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The most common mosquitoes at Al- Rayyan municipality (Qatar state) and their potential for transmitting malaria.

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ABSTRACT

During the period from December 2014 to April 2015, a mosquito survey (Diptera: Culicidae) was conducted at Al Rayyan Municipality, western region of Qatar. The survey aimed to identify the most common mosquitoes species in the study area and assess their potential in transmitting malaria. In all, 37 collection sites were visited throughout the study period revealing 312 mosquitoes. Larvae were collected as well as adults. The pH of larvae breeding sites was also measured in the laboratory. Also, Several maps have been produced using GIS, showing sampling locations. Species identification was carried out using the available standard keys. The survey revealed the presence of four mosquito species representing three genera: *Culex* 91% (3 species), *Anopheles* 6% (1 species) and *Coquillettidia* 3%. The species encountered were: *Culex vagans* (Wiedemann 1828), *Culex mimeticus* (Noe 1899), *Culex bitaeniorhynchus* (Giles 1901) and *Anopheles stephensi* (Liston 1901). Results showed that the most abundant mosquitoes were *Culex,* although there was no previous record of the new encountered species. Encountered breeding sites were sewage waste water bonds, construction water tanks, animals drinking containers and abundant small containers. Malaria in Qatar is not endemic, however, it is one of the utmost frequently introduced communicable diseases. The presence of malaria vector borne diseases (*Anopheles stephensi*), availability of breeding sites, and the presence of individuals who are infected with malaria protozoans (*Plasmodium* spp.), states the risk of a reintroduction of malaria to Qatar.

ملخص البحث

اجريت هذه الدراسة المسحية بهدف معرفة اجناس(انواع) البعوض الاكثر انتشارا في بلدية الريان بدولة قطر وذلك في الفترة من شهر ديسمبر 2014م الي شهر ابريل 2015 وخلال هذه الدراسة تم جمع العينات من 37 موقعا واشتملت العينات اليرقات بالاضافة الى البعوض البالغ والذي وصل عدده 312 فرد (بعوضة) كما قيست درجة حموضة مياه التوالد في المعمل. وبالاضافة الى ذلك تم تقديم خرائط لتوزيع الانواع المختلفة وذلك باستخدام احد برامج نظم المعلومات الجغرافية .وبناء علي احدث المفاتيح التصنيفية للبعوض تم تعريف ثلاثة اجناس:*Coquillettidia* and *Anopheles* *Culex,* واربعة انواع *Culex vagans Culex* *.bitaeniorhynchus Culex, mimeticus* and *Anopheles stephensi* . كما اظهرت النتائج ان بعوض *Culex*هو الاكثر انتشارا في منطقة الدراسة. ان وجود بعوض *Anopheles stephensi* المدرج كعامل اساسي لمرض الملاريا يمثل خطورة ويجعل دولة قطر عرضة لظهور مرض الملاريا مجددا بها

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1 Introduction

Qatar is aiming for an advanced society through its 2030 vision to become a knowledge base economy skilful of sustaining its development and offering a high customary of living for all of its people. Sound health and environment are considered one of the main areas under consideration for its people and those of the region. Particularly that Qatar will be hosting the world cup tournament in 2022. Research in the field of insect transmitted diseases area, would be in a line with the country's vision. Qatar is one of the nations in the Middle East that excluded indigenous malaria transmission, however, the risk of introduced malaria still occurs due to the big number of immigrant labours from malaria-endemic countries.

Mosquitoes, members of order Diptera, family Culicidae, are widely distributed throughout the world and exploit various water bodies for their breeding (Stone 1977; WHO 1982; Gautam *et.al*. 2006). Their well-developed adaptation mechanisms, enable them to thrive in various environments. They inhabit, impermanent and long-lasting, extremely contaminated and clean, big and little water bodies; even the slightest buildups like flower vases, buckets, tires, leaf axes and hoof prints are potential sources (Norbert *et al.* 2010). Therefore, they are the most abundant group that take place all over temperate and tropical areas of the world, and past the Arctic Circle. These insects comprise of a monophyletic taxon (Wood and Borkent 1989; Miller *et al.* 1997; Harbach and Kitching 1998). They are most likely the most extremely undesirable arthropods, which also known for being annoying stinging pests. At times, their irritation bites are so annoying that they make outside activities practically unbearable in numerous parts of the world. For example, a lot of coastal regions are made intolerable by salt marsh mosquitoes, which in the other hand, seriously affect the real estate expansion and the tourism business.

Mosquitoes known to have complete metamorphosis form of development, which include four stages, eggs, larvae, pupa and adults. The first three stages are aquatic. Suitable water site is very important to ensure a successful development of these stages to the adult formation. Parameters like water quality, occurrence of light, accessible food, present eggs, and vegetation are significant factors in choosing a satisfactory breeding site (Norbert *et al.* 2010).

At the present, about 3,490 species are known (Harbach and Howard 2007). Mosquitoes are commonly, identified either at the field or the lab as (fourth-instar) larvae or adults. Males on the other hand are particularly required to differentiate between various species since the females of many generic-level taxa are extraordinary alike in habitus. While the classification of many seemingly closely linked species is confused by obvious resemblance, bounds of many particular groups are disquieted by morphological variety. Some genera contain various features of unknown similarities that will predictably be documented as independent monophyletic lines when they are carefully studied (Belkin 1962; Judd 1996; Harbach and Kitching 1998; Reinert *et al*. 2004, 2006). All males and the females of numerous species nourish entirely on plant fluids, such as fruit juices, nectar, honeydew etc. Females of many species nourish on the blood of living animals, in order to produce mature eggs, although, some normally may produce eggs without a blood meal. The duration of flight and feeding activity is normally very explicit for utmost species. Some species are energetic at night time (nocturnal) or twilight (crepuscular) while others are energetic throughout the daylight time (diurnal). Regardless of our present understanding of mosquito biology, almost so little is recognized about the detailed bionomics of many species. The provocations or influences that draw mosquitoes to a human or animal hosts are complicated and are not completely implied (Norbert *et al*. 2010).

Mosquitoes are the most significant vectors of human infections and the most severe leechlike arthropods that feed on reptiles, amphibians, mammals and birds. Some species show significant host specificity whereas others have more wide-ranging sense of taste. Over a hundred species of mosquitoes are have the ability for transmitting numerous diseases to human and other animals. These diseases are of concern specially malaria (Plasmodia, i.e., *Plasmodium vivax*, *Plasmodium falciparum, Plasmodium ovale, Plasmodium malariae*), arboviral encephalitides (viruses, i.e., West Nile, dengue, yellow fever, Eastern Equine, Japanese, others) and lymphatic filariases (filarial worms, i.e., *Brugia, Wuchereria*) (Peters 1992).

Anopheles mosquitoes, for instance, alone transfer malaria. It is certainly the utmost severe arthropod vector-borne disease distressing humans. More than 90% of the total malaria deceases in the world happen in Africa. Since most of the cases in Africa are caused by *Plasmodium falciparum*, the most serious of the four human malaria parasites. In addition to that capability of malaria vectors (e.g., *Anopheles* *gambiae* Giles) which are prevalent in Africa and are very hard to govern (Rueda 2008).

Malaria is not prevalent in Qatar, but it is one of the most frequently imported communicable diseases. Since 1970 no indigenous malaria cases have been documented. However as most of gulf countries, imported malaria has been pretty common attributable to increasing immigration from and travel to malaria-endemic countries. Two potential vectors of malaria have been identified by some researchers that is *Anopheles stephensi* and *Anopheles multicolor* Cambouliu (Khan *et al* 2009). Both vectors known as primary and secondly vectors respectfully in the Southwest Asian Region (White 1989 and Zahar 1974).

Presence of such vector, assert the risk of the country to a reintroduction of malaria regardless of the fact that there is no present malaria transmission in Qatar (Khan *et al*. 2009). However, more study in this area is needed to provide and confirm such information.

The aim of this study is to determine the geographical distribution and density of the most dominant mosquitoes in Rayyan municipality, Qatar state and their potential for transmitting malaria. This goal can be achieved throw the following.

Specific objectives:

1. Identify the most common mosquito (genera) in the study area. In addition to the identification of *Anopheles* species.

2. Identify the distribution and density of *Anopheles*, and determine if the *Anopheles* in the study area is a potential carrier of malaria or not.
3. Produce a map using GIS to show mosquito geographical distribution and its habitat including breeding sites by focusing on *Anopheles* species.

We hope the results obtained from this study would be of value to update the knowledge about prevalence and distribution of mosquitoes in the region. Such knowledge will provide the basic information necessary for understanding the possible role of the mosquitoes in the study area in the transmission of human and animal diseases and hence their control.

2 Literature Review

2.1 Mosquito Habitats

Mosquitoes breed in a widespread range of habitats with different types of waters (WHO 1982; Abdull Allah and Merdand 1995 and Gautam *et.al.* 2006), which permit them to colonize different kinds of environments (Rueda 2008). Physical and chemical characteristic of the water are recognized to be specific for several species (Seghal and Pillai 1970) and (Eitam *et al.* 2002), although factors that govern the selection of breeding site by the females to lay their eggs remains mysterious for numerous species (Norbert *et al*. 2010). Parameters like water characteristics, occurrence of light, accessible food, eggs present, and vegetation are critical influences in choosing a satisfactory breeding site. (Norbert *et al.* 2010). Many species breed in both natural and non-natural sites (artificial containers) such as gutters, pools, tree holes, coconut shells, leaf axils, bamboo stumps, septic tanks construction tanks and so on (Mafiana 1989; Aigbodion and Anyiwe 2005).

Knowledge of larval habitats is critically significant in choosing suitable and effective vector control methods and programs (Asha and Aneesh 2014). In short time- flooded zones, lakes or rivers sideways, floodwater Mosquitoes like *Ochlerotatus sticticus* (Meigen 1830) or *Aedes vexans* (Meigen 1830) mature in great quantities and with a flight range of quite a few kilometers, which make them a remarkable annoyance even in areas that faraway from its breeding sites. Eggs of these mosquitoes are laid in seasonal or temporarily flooded zones (Mohrig 1969;

Becker and Ludwig 1981). Other mosquitoes, for example, *Ochlerotatus cataphylla*, (Dyar 1916) *Ochlerotatus cantans,* (Meigen 1818) *Ochlerotatus communis* (De Geer 1776) *Ochlerotatus hexodontus* (Dyar 1916) and *Ochlerotatus punctor,* (Kirby1837) which known as snow-melt mosquitoes, find ultimate breeding site in puddles and pools that are shaped after heavy rainfall or after the snow melts. Brackish or salt water habitats near coastal areas in floodplain favored by halophile's species such as *Ochlerotatus sollicitans* (Walker 1856), *Ochlerotatus taeniorhynchus* (Wiedemann 1821), *Ochlerotatus vigilax* (Skuse 1889), *Ochlerotatus detritus* (Haliday 1833) and *Ochlerotatus caspius* (Pallas 1771). *Anopheles* (Meigen 1818) larvae can typically be found in the presence of other mosquito species in edges of streams, grassy ditches, and in small temporary water collections. Tree-holes habitat favored by arboreal species such as *Aedes cretinus* (Edwards 1921), *Ochlerotatus geniculatus* (Olivier 1791), *Orthopodomyia pulcripalpis* (Rondani 1872) and *Anopheles plumbeus* (Stephens 1828). In addition, some species can breed in even small containers for example *Aedes aegypti* (Linnaeus 1762), *Culex pipiens* (Linnaeus 1758), *Ochlerotatus japonicus* (Theobald 1901) and *Aedes albopictus* (Skuse 1894).

Furthermore, some species have the ability to acclimate to a number of climatic factors or fluctuating environment, for example, tiger mosquito is a tropical species, but it thrived to develop evolutionary adaption in which a female could inhabit a temperate climate and lay special kind of eggs. In long days the female lay eggs that is dissimilar from the one that is been laid in shorter days. This ability helped these species to spread globally through international trade (Madon *et al.* 2002). These are just a few examples of different types of habitats in which mosquitoes can be found, (**Table 1**) is providing more information about mosquitoes and their habitats.

Table 1: Common habitats of the mosquito larvae (Laird 1988).

