>Siagra hoggarica</i> (Hemiptera)	<i>Cx. quinquefasciatus</i> (larvae and pupae)	The predatory efficacy was highest against first larval instar and it decreased as the larvae grew older under laboratory and field conditions	Alahmed <i>et al.</i> , 2009
Prey stage	<i>Tx. rutilus rutilus</i> (Diptera)	<i>Ae. aegypti</i>	More 4 <sup>th</sup> instar prey was consumed significantly than pupae or 1 <sup>st</sup> instars, but they killed without eating, significantly more pupae than 4 <sup>th</sup> instars and no 1 <sup>st</sup> instar killing was observed.	Padgett & Focks, 1981
	<i>Ochthera chatybesceens</i> (Diptera)	<i>An. gambiae</i> s.s.	Significantly more 2 <sup>nd</sup> instar larvae were consumed than pupae when they were both available.	Minakawa <i>et al.</i> , 2007
Prey size	<i>Buenaia</i> sp. (Hemiptera)	<i>Cx. pipiens quinquefasciatus</i>	Predation rate exhibited by <i>Buenaia</i> sp. was the same, and was independent of prey body size.	Rebollar-Tellez <i>et al.</i> , 1994
	<i>Culiseta longiareolata</i> (Diptera)	<i>Cx. quinquefasciatus</i>	4 <sup>th</sup> instar larvae of <i>Cs. longiareolata</i> fed on 1 <sup>st</sup> , 2 <sup>nd</sup> and 3 <sup>rd</sup> instar larvae of <i>Cx. quinquefasciatus</i> . The rate of prey consumption was 1.65 larvae/day/predator.	Al-Saadi & Mohsen, 1988
	<i>Corethrella appendiculata</i> (Diptera)	<i>Ae. albopictus</i> & <i>Ochlerotatus triseriatus</i>	Second instars of species were more vulnerable to predation than were 3 <sup>rd</sup> instars and the 3 <sup>rd</sup> instar <i>Aedes</i> was more vulnerable than <i>Ochlerotatus</i> of the same stage.	Kesavaraju & Juliano, 2004

<i>Notonecta hoffmani</i> (Hemiptera)	mosquito larvae	preference first increased and then decreased with increasing prey size	Scott & Murdoch, 1983
<i>Tx. towadensis</i> (Diptera)	mosquito larvae	Consumption of 2 <sup>nd</sup> instar prey increased convexly toward an upper asymptote, however sigmoid association was observed with 4 <sup>th</sup> instar prey. Although younger predaceous larvae consumed more 2 <sup>nd</sup> instar prey than 4 <sup>th</sup> instar, older predaceous larvae preferred 4 <sup>th</sup> instar prey	Yasuda & Hagimori, 1997
	<i>Cx. fatigans</i>	With increasing prey size, <i>Cx. raptor</i> require 1,6 and 62 minutes to handle single 2 <sup>nd</sup> , 3 <sup>rd</sup> and 4 <sup>th</sup> instar larva weighing 0.2, 1.2 and 4.3 mg respectively	Prakash & Ponniah, 1978
<i>Cx. raptor</i> (Diptera)	<i>Cx. quinquefasciatus</i>	1 <sup>st</sup> , 2 <sup>nd</sup> instars predators preferred the 1 <sup>st</sup> instar of the prey while instars 3 and 4 preferred prey instars 2 and 3	Thangam & Kathiresan, 1996
<i>Lacconectus punctipennis</i> (Coleoptera)	<i>Ae. albopictus</i>	Larvae could each consume up to 10 1 <sup>st</sup> instar and 10 4 <sup>th</sup> instar prey larvae per day.	Sulaiman & Jeffery, 1986
<i>Ochthera chalybesceus</i> (Diptera)	<i>An. gambiae</i> s.s.	Prey size does not affect predation capacity	Minakawa <i>et al.</i> , 2007
Prey stage abundance	<i>An. stephensi</i> , <i>An. stephensi</i> & <i>Cx. quinquefasciatus</i>	<i>An. stephensi</i> was preferred followed by <i>Cx. quinquefasciatus</i> and <i>Ae. aegypti</i>	Wattal <i>et al.</i> , 1996
	<i>Cx. fatigans</i>	The percentage of prey killed and left unconsumed increased with an increase in prey density	Prakash & Ponniah, 1978
Prey density	<i>Agabus erichsoni</i> & <i>A. opacus</i> (Coleoptera)	At a high density of prey larvae, larvae of all instars of the larger species <i>A. erichsoni</i> had significantly higher consumption rates than the smaller species <i>A. opacus</i> . At a low prey density the differences were smaller and only 3 <sup>rd</sup> instar predators larvae differed significantly.	Nilsson & Soderstrom, 1988
	mosquito larvae	Corixids fed less upon mosquito larvae than the other predators	Washino, 1969
	<i>Abedus indentatus</i> , <i>Belostoma flumineum</i> , <i>Corisella sp.</i> & corixids (Hemiptera)		

Predator species	<i>Aeshna flavifrons</i> , <i>Coenagrion kashmirum</i> , <i>Ischnura forcipata</i> , <i>Rhinocypha ignipennis</i> and <i>Sympetrum durum</i> (Odonata)	<i>Cx. quinquefasciatus</i>	The daily feeding rate varied among the odonate species. The mean number of IV instars <i>Cx. quinquefasciatus</i> larvae killed per day, ranged between 14 and 64 (64 mosquito larvae for <i>I. forcipata</i> , 57 for <i>A. flavifrons</i> , 45 for <i>R. ignipennis</i> , 25 for <i>S. durum</i> and 14 for <i>C. kashmirum</i> ).	Mandal <i>et al.</i> , 2008
	<i>Anisops bouvieri</i> , <i>Diplonychus</i> (= <i>Sphaerodema</i> ) <i>rusticus</i> and <i>Diplonychus annulatus</i> (Hemiptera)	<i>Cx. quinquefasciatus</i>	A single adult of <i>A. bouvieri</i> , <i>D. rusticus</i> and <i>D. annulatus</i> consumed 2-34, 11-87 and 33-122 fourth-instar mosquito larvae per day respectively. The predatory impact (PI) values were 14.77-17.31, 46.9-55.73, and 61.74-72.72 larvae/day for <i>A. bouvieri</i> , <i>D. rusticus</i> , and <i>D. annulatus</i> , respectively while the clearance rate (CR) value range was 9.06-13.25 for <i>A. bouvieri</i> , 13.64-15.99 for <i>D. rusticus</i> , and 13.50-16.52 larvae l/day/predator for <i>D. annulatus</i> . The values of mutual interference constant, "m," remained 0.06-0.78 for <i>A. bouvieri</i> , 0.003-0.25 for <i>D. rusticus</i> , and 0.09-0.27 for <i>D. annulatus</i> , and did not vary between the days. The difference in predatory efficiency, CR, and PI values varied significantly among the three predators, indicating the possible difference in the function as predators occupying the same guild.	Saha <i>et al.</i> , 2007
	<i>Colymbetes paykulli</i> , <i>Ilybius</i> <i>ater</i> & <i>I. fuliginosus</i> (Coleoptera)	<i>Culex</i> mosquitoes & <i>Daphnia</i>	<i>Colymbetes paykulli</i> chosed mosquito larvae more often but, both other predators preferred <i>Daphnia</i> spp.	Lundkvist <i>et al.</i> , 2003
	<i>Diplonychus</i> sp. & <i>Anisops</i> sp. (Hemiptera)	<i>Cx. annulirostris</i>	<i>Diplonychus</i> sp. preyed upon larval and pupal stages of <i>Cx. annulirostris</i> and more efficient than <i>Anisops</i> sp. that fed on larval stage only.	Shaalaa <i>et al.</i> , 2007
	<i>Dytiscus marginalis</i> and <i>Hydrophilus triangularis</i> (Coleoptera).	2 <sup>nd</sup> , 3 <sup>rd</sup> and 4 <sup>th</sup> stage larvae <i>Cx.</i> <i>quinquefasciatus</i>	<i>Dytiscus marginalis</i> was consistently more effective than <i>Hydrophilus triangularis</i> .	Nelson, 1977
	<i>Notonecta unifasciata</i> and <i>Buena scimitra</i> (Hemiptera)	4 <sup>th</sup> instar <i>Cx. pipiens</i> <i>quinquefasciatus</i> larvae	<i>N. unifasciata</i> adults have an overall higher daily killing of than <i>B. scimitra</i>	Stewart & Miura, 1978
	<i>Sphaerodema annulatum</i> and <i>S. rusticum</i> (Hemiptera)	<i>Armigeres subalbatus</i> (different ratios & densities of larvae and pupae)	Both bug species consumed both 4 <sup>th</sup> instar larvae and pupae of <i>Ar. subalbatus</i> in quite good numbers depending on their relative abundance	Aditya <i>et al.</i> , 2005