Habitats*	Examples of mosquitoes	Remarks
Flowing streams	*Culex fuscocephala; Culex gelidusn; Anopheles kochi; Anopheles spp.*	Include creeks, drainage and irrigation
Ponded streams	*Anopheles kochi; Culex annulus; Culex bitaeniorhynchus; Lutzia fuscanus*	Include flooded stream beds, Chlorophyta-rich habitats, polluted ponds
Lake edges	*Anopheles quadrimaculatus; Anopheles pseudopunctipennis; Culex annulirostris; Culex squamosus; Culex annulirostris.*	Margins of lakes
Swamps and marshes	*Aedes longirostris; Anopheles kochi; Anopheles sinensis; Culex gelidus; Culex tritaeniorhynchus; Mansonia uniformis; Mimomyia chamberlaini.*	Include fishponds, duckweed ponds
Shallow temporary pools	*Aedes communis; Aedes excrucians; Aedes hexodontus; Anopheles dirus.*	Include snowmelt pools
Natural containers (plant origin)	*Anopheles spp; Culex spp; Ficalbia spp; Haemagogus sp; Orthopodomyia spp; Sabethes spp; Toxorhynchites spp; Tripteroides spp; Uranotaenia spp; Wyeomyia spp.*	Include tree holes, internodes, leaf axils, flower bracts, fronds, nuts, pods, pitchers.
Natural containers (animal and other origins)	*Anopheles spp; Culex spp; Culiseta spp; Deinocerites spp; Eretmapodites spp; Uranotaenia spp.*	Include shells of snails, clams, arboreal ant nests, crab holes

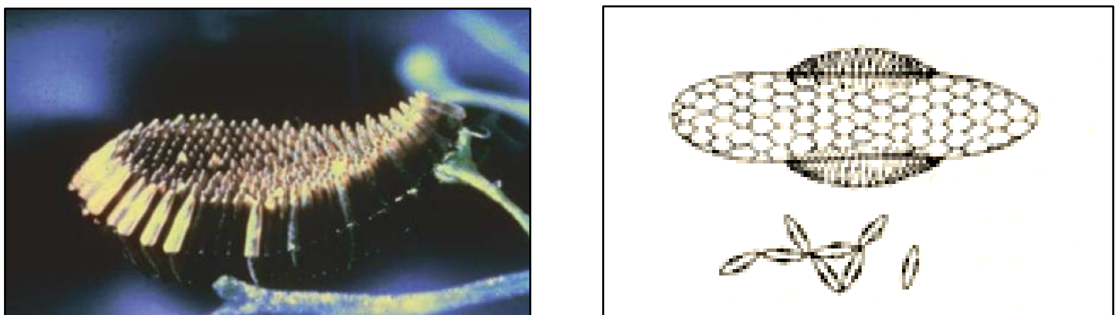
2.2 Biology of Mosquitoes

2.2.1 Mosquitoes Life Cycle

Mosquitos' life cycle described as a complete metamorphosis form of development. It includes four stages; namely, egg, larva, pupa, and adult. Larvae and pupa (second and third stages) need certain environment with flowing or standing water for successful development. The following is a description of all four stages.

2.2.1.1 Mosquito Eggs

After a full blood-meal, female mosquitoes in general lay between 50 and 500 eggs, during 2 to 4 days (this can be longer in cool temperate climates). Mosquitoes can be parted into binary groups, according to their egg-laying mechanisms. Females from first group, lay their eggs individually (Figure 1) on the water surface (*Anopheles*) or in bunches (*e.g. Uranotaenia, Culex, Orthopodomyia, Coquillettidia,* and subgenus *Culiseta* (Figure ***1***). First group embryos do not go into dormancy and hatch as soon as the embryonic development is done. This group typically has quite a few generations each year. The number of generation produced each year depend on several factors, breeding season length, biotic and abiotic circumstances, but temperature has the most significant effects on the speed of development. The second group, need to find the appropriate site to lay their egg. This is crucial to guarantee successful progress of the undeveloped stages. Clements described the mosquito embryogenesis in detail (Clements 1992). According to him the development of the embryonic start almost instantly after the eggs have been laid. The time needed for the eggs to hatch depend on the temperature, usually it take around two to seven days. *Culex pipiens* larvae, take one day to hatch after the eggs have been laid at a temperature of 30°C, three day to hatch at 20°C and ten days to hatch at 10°C, the larvae cannot be developed at 4°C (Figure 1).



Egg raft of *Culiseta annulata* (size approx. 5 mm) | *Anopheles* laid her eggs singly

Figure 1: Mosquito Eggs. (Norbert *et al.* 2010).

2.2.1.2 Mosquito Larvae

Larvae have an apodous body (legless) which divided into three discrete parts: The head which contains the eyes, the mouth fragments and the antennae; the thorax and the abdomen. In order to regulate the electrolyte level the abdomen poised three modified posterior segments that contain four anal papillae to control it. For oxygen intake a siphon is used in culicine or as in anopheline only spiracular lobes. For this reason, culicine larvae dangle downward from the water surface (Figure 2) while larvae of anopheline lie parallel to the surface of the water (Figure 2) (Norbert *et al*. 2010). Palmate setae, specialized setae, are responsible for holding the anopheline larvae in the horizontal position. However, larvae of some species as in *Mansonia* and *Coquillettidia* live underwater, hence, they have specialized siphon which is altered for penetrating under water parts of aquatic plant to get oxygen from the aerenchyma (Figure 2). Hanging downward while attached to the plant these larvae filter water for food, therefore will not be seen by predators) (Norbert *et al.* 2010). Larvae feed on microorganisms, protozoa, algae, detritus and other sort of organic materials. They can be classified as suspension or filter feeders, predators or browsers (Dahl *et al*. 1988). Larvae of some species nourish on other mosquito's larvae (e.g., *Toxorhychites, Lutzia*). However, first instar of some predator's species is filter feeder. In addition, mosquitoes' larvae are an essential source of food for some birds (Cameron *et al.* 2007) Therefor mosquitos' larvae play an important role in natural breeding habitats food chain. Larvae are very sensitive to any interference to the water, therefore they dive down for a little while when they feel any

(**a**) Larvae of *Aedes vexans*

(**b**) Larva of *Anopheles plumbeus*

(**c**) Larva of *Coquillettidia richiardii* attached to plant tissue (photo, Hollatz)

Figure 2: Mosquito Larvae (Norbert et al. 2010)

Depending on water temperature larvae of most species can go on feeding for one to three weeks or more before developing to the next stage. There are big variances in the most favorable temperature for the growth of various mosquito species for example, the snow-melt mosquitoes can develop successfully at temperatures as low as 10°C, while they are unable of complete their development at temperatures beyond 25°C.

2.2.1.3 Pupa

Pupae are the third aquatic stage following the larvae; usually this stage last in about two days, yet, this period can be extended or reduced at lower or higher temperatures respectively. Larva fat body is used as a source of vitellogenines after transferred to the adult stage. Pupae float immobile at the water surface when at rest, but it dives down into the water when disturbed. Unlike other insect, pupae of mosquitoes are pretty mobile Comparing to larvae, which swims energetically to the water surface, the pupa drifts inactively again to the surface after diving. Most mosquitos' pupae are quite tolerant of dehydration, and adults can develop well even if breeding sites have practically dried or pupae have been stranded. Moreover, pupae do not feed unlike larvae. Pupae of *Mansonia* and *Coquillettidia*, take the oxygen from the aerenchyma of the underwater parts of the plants (Norbert *et al.* 2010).

2.2.1.4 Moaquito Adults

Adult mosquitoes deliver food for wide variety of terrestrial birds, invertebrates, mammals, amphibians and reptiles, in addition to its role in the pollination of some plants (Cameron 2007). The final phase of transformation is completed when adult gradually develop from the pupal skin. To escape dropping onto the water surface, the emerging adult moves carefully. Newly emerged adults are very vulnerable to predators and strong winds. In the case of genus *Coquillettidia* because the pupa is attached to under water parts of the plant, they have to float to the surface of the water. In order to do that the pupa must detach its trumpets from the plant, float to the surface before the emergence (Mohrig 1969). Most of mosquito species (*Culex, Uranotaenia, Culiseta and Anopheles*) hibernate as adult females. They pursue overwintering housings throughout autumn and move out of these spots when the temperatures rise in spring. Commonly, females of these species utilize the left over larval fat body and nourish regorously on plant juices throughout autumn females of some species within the *Anopheles maculipennis* Complex can take infrequent blood-meals through winter to endure the extensive times of starvation (Clements 1992). After emergence the males need one to two day to be sexually mature, for that reason they emerge one day before females.

2.2.2 Feeding

Mosquitos' feedin-parts are well developed for piercing the skin and sucking blood of a host. Males feed on flower nectar to obtain carbohydrates, while the female mouthparts are well established to penetrate the skin of the host to attain blood for egg development (Magnarelli 1979 and Clements 1992). Female mosquitoes punctured the skin of the host, and start sucking the blood. Sucking organs, help to pump the blood or the nectar into the guts. In order for a female mosquito to take a blood-meal, the blood must remain in a liquid form to prevent clotting. Therefore, mosquito's saliva, which contains anticoagulants, is injected into the wound (Parker and Mant 1979). This action commonly stimulates resistant response that lead to inflammatory reaction at the site of the wound of the host. Temperature and

Thickness of the skin are most likely essential probing provocations for mosquitoes, as skin's surface temperature is linked to the quantity of blood veins in the skin (Davis and Sokolova 1975). Mosquito's males and females need plant fluids as an drive source, commonly for flight. Plant honeys like honeydew and floral nectar, are the most drive source used throughout the adult life of both sexes (Briegel 1973). However, mosquitoes vary in their feeding and relaxing behavior. Species which rather nourish indoors are called endophagic (endophagy), while others which nourish largely outdoors, are known as exophagic (exophagy).

2.2.3 Host Seeking

Oogenesis can only be completed in many mosquito species when the females take a blood-meal. Consequently, they have adapted a complicated host-seeking behavior to detect possible host. Mainly, three stimuli are used visual thermal and olfactory. Females have many antennal receptors that react to host odors. Lactic acid, carbon dioxide, octenol, acetone, phenolic and butanone compounds are the main olfactory provocations. The host-seeking process may vary among species since it rely on the availability of certain hosts and season (Sutcliffe 1987). Most mosquitos' females need to take blood meal in order to lay mature eggs. Therefore to locate a potential host they developed a complicated host seeking behavior. These host seeking mechanisms built on visual olfactory and thermal stumuli. Female mosquitoes, have a unique antennal receptors that respond to host odors. Carbon dioxide is the key olfactory stimuli in addition to lactic acid, acetone, octenol phenolic and butanone. Different species may have different host seeking behavior, depending on the potential host and the season.

According to a field study two types of dispersal mechanism are assisting the host seeking procedure, horizontal and vertical dispersal. Some species found on trap at 4m height from the ground whereas others found on 10 m height. There is a relation between the mosquitos' distribution and the appropriate host. Mosquito is quick to respond to the host odor, it can use the odor from more than 20 m to track down the host. The release of carbon dioxide as well as other stimuli provokes behavioral reactions. Mosquitoes are very sensitive to any minor variations in carbon dioxide concentrations. The receptors display responses to fluctuations as sm all as 0.01% (Kellogg 1970).

Moreover, other constituents of host odor and breath found to be a stimuli and can be detect by female mosquitoes antennal receptors when attended by carbon dioxide. For example, lactic acid can be a triggering and adjusting stimulus for mosquitoes, if carbon dioxide is also exists in the air (Smith *et al*. 1970; Price *et al*. 1979). These mechanisms are the results of evolution development between the insect and the target host (Murlis 1986). The compound eyes assist to distinguish between light concentration, movement, divergence and color. Mosquitoes react mostly to black, blue and red colors, while the least desirability is caused by yellow and white (Lehane 1991).

2.2.4 Survival during Dry Seasons and Hibernation

In tropical areas, survival mechanisms during dry season, especially for the major vectors of malaria such as *Anopheles gambiae* Giles, is the most disturbing part in understanding the biology of mosquitoes. Two survival approaches could be detected: constant reproduction all the way through the year and dormancy of embryo in wet soil for few days (Minakawa *et al.* 2001). In temperate regions mosquitoes have developed effective mechanism to egg, larvae and adults in order to overwinter the dry season. However, some species can overwinter the dry season in more than on stage for example *Ochlerotatus rusticus* Rossi and *Culiseta morsitans* Theobald. Several elements, especially hydrological conditions and the latitude (drought and cold) are very important in determining the period of hibernation which can vary even within one species.

2.3 Medical Importance of Mosquitoes

Mosquitoes are accountable for the communication of a lot of medically important parasites and pathogens (i.e. nematodes, bacteria, and viruses) which cause serious illness such as malaria, dengue, Chikungunya and yellow fever, filariasis or encephalitis (Kettle 1995; Beaty and Marquardt 1996; Lehane 1991; Eldridge and Edman 2000). Transmission mechanism can be biological or mechanical. The biological mechanism is more complicated since it require time for duplication and growth of the pathogen or parasite inside the vector. Because of their feeding behavior, mosquitoes can transfer pathogens or parasites from one vertebrate host to another, if other factors such as mosquito's ecology and physiology is fitting for transmission. Competent vectors have to be thoroughly connected with the hosts and their durability has to be adequate enough to permit the pathogens/parasites to multiply and/or to mature to the transferrable stages in the vector. Successful transmission aquire several blood-meals. Mosquitoes are the most dangerous insects threatening human. Over three billion people, in the tropical and subtropical regions are threaten by mosquitoes, inaddition, they have noticeably affected the development of mankind economically and politically. Certainly, these diseases in the past have weakening empirs, and affected its development. For, e.g. malaria was one of the main health problem that led to the fall of Roman Empire. (Bruce-Chwatt and de Zulueta 1980).

2.3.1 Malaria

Malaria which caused by protozoans (*Plasmodium* spp.), remains the most significant vector-borne disease. Over 100 tropical countries are affected by it. In 2006, about 3.3 billion people (over 40% of the world population) were threatened. The disease is accountable for about 300 million case and over one million deceases yearly, mostly within children under five years, over 90% of them live in the tropical of Africa (WHO 1997 c). Malaria is a major socio–economic problem due to the massive fatalities, lost days of labor, the expenses for treatment of patients, and the adverse effect of the disease on development. The twelve-monthly expenses of malaria in Africa were about nearly two billion US$ (WHO 1993, 2008).

Four species of the genus *Plasmodium* (*P. falciparum*, *P. vivax*, *P. ovale* and *P. malariae*) are known to cause human malaria. Only anopheline mosquiroes are responsible for the transmition of the causative agent of human malaria. Around 400 species of *Anopheles* are known worldwide, 40 species of these are significant vectors of human malaria. In Sub-Saharan Africa, *Anopheles gambiae* Complex, are the most vital vectors and the most effective malaria vectors around the world. The most dangerous from this complex are *Anopheles arabiensis* (species B and A). Both are the most severe vectors of *P. falciparum* (the most severe malarial pathogen) in Africa. Because of their anthropophilic behavior and physiological probabilities, their ability as a vector is higher than other closely related sibling species. Speciously, vector (mosquito) and pathogen/parasite, both are either reformed to each other throughout the course of evolution, causing a co-occurrence, or the pathogen/parasite was resisted. Identification of vector populations is very difficult due the phenotypic and genotypic plasticity, which effect the implementation of surveillance and control/management strategies. But, PCR – technique, allows researchers to use explicit genetic markers to differentiate sibling species from each other, and also in the examinations of well-defined populations. Around 20 *Plasmodium* species found in other primates, about the same quantity found in other mammals, and nearly 40 each in reptiles and birds (Garnham 1980, 1988).

The *Plasmodium* species have a multipart replication, consist of asexual replication in vertebrates' and sexual replication in mosquitoes. After a blood meal from infected vertebrates the mosquitoes will ingest the sexual form of the parasite, which will get into the mosquito gut and develops into a motile ookinete. Move to settle outside of the midgut epithelium, and make an oocyst. Meiotic and succeeding mitotic divisions will follow and produce many habloid, which erupt the wall of the oocyst and move over the haemocele and gather in the salivary glands. At this point the diseased mosquito is capable of inserting the sporozoites with saliva into the following host. After this primary replication, the parasite endures asexual multiplication in the erythrocytes. Grown schizont, yields a small quantity of new merozoites (Garnham 1966). Which burst into the blood stream and invade new erythrocytes and repeat the cycle. This discharge of merozoites causes the fever attack along with other clinical symptoms. The interval between the fever attacks determined by the length of the schizogonic cycle. When infected with *Plasmodium falciparum and P. ovale* the fever attack after 48 h, while infected with *P. malariae* the fever will attack after 72 h (Kettle 1995) (Figure 3). After a few cycle, some trophozoites produce gametocytes instead of merozoites which have to be consumed by anopheline mosquitoes to complete the cycle of development. Malignant malaria is the most serious type in human, resulting in life- threatening malaria. It is a common cause of mortality in children, and cause mortality in 25% on non- immune adults (Wernsdorfer 1980).

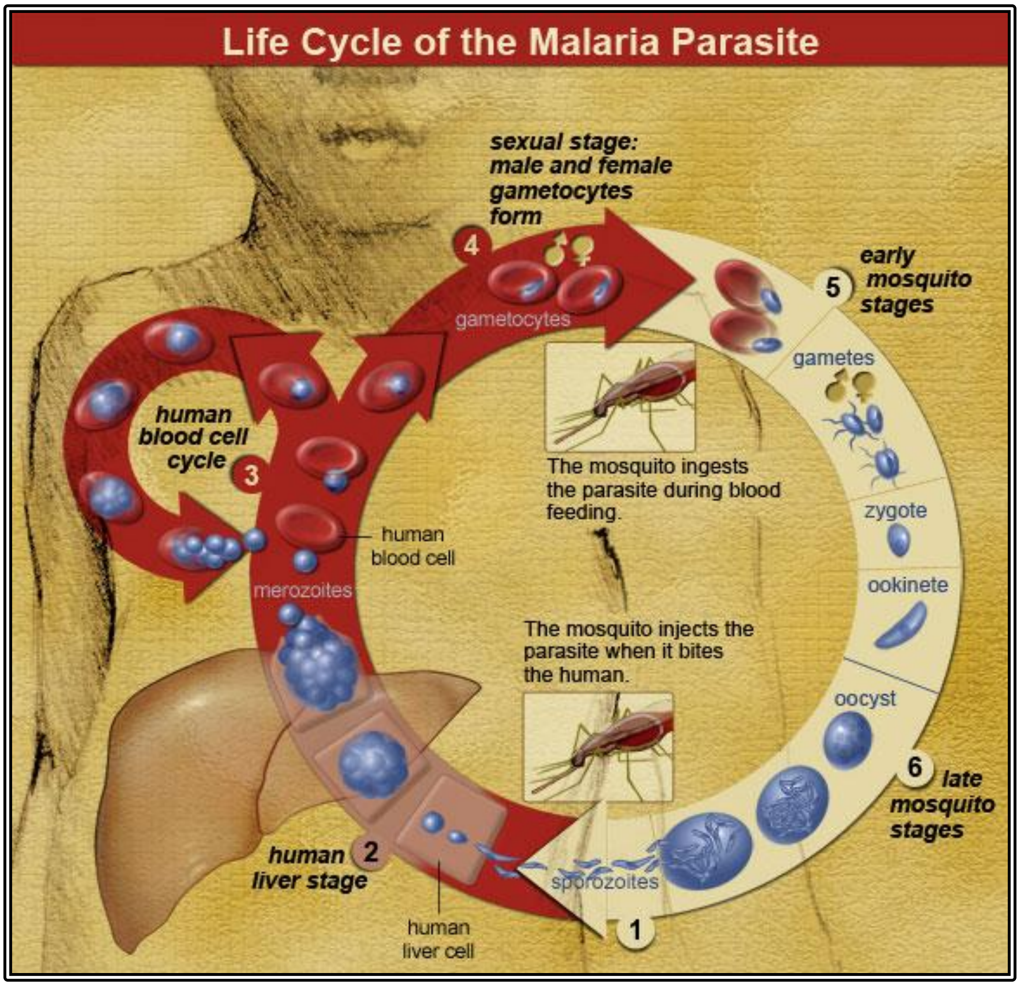


Figure 3: Life Cycle of the Malaria Parasite (National Institute of Health 2012).

Deficiency of proper infrastructure, resource shortages, lack of knowledge and training, resistance to chloroquine and other antimalarial drugs, environmental limitations and usage of pesticides, all together inhibited the progress in malaria prevention and control particularly in Africa for a lot of years (WHO 2000, 2008; Etang *et al*. 2004 and Mukabana *et al*. 2006). Roll Back Malaria (RBM) program, one of the international campaigns against malaria, directed by main international organizations, target to lessen these long-standing weaknesses (Makundi *et al.* 2007; Mboera *et a*l. 2007; Protopopoff *et al*. 2007 a. b; RBM 2005). Control of mosquito populations in their breeding sites provide further chance to considerably improve the protection provided by current vector management approaches and resulting in malaria decline (Killeen *et al.* 2000a,b; Fillinger and Lindsay 2006; Dongus *et al*. 2007; Walker and Lynch 2007).

Even in Northern Europe, it is well recorded that malaria was a threat to human life (Marchant *et al.* 1998). Mainly, *Plasmodium vivax* and *P. falciparum.* Found in Europe, the first one happened throughout the continent while the other one limited to the Southern Europe (Jetten and Takken 1994). In the present time, *Plasmodium vivax* rarely causes a deadly disease, which proposes that *P. vivax* has developed into a reduced virulence over the previous century (Kettle 1995). Malaria was gradually wiped out from Europe right after War II. This was largely because of the decrease in natural breeding habitats through developed agricultural methods, upgraded socio-economic settings and improved hygienic practices. The latest reported incident of indigenous malaria in continental Europe was in Greek Macedonia in 1975 (Bruce-Chwatt *et al.* 1975).

Rising slope in international travel has further added to the cmplecity of this problem, not only diseased people import the parasite, but also infected anopheline may be transported by aircraft from one continent to another. Which make them a threat to people that live close to the international airport. Over 60 cases of "airport malaria" occurred in Europe since 1963. For example, in 1994, seven people were reported to with *Plasmodium falciparum* infection in the vicinity of the Charles de Gaulle airport in Paris alone, even though they had never been to the tropics. Norbert *et al*. 2010).

In Central and South America, malaria has been a main problem regardless of current advances, and remains as a vital hazard. Through the region, a number of anopheline species are elaborate in transmission. *Anopheles albimanus* Wiedemman is very common, taking place from the southern borderes of the USA in the north, to Northern Peru borders in the south, which include the Caribbean region. It is a tropical lowland species, existing most generally on coastal areas and along waterways. *Anopheles darling* Root is also an essential vector, originate mostly in forested areas, and spreading from Mexico to Argentina, and from the Atlantic to the Pacific coasts. Also, other species such as *Anopheles aquasalis* Curry, *Anopheles albitarsis* Arribalzaga, and *Anopheles pseudopunctipennis* Theobald are important. The most common parasite in the Americas, is *Plasmodium vivax* causing about 75% of cases. The rest is caused by *Plasmodium falciparum*, however, *P. malariae* also taking place in restricted areas. Malaria control has confronted a range of challenges including budgetary issues, insecticide resistance, human migration and the emergence of urban malaria. However, a descending pattern in malaria cases from 1.1 million in 2,000, to ~770,000 in 2007 (PAHO).

In Australia, malaria previously happened infrequently in the northern areas. In 1934 an unexpected malaria outbreak caused by *Plasmodium falciparum* ocuured at Fitzroy Crossing (Western Australia) lead to 165 deaths, however, by 1981 the disease was eliminated. The vectors initially accountable for transmission are not known, but *Anopheles farauti* Laveran was possible to have been significant vector, with other species such as *Anopheles amictus*, *Anopheles bancroftii* (Giles 1902) and *Anopheles hilli* (Woodhil and Lee 1944) also playing a role. Presently, around 700–800 malaria cases are imported every year, and low number of locally transmitted malaria as well (Norbert *et al*. 2010).