<i>Sympetrum striolatum</i> and <i>Coenagrion puella</i> (Odonata)	3 <sup>rd</sup> instar larvae of <i>Culex pipiens</i>	The anisopteran <i>Sympetrum striolatum</i> was more voracious than the zygopteran <i>Coenagrion puella</i> .	Onyeka, 1983
<i>Tx. Rutilus</i> & <i>Corethrella appediculata</i> (Diptera)	<i>Ae. Albopictus</i> & <i>Ochlerotatus triseriatus</i>	Overall survivorship of both preys decreased greatly in the presence of the top predator <i>Toxorhynchites</i> whilst the intermediate predator <i>Corethrella</i> increased the survivorship of the native prey species <i>Ochlerotatus</i> and decreased survivorship of the invasive prey species <i>Aedes</i> compared to treatments without predators in artificial analogues of water-filled tree holes	Griswold & Lounibos, 2006
<i>Tx. Splendens</i> & <i>Rhantus silkkimensis</i> (Diptera & Coleoptera)	<i>Cx. quinquefasciatus</i>	Predation rate of <i>Rhantus</i> ranged between 21.56 & 86.89 larvae/ day depending on prey-predator densities. The predator impact (PI) remained between 18.67 & 35.33 larvae/day depending on prey densities, while the clearance rate (CR) ranges between 2.21 & 2.23 larvae litres/day/predator. Comparatively, the <i>Toxorhynchites</i> consumed prey larvae at the rate of 0.67 to 34.22 larvae/ day, depending prey-predator densities. The predator impact (PI) ranges between 7.67 & 11.33 larvae/day, and the clearance rate (CR) ranged between 1.41 & 1.76 larvae litres/day/predator.	Aditya <i>et al.</i> , 2006
<i>Enithares indica</i> (Hemiptera)	<i>An. stephensi</i> & <i>Cx. quinquefasciatus</i>	No marked difference in predation by 1 <sup>st</sup> to 4 <sup>th</sup> nymphal instars of the predator while predation of 5 <sup>th</sup> nymphal instar and adult bugs was noticeably low for <i>Anopheles</i> larvae but quite high for <i>Culex</i> larvae	Wattal <i>et al.</i> , 1996
<i>Orithemis ferruginea</i> (Odonata)	Mosquito larvae	Larger naiads ate more larvae without showing any preference for 1 <sup>st</sup> or 4 <sup>th</sup> instar while smaller naiads preferentially ate 4 <sup>th</sup> instar.	Cordoba & Lee, 1995
<i>Tx. splendens</i> (Diptera)	<i>Ae. aegypti</i> larvae	Second instar larvae of the predator consumed <i>Ae. aegypti</i> larvae significantly at a higher rate than the other instars. Except for 2 <sup>nd</sup> instar predators, other instars showed a significant reduction in attack rate and an increase in handling time	Wattal <i>et al.</i> , 1996

	<i>Ae. aegypti</i> , <i>An. stephensi</i> , <i>Armigeres subalbatus</i> and <i>Cx. quinquefasciatus</i>	The 1st and 3rd instars of <i>Tx. splendens</i> showed lowest and highest predation rate, respectively. Predation rate by any stage of the predator was highest in 1st instar larvae and lowest in 4th instar larvae of all prey species. The variations in consumption rate seemed to be related with the size of the prey larvae offered rather than to the preference for any species.	Pramanik & Raut, 2003
	<i>Acilius sulcatus</i> (Coleoptera)	<i>Cx. quinquefasciatus</i>	Chandra <i>et al.</i> , 2008
Predator prey density	<i>Diplonychus indicus</i> (Hemiptera)	<i>Ae. aegypti</i> and <i>Cx. fatigans</i> (4 larval instars of the preys at varying densities)	Venkatesan & Sivaraman, 1984
	<i>Enallagma civile</i> (Odonata)	<i>Cx. tarsalis</i>	Miura & Takahashi, 1988
Aquatic vegetation	<i>Diplonychus sp.</i> & <i>Anisops sp.</i> (Hemiptera)	<i>Cx. annulirostris</i>	Shaalan <i>et al.</i> , 2007
Temperature	<i>Notonecta hoffmani</i> (Hemiptera)	Mosquito larvae	Murdoch <i>et al.</i> , 1984
	<i>Agabus erichsoni</i> & <i>A. opacus</i> (Coleoptera)	<i>Ae. communis</i>	Nilsson & Soderstrom, 1988
	<i>Aeshna flavifrons</i> , <i>Coenagrion kashmirum</i> , <i>Ischnura forcipata</i> , <i>Rhinocypha ignipennis</i> and <i>Sympetrum durum</i> (Odonata)	<i>Cx. quinquefasciatus</i>	Mandal <i>et al.</i> , 2008