Sub-Saharan Africa is the most difficult and challenging malarious region. Political unsteadiness, military movement, relocated human populations, urbanization and poverty, all together add to the situation. In some previous malaria-free areas in East Africa, cases are presently being reported, which might suggest a climate change effect. Most of the population, about 93% lives in areas with endemic malaria or in areas at risk of epidemics hence, around 270 million cases occurred yearly. Therefore, fatality rise to up to 95% of the worldwide malaria deaths in this continent. The main vectors are *Anopheles gambiae* (Gilels 1902), *Anopheles arabiensis* (Patton1905) and *Anopheles funestus* Giles, with *Anopheles pharoensis* Theobald also important in some areas. The *Anopheles gambiae* Complex (Gilels 1902) comprises some of the most effective malaria vectors (anthropophilic and endophilic) and take place in both urban and rural areas. *Plasmodium falciparum* is still the most important parasite, with *P. malariae* and *P. ovale* also taking place. *Plasmodium vivax* occurs in East and Southern Africa, but not reported in West Africa (Norbert *et al.* 2010).

Asia include a wide range of areas, with great variety of habitat, climate, and socio-economic rank. Vector importance and parasite occurrence is varied too. Main vectors in Central and Western and Asia comprise *Anopheles sacharovi* Favre, *Anopheles stephensi* Liston, *Anopheles culicifacies* Giles, *Anopheles fluviatilis* James, and *Anopheles superpictus* Grassi. In the Far East tropical, an inclusive variety of important species take place, including *Anopheles dirus* Peyton and Harrison, and *Anopheles minimus* Harbach and Mangui. This collection inhabits a wide-ranging of habitats. *Plasmodium falciparum*, *P. malariae* and *P. vivax*, all take place in this continent, with the quantity of *P. falciparum* growing in South East Asia (WHO 2000). ~2.5 million cases were reported yearly, with the majority taking place in India, with ~4% of the world malaria mortality occuring in this region. Number of cases has been decreasing since 1996. As in other parts of the world, military confilect and human relocation add to the malaria problems (Norbert *et al.* 2010).

2.3.1.1 Malaria in Qatar and Gulf Countries

Malaria is not endemic in Qatar, but it is one of the most regularly imported infectious diseases; it has been terminated from the country since 1970. However, the country is at risk of such diseases and necessitates full-bodied surveillance and preparedness to address any potential outbreaks, such risk is mostly due to the increase in arrival of foreign workers from endemic countries, in addition to the five-fold increase in the number of international arriving tourists between 1995 and 2009, reaching 1,650,000 visitors. In the present, The State is sustaining the zero reporting of indigenous malaria cases, while imported malaria is prevalent (Figure 4).

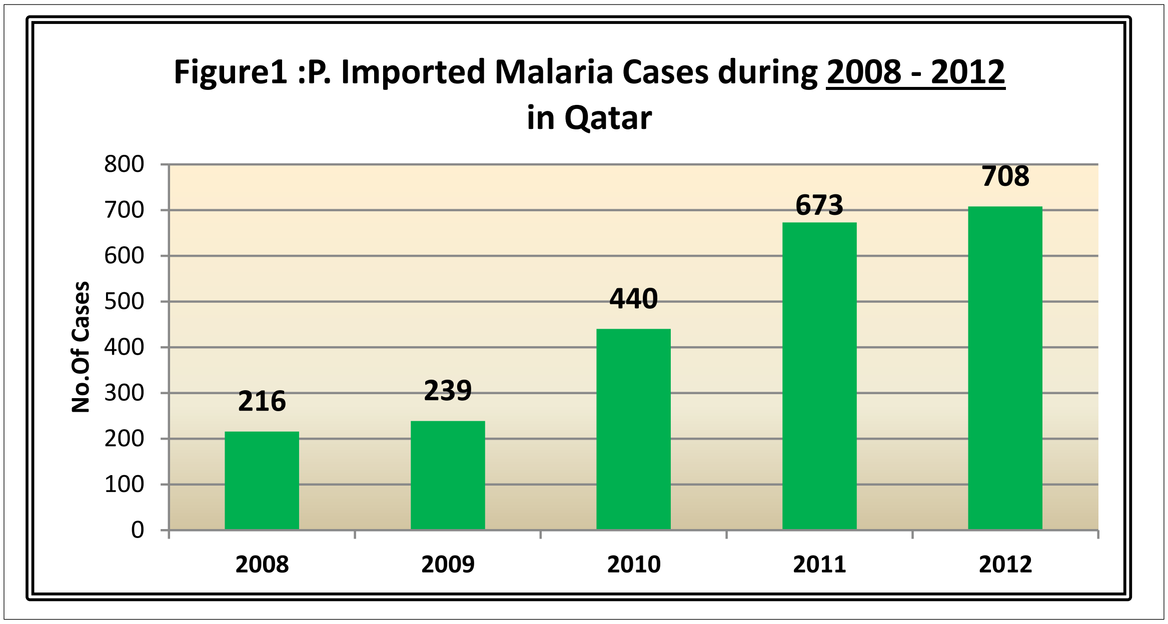


Figure 4: Imported malaria cases during 2008 – 2012 (Supreme Counsil of Health 2013).

All indigenous cases of malaria were reported for the last few years are attributed to importation by labor force, recruited on the large scale particularly from the countries of Indian subcontinent (India, Bangladesh, Pakistan, Sri Lanka and Nepal), Western Pacific (Philippines), add to that imported cases from Sub-Saharan Africa, including Tanzania, Sudan, Tanzania, and Zanzibar (Mikhail *et al.* 2009). However, only limited malaria cases are being imported by the residents (Qatari or Non Qatari), returning from malaria endemic areas, being there either on business trip, or on vacations. A one year study shows that 81 cases of malaria has been confirmed according to Hamad Medical Corporation (Figure 5) 64 (79%) males and 17 (21%) females. Among qatari residents four cases have been reported while in non-Qatari residents 45 cases were reported, in addition to 32 cases within newly immigrants and visitors.

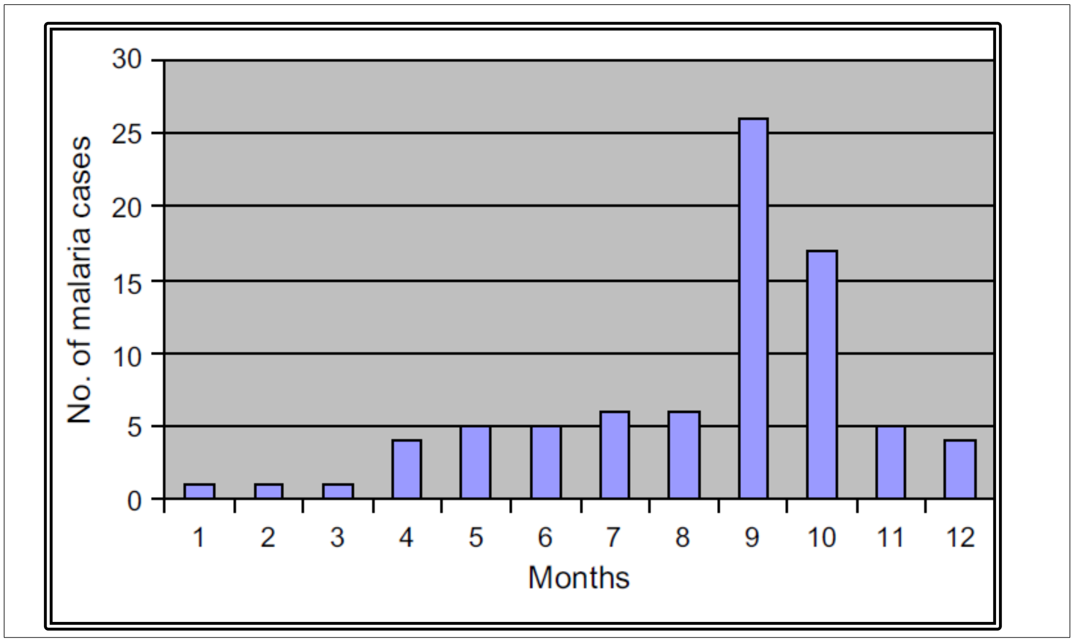


Figure 5: Number of malaria cases in 12 months (Supreme Counsil of Health 2013).

Plasmodium vivax was found to be the cause for most cases than *P. falciparum* with the majority brought from the Indian subcontinent where *P. vivax* is the main malaria species. However, the prevalence of *Plasmodium vivax* can be clarified somewhat by the arrival of huge number of workers during the years 2004 and 2005 from India, Nepal and Pakistan to assistance the construction of the sport city which for Asian games that held in Qatar in 2006. The inflow of such workers alters the demographic features of Qatar considerably (Al Tawfig 2006). Likewise, numerous reports from Saudi Arabia, United States and some European countries (WHO 2000 and Al Tawifiq 2006), showed that most patients that coming from

India and Pakistan had a majority of *Plasmodium vivax* while Patients with *Plasmodium falciparum* infections were most likely had come from or visited Africa.

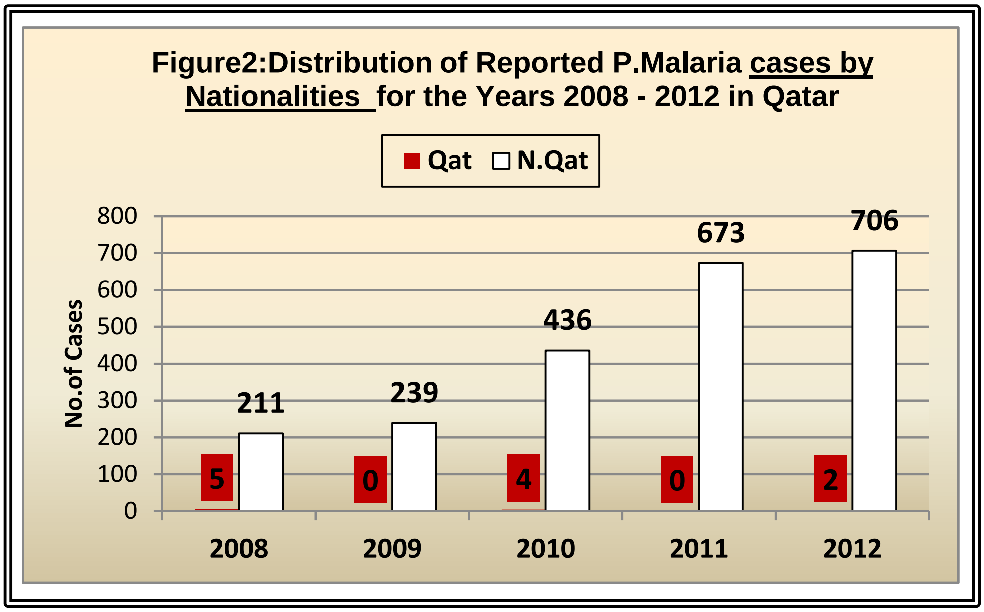


Figure 6: Distribution of Reported *Plasmodium malaria* cases by Nationalities (Supreme Counsil of Health 2013).

Distribution of malaria cases by nationality was Indians (48.5%), %), Pakistani 30.5%, Sudanese (3%), Nepali (14%), in 2012 two Qatari encountered the disease after traveling to endemic countries (Figure 6). Some reports (Jensenius *et al.* 1999) found that some factors are linked to the expansion of complicated falciparum disease such as age, doctor's delay, non – immunity and total diagnosis delay. Sever cases of falciparum malaria was more found among Qatari than non-Qatari patients (50% vs. 5%). This can be explained due to lack of immunity for malaria. Two possible vectors of malaria have been recognized in Qatar that is *Anopheles stephensi* Liston and *Anopheles multicolor* (WHO) presence of such victors increase the risk of malaria transmission in Qatar dispite the fact that there no active malaria in the country. Thus, the epidemiological surveillance system of the country must be alert to report and investigate all malaria cases (Khan 2009).