Physical	<i>Acilius sulcatus</i> (Coleoptera)	<i>Cx. quinquefasciatus</i>	Feeding rate decreased with the volume of water.	Chandra <i>et al.</i> , 2008
Foraging area	<i>Cx. raptor</i> (Diptera)	<i>Cx. fatigans</i>	The predation capacity was not influenced by changes in water volume	Prakash & Ponniah, 1978
	<i>Diplonychus sp.</i> & <i>Anisops sp.</i> (Hemiptera)	<i>Cx. annulirostris</i>	Significant effected <i>Anisops</i> capacity but effect of foraging area was pronounced in <i>Diplonychus</i> nymphs only.	Shalan <i>et al.</i> , 2007
	<i>Tx. Splendens</i> (Diptera)	<i>Ae. aegypti</i> larvae	Foraging surface did not influence the predation rate	Analraj & Das, 1998
Water temperature	<i>Tx. splendens</i> (Diptera)	<i>Ae. aegypti</i> larvae	Predation was high at high water temperature however; it did not influence prey handling time.	Analraj & Das, 1998
Water Depth	<i>Ochihera chalybesceens</i> (Diptera)	<i>An. Gambiae</i> s.s.	The predation capacity was not influenced by changes in water depth	Minakawa <i>et al.</i> , 2007
	<i>Aeshna flavifrons</i> , <i>Coenagrion kashmirum</i> , <i>Ischnura forcipata</i> , <i>Rhinocypha ignipennis</i> and <i>Sympetrum durum</i> (Odonata)	<i>Cx. quinquefasciatus</i>	The feeding rates varied significantly between dark and light conditions, in all the odonate species. Darkness had negative influence.	Mandal <i>et al.</i> , 2008
	<i>Acilius sulcatus</i> (Coleoptera)	<i>Cx. quinquefasciatus</i>	Feeding rate of did not differ between the light-on and dark.	Chandra <i>et al.</i> , 2008
Illumination	<i>Brachytron pratense</i>	<i>An. subpictus</i>	The consumption rate was significantly higher during the lights-on phase than during the lights-off phase	Chatterjee <i>et al.</i> , 2007
	<i>Dytiscus marginicollis</i> , <i>Lestes congener</i> ; <i>Notonecta shootrii</i> , <i>Tramea lacerate</i> , <i>T. torosa</i> (Coleoptera, Odonata & Heiptera)	<i>Cs. incidens</i>	Darkness did not significantly affect the predation activity but the odonate naiads, <i>Tramea</i> , have consumed more larvae in darkness than in normal illumination	Lee, 1967



predacious beetle larvae. Contrarily, treatment with parathion 7 days after the *B.t.* H14 application severely reduced the numbers of the beetle larvae. *Cx. pipiens quinquefasciatus* larvae predation was greater when a combination of the hemipteran predator *Buena* sp. and the bacteria *B. t.* var. *israelensis* were present than when each was used separately (Rebollar-Tellez *et al.*, 1994). The predaceous backswimmer *N. irrorata* and the bacterium *B. t.* var. *israelensis* were assessed separately and in combination with each other to suppress mosquitoes on larval population of mosquitoes maintained under experimental field conditions (Barbosa *et al.*, 1997). The combination treatment of both bacterium and predator gave the best result with no harmful effect on the predators. Zero densities of *Ae. aegypti* larvae per dip occurred more frequently in plastic containers treated with both agents than with individual agents. Painter *et al.* (1996) mentioned that repeated applications of *B. t. i.* to the mosquito predator *Erythemis simplicicollis* (Odonata: Libellulidae) from hatching to final instar did not affect development to the adult stage, morphology or maiden flight capability. A 3-year study, 2000-2002, field study for mosquito control with *B. s.* in southeastern Wisconsin revealed that no detrimental effects to nontarget organisms, in particular predaceous insects, could be attributed to this microbial insecticide (Merritt *et al.*, 2005).

Although both *Bti* and *Bs* are safe to other non-target organisms (Mittal, 2003) and recommended as ideal control agents in integrated mosquito control (Lacey, 2007), Collins & Blackwell (2000) reported that, problems have arisen in combining them with some *Toxorhynchites* mosquitoes. Lacey & Dame (1982) showed that fourth instar *Tx. r. rutilus* larvae exposed to 1, 5 and 10 ppm of *Bti* in the presence of excess prey (20 *Ae. aegypti* larvae) responded with 23, 62 and 95% mortality respectively after 10 days. In the presence of excess larvae 98% mortality was observed 10 days after exposure to 0.5 ppm. A positive correlation between concentration of *Bt* (H-14; IPS-78) and mortality was observed in fourth instars

of *Tx. amboinensis* and *Tx. brevipalpis* in the presence of *Ae. aegypti* larvae but *Bs* toxins were lethal only to *Tx. r. rutilus* (Lacey, 1983).

Combinations of insecticides and predators to control mosquito vectors showed a wide range of risk to predators. In some studies there was no or little risk to the predators. Djam & Focks (1983) found that, except for resmethrin, the ED<sub>90</sub> for fenithion, chlorypyrifos, naled and malathion for *Tx. amboinensis* were 1.6 times greater than *Ae. aegypti* and females of the *Toxorhynchites* mosquito were somewhat less susceptible than the males to all of the compounds tested. These results suggest that there is little possibility of applying those insecticides (except resmethrin) at a level sufficient to control *Ae. aegypti* adults without affecting the *Tx. amboinensis* adult population. The relatively short lifespan of *Tx. amboinensis* suggests that the optimal time for insecticide application would be just prior to the release of the predators. In another similar investigation, the concentrations of resmethrin, malathion and naled caused 50% mortality to first instar *Tx. splendens* larvae were 2.87, 69.1 and 623 ppb respectively (Tietze *et al.*, 1993). The integrated treatment using a ground application of ULV-applied malathion and weekly release of the predaceous mosquito *Tx. amboinensis* reduced the *Ae. aegypti* population by 96% compared to 29% for malathion alone during the 14-week study in residential neighbourhoods in New Orleans, Louisiana, USA (Focks *et al.*, 1986). Rawlins & Ragoonansingh (1990) found that predaceous larvae, *Toxorhynchites moctezuma*, from Trinidad were less susceptible to temephos insecticide than *Ae. aegypti* larvae, indicating its possible usefulness in an integrated management program. In a laboratory study Focks (1984) investigated the impact of sublethal exposure on subsequent longevity, fecundity and egg hatch on *Tx. r. rutilus* if the pyrethroid insecticide resmethrin was used without regard to the date of predator release. The exposure of *Tx. r. rutilus* to resmethrin at the LD<sub>90</sub> dose for *Ae. aegypti* reduced neither the adult survival nor egg

hatch. Contrarily, average fecundity was reduced from 5.6 to 2.3 eggs/female/day during the first three or four days of oviposition. Accordingly, the author concluded that; minimizing the reduction in fecundity of *Tx. r. rutilus* in integrating use with resmethrin requires certain adjustments particularly limiting insecticide application prior to predator release. If this practice were followed, only those predators which had already been in the field for several days would be exposed and consequently the effect on fecundity would be minimized. Although previous studies showed no or little risk of insecticides to the predators, other studies showed highest levels of risk. In field study using insecticide to control rice field mosquitoes in California, Schaefer *et al.* (1981) reported that a single application of non-selective toxic agent to rice fields could sufficiently disrupt the predator complex so that resurgence of mosquito larvae populations can continue for a long period. The spraying of the Kenyan rice fields killed both *An. gambiae* and predators (Service, 1977). Moreover, the mosquitoes re-established themselves very quickly but recolonization by the predators was slower. Jebanesan & Vadani (1995) found that an increase in the concentration of the pyrethroid insecticide, K-Othrine, resulted in a decrease in the predation of *Cx. quinquefasciatus* larvae by *Diplonychus indicus*. A reduction in predation was noticed at the highest concentration and was proportional to the interference of the insecticide in the nervous co-ordination of the bug. The application of fipronil and lambda-cyhalothrin insecticides for control of the rice water weevil, *Lissorhoptrus oryzophilus*, in Arkansas rice fields produced deleterious effects on nontarget predaceous insects (Dennett *et al.*, 2003). A marked difference in susceptibility was found between selected nontarget insects. Lambda-cyhalothrin adversely affected populations of nontarget beneficial insects, such as the scavenger beetle *Tropisternus lateralis* and the backswimmer *N. indica*, whereas nontarget pestilent species, such as *Anopheles quadrimaculatus*, proliferated. Contrarily, Fipronil achieved higher