In 2005 the number of imported malaria cases reported by who in the Gulf countries, were 1054 in Saudi Arabia, 1544 in the United Arab Emirates, 544 in Oman, 302 in Kuwait and 71 in Bahrain. In general, all the Gulf countries have very high accessibility and liability with regard to malaria, since they receive constantly big numbers of imported malaria cases among the enormous arrival of labor force coming from areas as mentioned regarding Qatar, such as India, Pakistan, Bangladesh, Nepal, Sri Lanka and Philippines (Khan 2009).

2.4 Morphological and Taxonomic Techniques

2.4.1 Morphology and Systematics

Mosquitoes belong to the order Diptera (flies), sub-order Nematocera and family Culicidae. There are three subfamilies which can be distinguished from each other by unique characteristics,Figure 7) sumarise some of these characteristics (Jupp 2004).

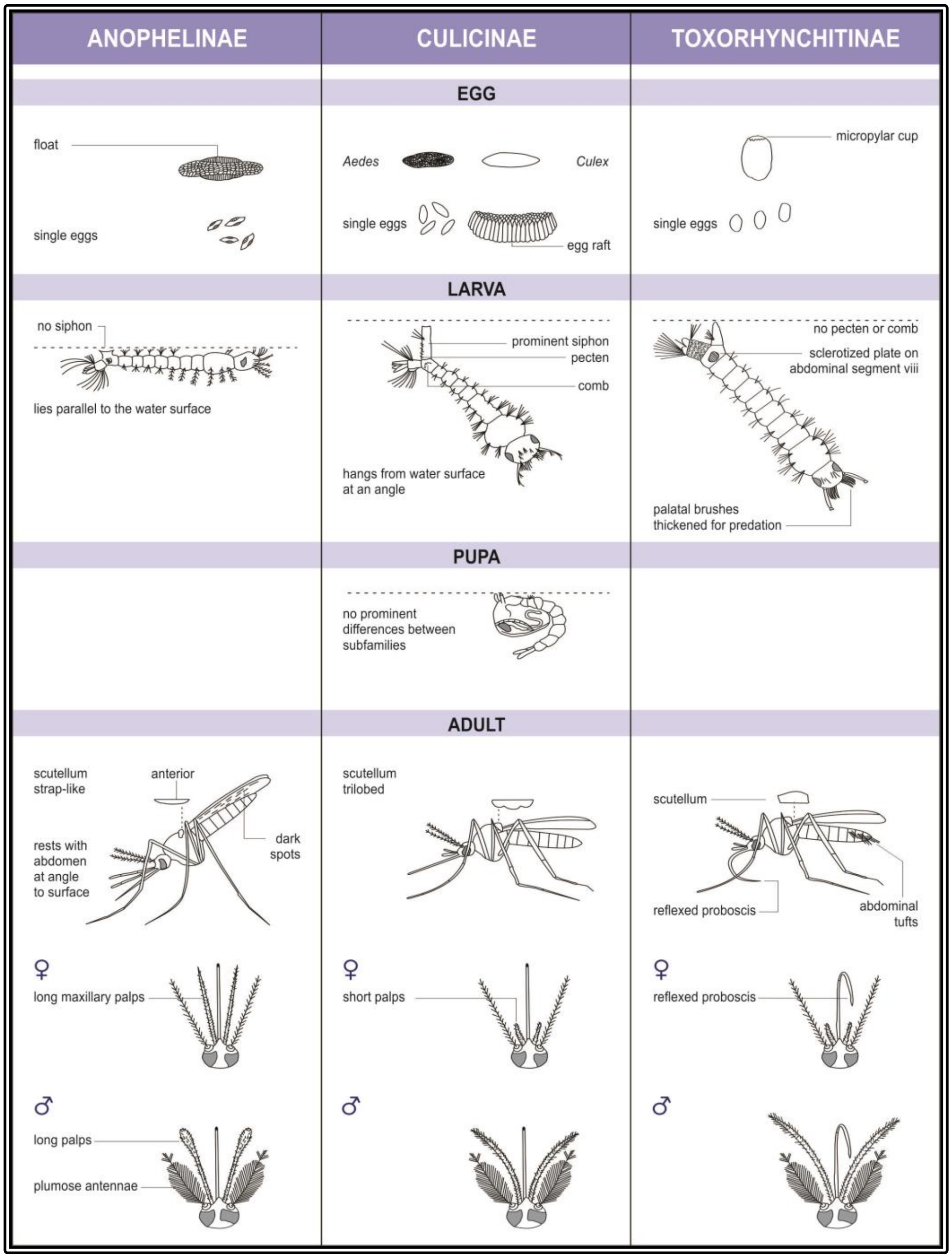


Figure 7: Separation of the different life stages of subfamilies Anophelinae, Culicinae and Oxorhynchitinae (Jupp 2013).

2.4.2 Morphology of Mosquitoes

2.4.2.1 Adults

The long scaled proboscis (labium and stylets) is what differentiate mosquitoes from other Nematocera members , longer than thorax and laying forward along with maxillary palps (Figure 8). Most mosquitoes males and the females of genus *Anopheles* have palps that are the same length or longer than the proboscis. Scales and setae are covering the three parts of the body. The wing margins, wing veins and legs, are usually covered with scales too. The families of slender have the closest similarity of the body shape, nonbiting midges (Chironomidae) and the long-legged crane flies (Tipulidae)), the first one are frequently being incorrectly thought of as mosquitoes, however, these families do not have mouthparts for piercing and sucking.

Visible unique features are what differentiat males of most mosquito species from females for example presence of long, hairy maxillary palps and plumose antennae. The males antenna have thick, long setae all along the first 12 flagellomeres unlike the last flagellomere which has shorter setae. Abnormally for Diptera, the mosquito integument is rather widely shielded with scales. Basically, scales are compacted setae comprising pigment and frequently have a striate external, and that what gives mosquitoes the physical or physicochemical coloration. Tropical species famous for its metallic green, blue and purple look (*e.g. Sabethes* spp., *Haemagogus* spp.). However, the scales' color of most other species may fluctuate from white to nearly black but is commonly mentioned as pale or dark when descriping species.

Both pale and dark scales may found combined together. They can also be assembled together, creating the particular contrasting arrangements, that will be stated to as stripes on the scutum, rings on all legs, and bands on the abdominal terga. Scale coloration can transform in dissimilar light sources and when stored for long period. The whole body color can be inclined by the color of the integument which can glow all over the scales. In the Culicinae subfamily, the abdominal terga and sterna are heavily concealed by scales, whereas the sterna, and commonly the terga, are entirely or mostly lacking scales in the Anophelinae subfamily. In general, mosquito head, thorax, and abdomen are concealed with setae of different shape, length, and coloration that gives genus and species-specific taxonomic significance (Norbert *et al.* 2010).

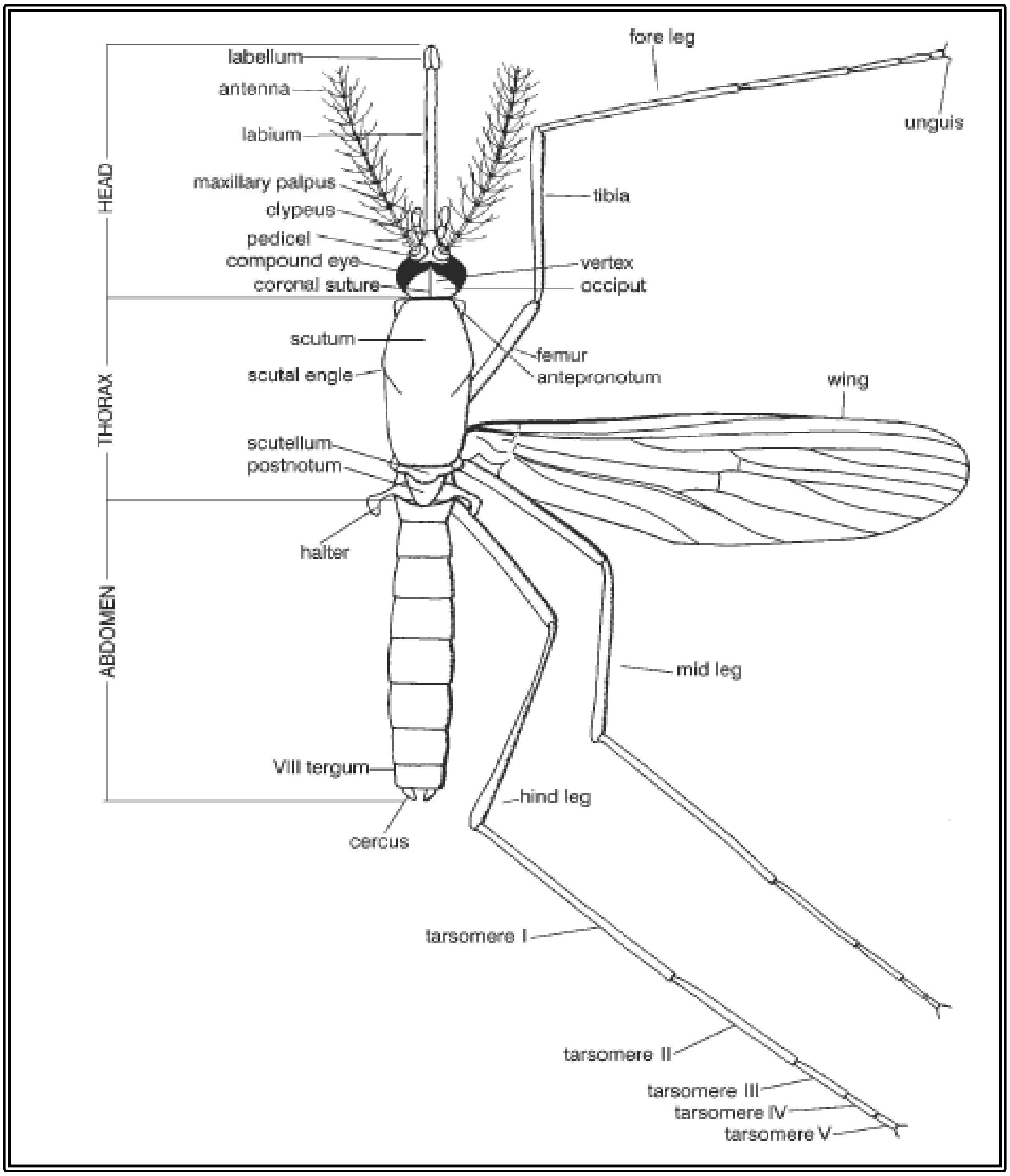


Figure 8: Adults Morphology (Norbert *et al.* 2010).

2.4.3 World Distribution

Mosquitoes have worldwide distribution, it has been encountered throughout temprate and tropic rigions (Mikhail 2009). They do not exist only in a few islands (e.g. Iceland and Faroe Islands) (Adam 2011) and Antarctica. Mosquitoes can breed in a widerange of habitats with fresh, brackish, or any water (turbid, clear, or polluted) excluding habitats that have high-salt concentration water such as marine habitats. Around 3,500 species and subspecies are known (Rueda 2008). The diversity of mosquitoes is very obvious, with numerous genera having universal spreading and some genera with more restricted distribution. Diverse geographical areas of the world have different mosquito species. The highest variety of mosquito species is originate in the Neotropical (NT) it contain about (31% of entire recognized species; 1069/3492) (Rueda 2008), tailed by the Oriental region with (30%), Afrotropical (22%), and Australasian (22%), while least percentages found in the Nearctic region with only (5%) (Figure 9). In the Neotropical, the highest quantity of species in Culicinae is originate under tribe Culicini, tailed by Aedini and Sabethini. While in the Oriental, Aedini has the highest quantity of species, tailed by Sabethini and Culicini. Orthopodomyiini, Culisetini, Hodgesiini, and Aedeomyiini have lesser quantity of species in the Oriental. In the Afrotropical, Nearctic, Australasian and Palearctic region, Aedini and Culicini have the highest quantity of species.