percentages of control against *An. quadrimaculatus* and was less harmful to both nontarget predators.

Unlike the use of combined insecticides and *Bacillus* bacteria, the reported use of combining IGRs with predators in integrated mosquitoes management is rare. Application of Methoprene, Stauffer-20458 and Thompson-Hayward-6040 at 0.025 Ib AI/acre for controlling *Psorophora columbiae* in rice fields caused significant reductions in certain predaceous aquatic insect populations (*Tropisternus* spp. adults and libellulid immatures) while no significant reductions in other predaceous aquatic insects “*Votonecta* spp. adults and immatures, corixid adults and immatures and *Thermodieclus* spp. Adults” occurred at 0.25 Ib AI/acre (Steelman *et al.*, 1975). In another field study evaluating safety and integration of methoprene and predaceous insects, Miura *et al.* (1978a) stated that combined effect of methoprene briquet treatments and the notonectid bugs, *N. unifasciata* and *B. scimitar*, suppressed *Cx. tarsalis* populations in the breeding sites and the treatments did not affect the reproductive, developmental or predatory activities of both predators. Impact of the insect growth regulator hexaflumuron was studied against *Anisops bouvieri* and *Diplonychus rusticus*, which are potential predators of mosquito immatures (Vasuki, 1996). These predators were not susceptible to hexaflumuron at a dose range from 0.0001- 1.0 mg/l and their efficacy did not significantly alter at sublethal concentrations. Other predators (*Ranatra* sp., dragon fly larvae and a cyclopoid copepod, *Mesocyclops leukarti*) also survived at 1.0 mg/l which indicated the safety and utility of hexaflumuron in integrated mosquito management.

It could be concluded that contributions of predators in integrated mosquito control will reduce the percentage of nuisance mosquitoes emergence and in terms of mosquito vectors transmitted disease will also reduce the probability of diseases transmission. The lack of interaction between larvae of mosquito vectors and their natural enemies and/or lower predator survivorship in certain habitats, particularly

shallow water pools and cement tanks (Das *et al.*, 2006) and urban environments such as temporal habitats (Carlson *et al.*, 2004), may cause a sudden increase in mosquito vectors densities and subsequent disease transmission. Furthermore, utilizing predaceous aquatic insects with *Bacillus* bacteria was more successful than combinations of predaceous insects and insecticides in particular against container breeding mosquitoes such as the dengue vector mosquito *Ae. aegypti*. Contrarily, combinations of predaceous insects and insecticides for controlling both rice field and container breeding mosquito vectors are not risk free because some insecticides produce predators' mortalities and predators' re-colonizing is slower than mosquitoes re-establishing. Preliminary results of the IGRs, in particular hexaflumuron, suggest their safety and adaptability in integrated mosquito control.

#### **Difficulties for utilizing predaceous insects for mosquito control**

Although these are successful examples of predators, there are difficulties associated with rearing; colonization and handling which are obstacles to a more widespread utilization of predaceous aquatic insects (Garcia, 1982). The second difficulty is polyphagy that has advantages and disadvantages (Murdoch *et al.*, 1984). An advantage is that these predators can survive when mosquito larvae are rare or absent, while a disadvantage is that they may not reduce mosquito larvae due to availability of alternative preys. The third difficulty is the presence of other invertebrates and vertebrates predators that may reduce the abundance of the predaceous insects (Larson, 1990). The fourth difficulty is predators may interfere through chemical or other cues; for instances the hydrophilid *Tropisternus lateralis* (Resetarits, 2001) and the phantom midge *Chaoborus albatrus* (Petranka & Fakhoury, 1991) avoid laying eggs in pools with fish. The fifth difficulty is the avoidance by mosquitoes of water containing invertebrate predators such as backswimmers and dragonflies and makes predator's impact more complicated.