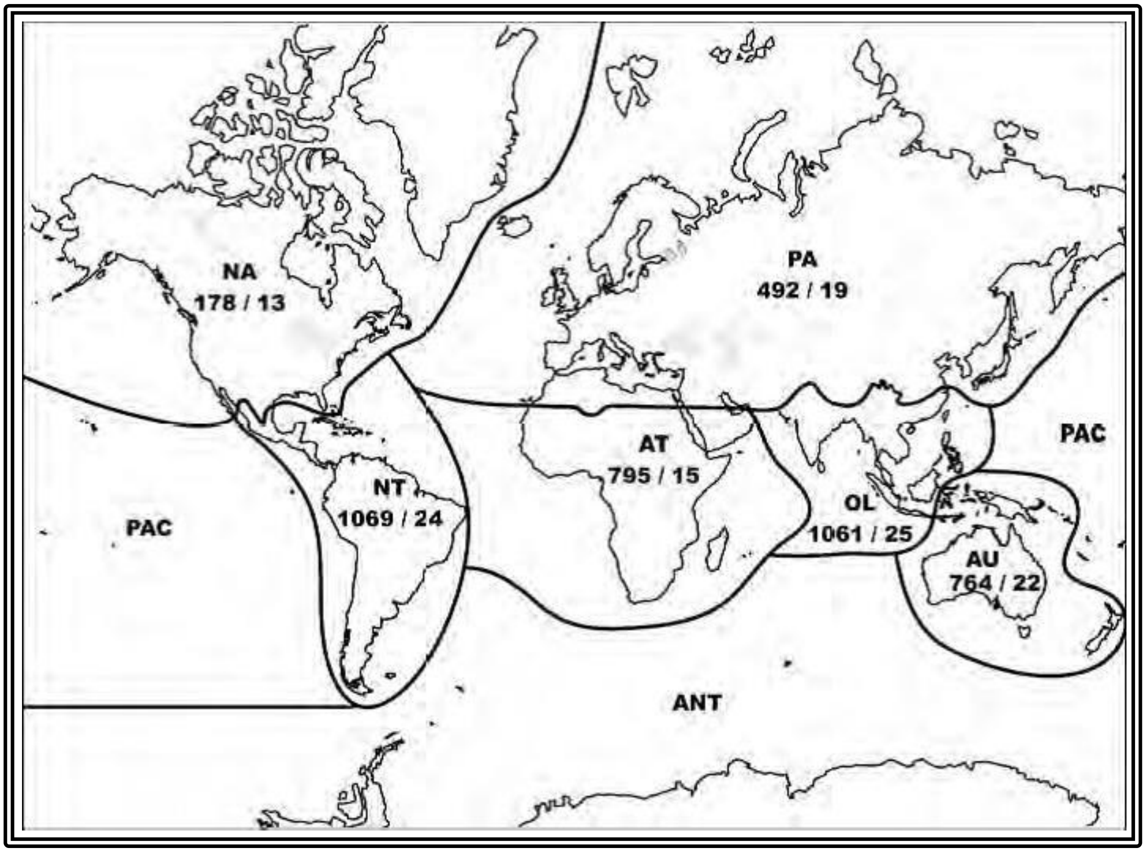


Figure 9: Number of species and genera in each zoogeographical region (from Tables 2 and 3). PA, Palaearctic; NA, Nearctic; NT, Neotropical; AT, Afrotropical; OL, Oriental; AU, Australasian region including South Pacific Islands (PAC: Pacific Oceanic Islands; ANT: Antarctic) (Rueda 2008).

47 species are below undefined subgenera in four genera, i.e. *Culiseta* (1 species in AU), *Aedes* (1 species in undefined genus in AU), *Wyeomyia* (37 species in NT), and *Culex* (7 species in NT, 1 in AU). Of the 81 identified subgenera and 42 genera, The Neotropical has 58% and 57% respectively. worldwide; Nearctic region, 23% and 31%; Oriental, 52% and 60%; Afrotropical region, 33% and 38%; Australasian region (counting South Pacific Islands), 53% and 55%; and Palearctic region, 39% and 45%. In altogether regions, subfamily Culicinae has bigger quantity of species comparing to subfamily Anophelinae. In the subfamily Culicinae, tribe Aedini within subfamily

Culicinae, has the highest quantity of, excluding the NT. In the Oriental, Afrotropical and Australasian regions, species from the entire 10 tribes of the subfamily Culicinae are encountered. Numerous countries or island in certain regions have prevalent species of mosquitoes. For instance, in the Philippines over 40% of the mosquito are endamic in that country (Tsukamoto *et al.* 1985). While in the South Pacific Islands, over 84% of the native species are common to the islands (Belkin 1962). Because of the quite poor knowledge of several mosquito groups, it is very hard to determine the precise affinities of mosquito fauna from many areas. An efficient list of widespread species regarding each country or the overall distribution record of species is established in the online Culicidae systematic catalog.

2.4.4 Arabian Peninsula Distribution

Distribution of mosquitoes (Diptera: Culicidae) in Saudi Arabia has been explored by many workers. Mattingly and Knight (1956) studied the distribution of mosquito in the Arabian Peninsula and documented 46 species and subspecies. In southwest of Saudi Arabia seven genera and 16 species were identified (Bakr *et al.* 2014; Abdullah and Merdan 1995), namely; *Aedes (Stegomyia) aegypti* (Linnaeus 1762), *Aedes caspius* (Pallas 1771), *Aedes (Aedimorphus) vexans arabiensis* (Patton 1905), *Anopheles pretoriensis* (Theobald 1903), *Anopheles d'thali* (Patton 1905), *Anopheles turkhudi* (Liston 1901), *Anopheles arabiensis* (Patton 1905), *Culex (Lutzia) tigripes* (Grandpre and Charmoy, 1900), *Culex decens* (Theobald 1901), *Culex sitiens* (Wiedemann 1828) *Culex bitaeniorhynchus* (Giles 1901) , *Culex quinquefasciatus* (Say 1823), *Culex pipiens* (Linnaeus 1758), *Culex tritaeniorhynchus*

(Giles 1901), *Culex sinaiticus* (Kirkpatrick 1924) and *Culliseta longiareolata* (Macquart 1838). The most common species in this region is *Culex pipiens* (Linnaeus 1758).

In the Eastern areas of Saudi Arabia, Wills *et al*. (1985) recorded *Anopheles fluviatilis* (James, 1902), *Anopheles sergentii* (Theobald 1907), *Anopheles tenebrosus* (Donitz 1902), *Culex pipiens* (Linnaeus 1758), *Culex quinquefasciatus* (Say 1823), *Culex tritaeniorhynchus* (Giles 1901), *Culex univittatus* (Theobald 1901), *Uranotaenia unguiculata* (Edwards 1913) and *Culiseta longiareolata* (Macquart 1838). According to Abdoon and Alshahrani (2003), *Anopheles arabiensis* (Patton 1905) is the most abundant mosquito in the southern part of the region also in addition to six species of anopheline mosquitoes in this Region, which were: *Anopheles d'thali* (Patton 1905), *Anopheles rupicolus* (Lewis 1937), *Anopheles sergentii* (Theobald 1907), *Anopheles multicolor* (Cambouliu 1902), *Anopheles turkhudi* (Liston 1901) and *Anopheles pretoriensis* (Theobald 1903). In the region of Riyadh, Al Kuriji *et al.* (2007) listed 15 species which comprised *Aedes caspius* (Pallas 1771), *Anopheles stephensi* (Liston 1901), *Anopheles coustani* (Laveran 1900), *Anopheles d'thali* (Patton 1905), *Anopheles pretoriensis* (Theobald 1903), *Culex laticinctus* (Edward 1913), *Culex perexiguus* (Theobald 1903), *Culex pipiens* (Linnaeus 1758), *Culex quinquefasciatus* (Say 1823), *Culex simpsoni* (Theabald 1905), *Culex sinaiticus* (Kirkpatrick 1924), *Culex theileri* (Theobald 1903), *Culex tritaeniorhynchus* (Giles 1901), *Culex univittatus* (Theobald 1901) and *Culiseta longiareolata* (Macquart 1838). AI-Ali *et al.* (2008), studied the mosquitoes in El

Madina district reporting 7 species: *Culex duttoni* (Theobald 1901), *Culex decens* (Theobald 1901), *Culex pipiens* (Linnaeus 1758), *Culex quinquefasciatus* (Say 1823), *Culex univittatus* (Theobald 1901), *Culex sinaiticus* (Kirkpatrick 1924), *Culex laticinctus* (Edward 1913), *Culex pipiens* (Linnaeus 1758) and *Culex quinquefasciatus* (Say 1823) were the most abundant. Through the 2000 epidemic of Rift Valley fever in the southern area of Saudi Arabia, Jupp *et al*. (2002) and Miller *et al.* (2002) collected 7 species of mosquitoes:*Aedes (Aedimorphus) vexans arabiensis* (Patton 1905), *Culex tritaeniorhynchus* (Giles 1901), *Aedes vittatus* (Bigot 1861), *Anopheles azaniae* (Bailly-Choumara 1960), *Culex pipiens* (Linnaeus 1758), *Aedes (Ochlerotatus) caballus* (Theobald 1912) and *Aedes (Ochlerotatus) caspius* (Pallas 1771). The distribution of mosquitoes in Mekkah Region was studied by Alahmed *et al.* (2009), record the following species: *Aedes caspius* (Pallas 1771), *Aedes aegypti* (Linnaeus 1762), *Anopheles d'thali* (Patton 1905), *Anopheles gambiae* (Giles 1902), *Anopheles multicolor* (Cambouliu 1902), *Anopheles rhodesiensis* (Theolbald 1901), *Anopheles sergentii* (Theobald 1907), *Anopheles stephensi* (Liston 1901), *Anopheles subpictus* (Grassi 1899), *Anopheles turkhudi* (Liston 1901), *Culex arbieeni* (Salem, 1938), *Culex laticinctus* (Edwards 1913) *Culex pipiens* (Linnaeus 1758), *Culex quinquefasciatus* (Say 1823), *Culex sinaiticus* (Kirkpatrick 1924), *Culex (Lutzia) tigripes* (Grandpre and Charmoy 1900) , *Culex tritaeniorhynchus* (Giles 1901), *Culex univittatus* (Theobald 1901) and *Culiseta longiareolata* (Macquart 1838). Members of the genus *Culex* were the most dominant also in the Eastern area, Hafr El Batin, Ahsaa and El Dammam, of Saudi Arabia, *Aedes caspius* (Pallas 1771), *Anopheles* *cinereus* (Theobald 1901), *Anopheles coustani* (Laveran 1900), *Anopheles d'thali* (Patton 1905), *Anopheles fluviatilis* (James 1902) *Anopheles gambiae* (Giles 1902), *Anopheles multicolor* (Cambouliu 1902), *Anopheles pretoriensis* (Theobald 1903), *Anopheles rhodesiensis* (Theolbald 1901), *Anopheles sergentii* (Theobold 1907), *Anopheles stephensi* (Liston 1901), *Anopheles subpictus* (Grassi 1899), *Anopheles superpictus* (Grassi 1899), *Anopheles tenebrosus* (Donitz 1902), *Culex laticinctus* (Edwards 1913), *Culex perexiguus* (Theobald 1903), *Culex pipiens* (Linnaeus 1758), *Culex quinquefasciatus* (Say 1823), *Culex simpsoni* (Theobald 1905), *Culex torrentium* (Matrini 1925), *Culex tritaeniorhynchus* (Giles 1901), *Culex univittatus* (Theobald 1901), *Culiseta longiareolata* (Macquart 1838) and *Uranotaenia unguiculata* (Edwards 1913) were recorded by Alahmed (2012).

Table 2: Distribution of *Anopheles (Cellia)* in southwestern Asia and Egypt (Harbach 1985).

	Afghanistan	Bahrain	Cyprus	Egypt	Iran	Iraq	Israel	Jordan	Kuwait	Lebanon	Oman	Pakistan	Qatar	Saudi Arabia	Syria	Turkey	U.A.E.	Yemen
annularis	•											•						
apoci					•	•												
arabiensis														•				•
azaniae																		•
cinereus				•			•	•			•			•				•
culicifacies	•				•	•					•	•					•	•
demeilloni																		•
dthali	•			•	•	•	•	•		•	•	•		•	•		•	•
fluviatilis	•	•			•	•					•	•		•				•
maculatus	•											•						
moghulensis	•				•							•						
multicolor	•		•	•	•	•	•	•		•	•	•	•	•	•			•
paltrinierii											•						•	
pharoensis				•			•	•						•	•			•
pretoriensis														•				•
pulcherrimus	•	•			•	•	•		•	•		•		•	•	•		
rhodesiensis rupicola				•			•	•		•	•			•	•			•
sergentii				•	•	•	•	•		•	•	•	•	•	•		•	•
splendidus	•											•						
squamosus																		•
stephensi	•	•			•	•			•		•	•		•			•	
subpictus	•				•							•						
superpictus	•		•	•	•	•	•	•		•		•		•	•	•		
turkhudi	•			•	•	•	•	•			•	•		•				•
willmori	•											•						
n. sp.				•														

2.4.5 Mosquitoes' Distribution in Qatar

A study in Qatar reported six indigenous species in five Municipalities, these species were *Culex pipiens* (Linnaeus 1758), *Culex univittatus* (Theobald 1901), *Culex pusillus* (Macquart 1850), Culex *caspies*, *Anopheles multicolor* (Cambouliu 1902) and *Anopheles stephensi* (Liston 1901). The survey showed the presence of *Culex pipiens* (Linnaeus 1758) and *Culex univittatus* (Theobald 1901) in Al Rayyan, Doha, Al Khor and Al Zakhira. *Culex pipiens* (Linnaeus 1758) had the highest density in all Municipality, *Culex pusillus* was found in Doha and Al Daayan. *Aedes caspies* (Pallas 1771) was found in Al Daayan and Al Shamal. Results also showed that *Anopheles multicolor* (Cambouliu 1902) and *Anopheles stephensi* (Liston 1901) were mainly present in Al Samal and had low density in Al Rayyan (Ain Khalid) (Mikhail *et al*. 2009). In brief, *Culex pipiens* complex in Qatar is the most important insect with high incidence and prevalence due to availablility of pools sewage swamps for breeding water. Moreover, *Culex quinquefasciatus* (Say 1823) were also reported in Qatar (Harbach 1985) in addition to two *Anopheles* species, *Anopheles multicolor* (Cambouliu 1902) and *Anopheles stephensi* (Liston 1901) (Harbach 1985; and Khan *et al.* 2009). Glick, 1992 reported that two *Anopheles* species has been found in Qatar *Anopheles multicolor* (Cambouliu 1902) and *Anopheles sergentii* (Theobald 1907).

3 Methodology

3.1 Study area

This study was carried out at Al Rayyan municipality, Qatar State. (Figure 10).

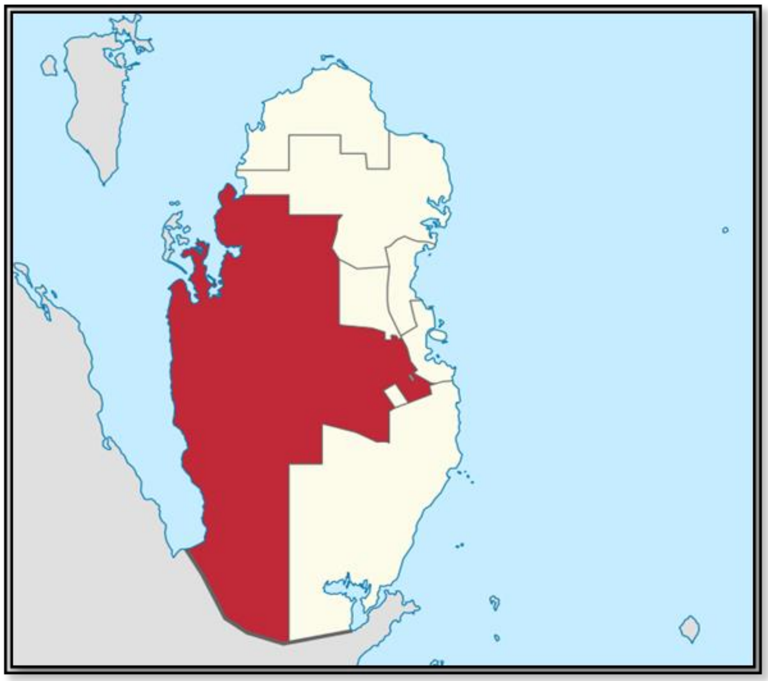


Figure 10: Rayyan Municipality, State of Qatar (Qatar Meteorology Department 2013).

3.1.1 Geography

Qatar State, a peninsula located on the western coast of the Arabian Gulf, boarded only by Saudi Arabia to the south and the rest of the region is surrounded by the Arabian Gulf. The state covers an area of 11,525 Km^2 including some islands. It stretches from north to south in about 187 Km, with a coastline length of 563 Km.

The state lie between °24 ′27 and °26 ′10 north latitudes and °50 ′45 and °51 ′40 east longitudes. The population in 2015, is 2,334,029 persons

3.1.2 Topography

Qatar mostly comprises of flat rocky surfaces. But also, consist of some hills and sand humps which reach an elevation of 40m above sea level in the northern and western areas of the country. The north and central areas consist of some geographical features that are unusual to the Arabian Gulf. These comprise rainwater-draining basins, which considered the most productive and have involved substantial agricultural investment.

3.1.3 Climate

The climate in general is characterized by hot summers and mild winters. Slight winter rainfall averaging about 80 millimeters a year (Figure 11).

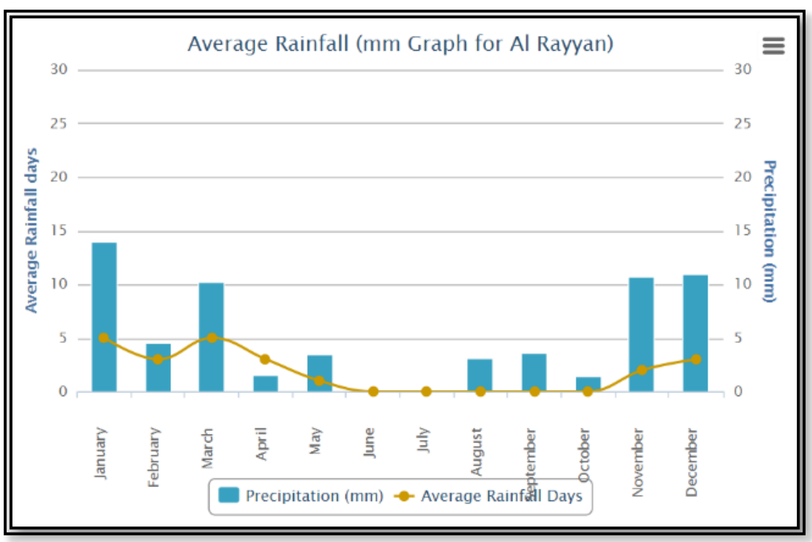


Figure 11 AlRayyan Avrage Rainfall (Qatar Meteorology Department 2013).

Temperatures vary from 45 °F in January to about 113 °F at the height of summer (Figure 12). However, it is usually pleasant during winter (Qatar Embassy 2015).

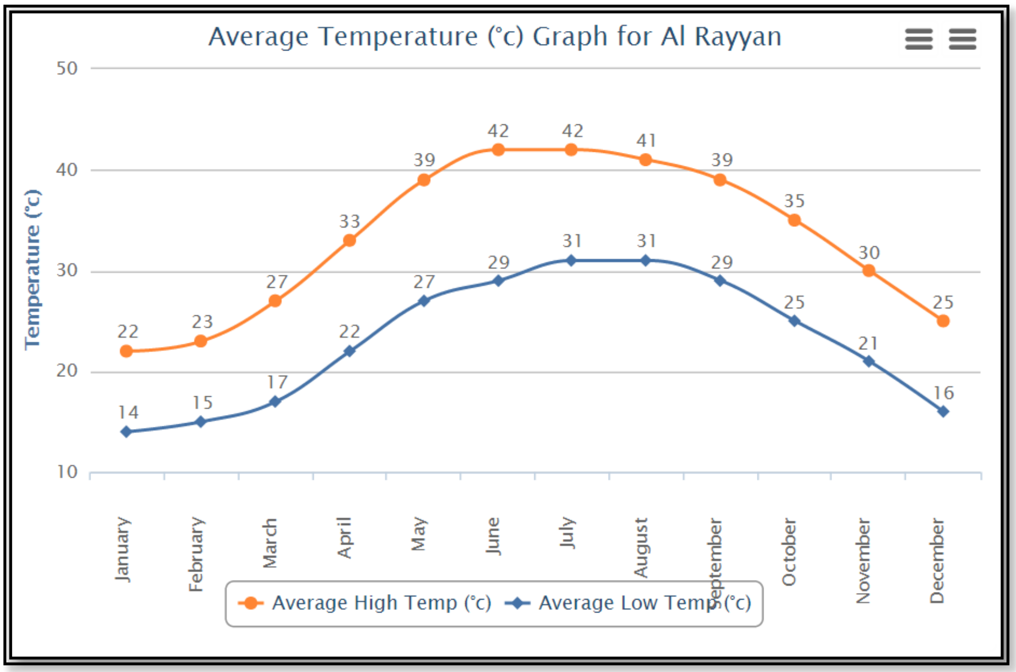


Figure 12: Al Rayyan Average Temprature. (Qatar Meteorology Department 2013).

3.2 Materials

3.2.1 Glassware and Plastic Containers

For rearing of mosquitoes, plastic containers with volumes of 3L, 2L, 1L and 1/2L were used. Throughout the process, the plastic containers were covered with muslin cloth. Mosquitos' larvae and pupae collected from different locations were kept in the plastic containers and reared up to adult's stage.

Glass containers with volume of 150 ml covered with plastic covers, were used for conserving mosquitoes adults, moreover, a small glass bottles size x were used for reserving mosquitoes after identification (Figure 13).

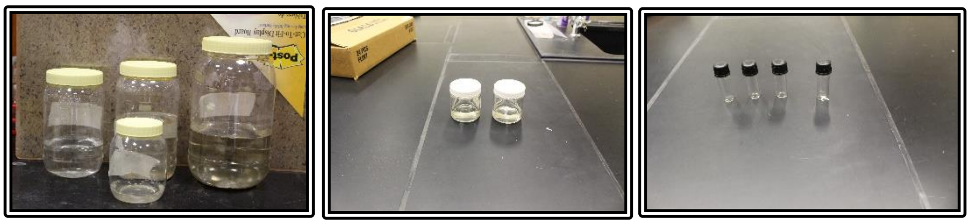


Figure 13: Glassware and Plastic Containers

3.2.2 Equipment’s Apparatus

3.2.2.1 Golobal Posistioning Systyem

Garmin GPS model MONTANA 650, space-based satellite navigation system device that delivers location and time information in all weather circumstances, anywhere on or near the earth where there is clear sight to stleast four or more GPS satellites, were used to identify the location of sampling sites. Also, Canon camera was used for taking photographs throughout the process (field and lab).

3.2.2.2 Collecting Adult Mosquitoes

Light traps (Pest Trap-GM950) was used to capture adult’s stage with big volume design (appropriate for breeding and planting areas), DC fans with ultra-long life, and funnel-type of anti-mosquito outflow device and dumet UV source with long-term and steadiness wavelength (Figure 14). Also, insect net with plastic handle of 80 cm long, circular iron ring of 22 cm diameter and mosquitoes bag of 70 cm in depth were used to collect adult mosquitoes at sites with no electricity source.

Figure 14: Light trap

3.2.2.3 Tools for Collecting Larvae

Dipper with a handle of 2.3 m and a mouth of 300 ml collecting mosquitos' larvae from potential habitats, white bowl of 350 ml were also used during larvae sampling to observe presence of larvae. Muslin cloth was used during the rearing process to prevent the developed adults from escaping in addition to providing larvae with oxygen (Figure 15).

3.2.2.4 Equipments and Tools Used at the Lab

Stereomicroscope with objectives r X t, 80 was used for identifying mosquitos' genera. In addition, an advanced Microphotographic Stereomicroscope was used to take some morphological photographs for our adult collection. Camel hair brushes were used for handling mosquitoes during several processes (separating emerged adult from larvae, emptying light traps and examination) (Figure 15). Also, Wooden storage box with capacity of 150 samples' bottles were used to store all mosquitoes samples' after examination.