Additionally, Washburn (1995) pointed out that control of ground pool mosquitoes using biological control agents is more feasible than container breeding mosquitoes due to the following physical and biological features: (1) Natural enemies limit mosquito larvae in ground pools whereas those in containers are limited by resource availability, (2) Containers are smaller than ground pools and lack internal primary productivity, (3) Container habitats support smaller populations of fewer species compared with ground pools, implying that it may be more difficult to establish natural enemies in small container habitats, (4) The lack of primary productivity within containers may limit the number of trophic levels and reduce the likelihood of establishing and maintaining predator population, and (5) Larval mosquito populations in containers are regulated by competitive interactions and mortality from natural enemies is likely to be compensatory.

These habitat and population characteristics, combined with difficulties in locating and treating containers have limited the implementation of biological control agents to suppress mosquitoes developing in water filled containers. Contrarily, Kumar & Hwang (2006) pointed out in their review that only biological control agents such as aquatic predaceous insects carry the potential for overcoming such obstacles and have the ability to adapt to various aquatic bodies including containers. The successful control strategy for container breeding mosquitoes that they pointed in their review was eliminating *Ae. aegypti* populations by introducing dragonfly larvae into domestic containers accommodating *Ae. aegypti* larvae in Myanmar (the experiment was conducted by Sebastian *et al.*, 1990). They have also pointed out that the selection of a biological control agent, mainly predator, in any vector suppression program should be based on: (1) Its self-replicating capacity, (2) Preference for the target mosquito vector population in the presence of alternate natural prey, (3) Adaptability to the introduced environment, and (4) Overall interactions with the indigenous organisms.

## CONCLUSION

In conclusion, predaceous insects are closely associated with mosquito immatures as they cohabit in a wide variety of aquatic habitats such as rice fields, tree holes, man-made ponds, snowmelt pools, temporary lagoons, floodwaters and rain pools. Those predators significantly affect the survival, development and recruitment levels of mosquitoes which most likely has an influence on vector-borne disease transmission rates. Biological and physical conditions were found to influence capacity of such predators. Biological conditions divided into predator and prey factors. Species, competence and predator-prey density were the most common predator factors while species, stage and prey density were more likely prey factors. Illumination, temperature, container size and foraging area were the physical conditions that have been searched. Also field studies and implementation of predaceous aquatic insects in integrated vector control were documented in some circumstances. As can be expected, further studies are needed to ensure successful and satisfactory mosquito control with predaceous insects.

Another important advantage of predators is their released kairomones that have the potency to repel ovipositing female mosquitoes for over a week. If these kairomones were commercially produced, they may provide eco-friendly and effective mosquito control, but more research is necessary to determine total impact. Thus, understanding the interaction between mosquito vectors and their aquatic predaceous insects is imperative for developing and implementing successful biological or integrated control measures that include the use of predators and/or their kairomones.

Utilizing biological organisms to control mosquito larvae is not only eco-friendly, but constitutes a means by which more effective and sustainable control can be achieved. This would be preferable to relying solely upon synthetic insecticides which are not being developed fast enough to combat resistance. As is always the case, the

elimination of aquatic larval stages is a proactive measure whereas control of potentially infective adult mosquitoes is a reactive response necessitated by inadequate management. In this context, predators should be seriously considered for they have the advantage that they can adapt to various water bodies that are enormously scattered around and within human settlements. Once established and effectively auto-reproducing, predators can achieve sustainable mosquito control to a degree that no chemical can hope to aspire.

Finally and likewise Quiroz-Martinez & Rodriguez-Castro (2007), we also recommend certain factors to must be taken into account when considering predaceous insects for mosquito control. These factors include: preference or selectivity of the prey by the predator, species diversity in mosquito breeding site, stability of the aquatic system, larval density, position of the predator in the water column, appropriate number of predators to be released, recovery of the larval population, predator-prey co-evolution, predator-prey synchronization, refuge and community participation.

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