3.2.2.5 Chemicals

Ethanol with 70% concentration were used for killing and preserving adults' mosquitoes or for rearing mosquitoes (Figure 15).

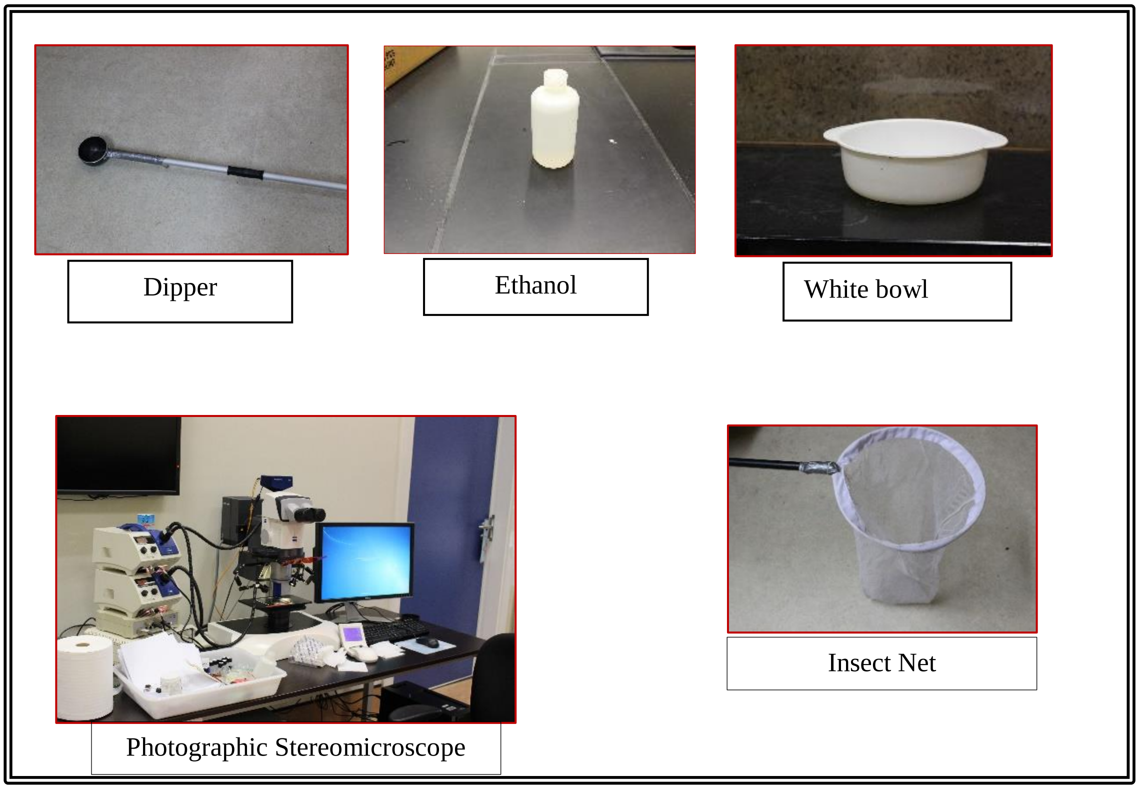


Figure 15: Material and Apparatus.

3.3 Methods

Mosquitos' larvae and adults were collected from different locations. Selection of sampling sites was based on the presence of stagnant water. Several visits were made to collect larvae in potential breeding sites. Eighteen larvae samples were collected from mostly rural areas namely, Shahaniya, Rodat Rashed, Um Alzubara, Um Almwagia, Abu Nakhla, karana, Miazzer. Samples were kept separately and reared for their adult emergance in the laboratory. Adults' mosquitoes were collected using light traps and insect net, from selected urban and rural areas in addition to two other sites used for conserving sewage waste water (Figure 20) is showing the percentage between sites visited from rural areas and urban areas. These sites were namely, Abu Nakhla sewer pond, Karaanah Sewer Pond, Shahaniya, Rodat Rashed, Um Almoaga, Um Alzubara, Miaazer, Alganim, Alazaziya. Alwab, Dahil, Alsylia, Miseemir and Dokhan. Adult specimens identified using systematic keys and catalogues of (Harbach 1985; and Gilck 1992). Collection sites were described to determine it is location characteristic as well as it is natural, physical and chemical characters. These characters include water movement described as running water or stagnant; light intensity described as sunny, partially sunny and shaded; depth of breeding site described as less or more than one meter; types of breeding site described as rain water, sewer water, irrigation etc.

3.3.1 Samples Collection

3.3.1.1 Collecting Larvae

Several visits have been carried out to potential breeding sites for collecting larvae. Selection of breeding sites was based on Knowledge and information from literature in addition to information from Al Rayan Municipality. A dipper with a long handle is used in big breeding sites to reach far part of the water body and to avoid disturbing larvae during sampling which will cause larvae to dive down (Figure 16). Numbers of dips depend on the size of breeding site. Samples were taken from the edges and centers of breeding sites. For sampling from small breeding sites such as buckets, construction tanks and small ponds a white bowl is used. Larvae were put in plastic containers with a lid, which were filled about 3/4 with water from the breeding site. (Norbert et al. 2010). Samples were labeled and transported to the lab.

3.3.1.2 Collecting Adults

Light traps (with UV light) were used mostly in collecting mosquito's adults. The traps need electricity power to operate, electricity extensions were used to allow sampling from locations far from electricity power source. Samples were collected from urban and rural areas. The traps were set in the selected locations before sunset and collected two to three hours later (rural areas) or collected next day after sunrise (urban areas). One month monitoring survey was carried out in one location where light traps were set all day long. Three samples were collected every day 7 am to 5 pm; 5 pm to 10 pm and 10 pm to 7 am. Weather observation was recorded during the period sampling such as temperature, humidity, rain sand storms etc. Temperature was taken at certain times during each sampling period as follow 5 am, 2 pm and 6 pm). Insect net was used to collect adults in areas with no electricity power source (Figure 17).

3.3.2 Lab Work

3.3.2.1 Larvae

The water pH was measuerd immediately after arriving at the lab using Denver Instrument, Ultra Basic pH meter. Larvae were kept in the plastic containers in the fields and covered with muslin cloth to prevent the developed adults from escaping as well as to allow oxygen to reach the larvae. Larvae were reared up to adults' emergence, the emerged adults were killed, using ethanol and preserved in ethanol with 70 % concentration, in small glass containers, covered with plastic lid to keep ethanol from evaporating.

3.3.2.2 Adults

Light traps were brought to the lab after collecting samples. Ethanol was used for killing the insect. A piece of light colored plastic was laid on the counter (to aid accurate view). The traps were emptied gently over the light colored plastic, big insect were separated first (different insects were caught in the light traps). The rest was transferred into white bowl to be examined under insect magnifier. For handling and moving insect a camel brush was used. The magnifier helps to separate mosquitoes from other insect although some time it was hard to determine if it was a mosquito or not (light traps damage samples). After that mosquitoes were put in small glass containers with a label and reserved in 70% ethanol.

3.3.3 Analyzing Samples

Adult's mosquitoes were examined under stereomicroscope. Identification was carried out using the standard keys (Harbach 1985; and Glick 1992). Adults were examined individually, handled with camel brush or thin entomological pin. Mosquitoes were then reserved individually each in small bottle in ethanol of 70% concentration. A Parafilm were used to secure the cover and to keep the ethanol form evaporating. A label with all the information (Sample type and No., genus, species and sex of the mosquito) was attached to the samples (Figure 18).

3.3.4 Analyzing the Data

A t–test was conducted to test if there is no significant difference in the pH concentration between type of mosquitoes (*Culex* and *Anopheles*). In the other hand, a chi square test was performed to conclude if there is a significant variance in the association between mosquitoes types found and the area where they have been captured.

3.3.5 Producing Maps Using GIS

Geographic information system (GIS) is a system intended to capture, manipulate, store, manage, analyze and present all types of spatial or geographical data. Many disciplines can benefit from GIS technology. In this study maps were produced using GIS tools. The maps showing all samples sites, in addition to some chosen attributes such as (type of breeding site, number of mosquitoes collected types of mosquitoes etc.) Arc GIS program, 10. 2 was used to produce the maps.

Collecting Larvae Samples

Two small containers with larvae at Rodat Rashid

Abu Nakhla sewage waste water pond

Leaking water from underneath some houses in Meaazer

Sewer waste water at Shahania Animal complex

Water tank from construction site in Al wajba

Animal drinking water at Rodat Rashid

Figure 16: Collecting Larvae

Collecting Adults Samples

Using light trap to collect adults at plant market. Al Garafa

Using the net to collect adults at sewage waste water pond Alkaraana

Using net to collect mosquitoes while resting at sewage waste water pond Alkaraana

Using light trap to collect adults at Maayzer were sewer waste water were leaking

Using light trap to collect aduts from animals and plant farm at Rodat Rashed

Using light trap to collect adults from residential area at Al azeezya

Figure 17: Collecting Adults

Figure 18: Lab Activities

4 Results

Mosquito survey was conducted in Al Rayyan Municipality, Qatar State, for five months (December, 2014 to April, 2015). A total of 37 collection sites were visited throughout of the study period revealing a total of 312 mosquitoes from which 115 (37%) mosquitoes has been reared from larvae (L) in the lab while 197 (63 %) mosquitoes have been collected as adults (At) (Figure 19) **.**

Out of the 312 mosquitoes, 155 were female, from which 89 have been captured as adults and 66 has been reared from larvae, while 157 mosquitoes were males, from which 108 mosquitoes have been captured as adults and 49 have been reared from larvae (Figure 19).

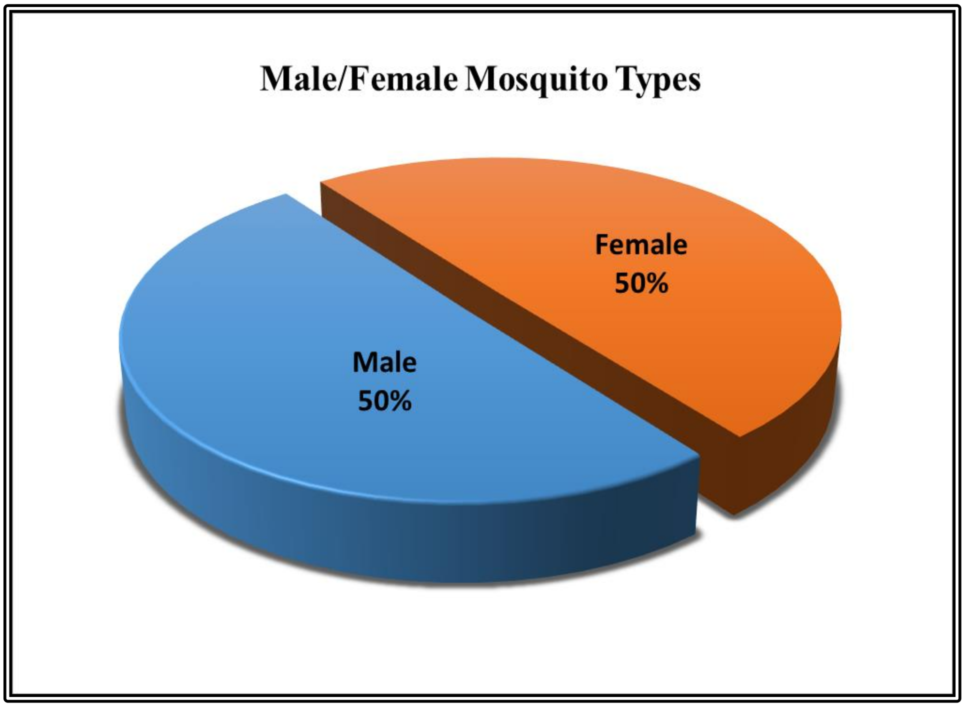


Figure 19: Female vs. Males

Percentage of mosquitoes collected from urban sites visited during the study period compared to rural sites was 58% to 42% respectively (Figure 20). Moreover, most of urban sites were positive, (where mosquitoes has been captured or reared from larvae to adults), only one site was negative, while a number of rural site were negative (no mosquitoes were captured or successfully reared to adult formation), where nine sites out of 23 was negative (Figure 21), (Larvae did not developed into adult formation).

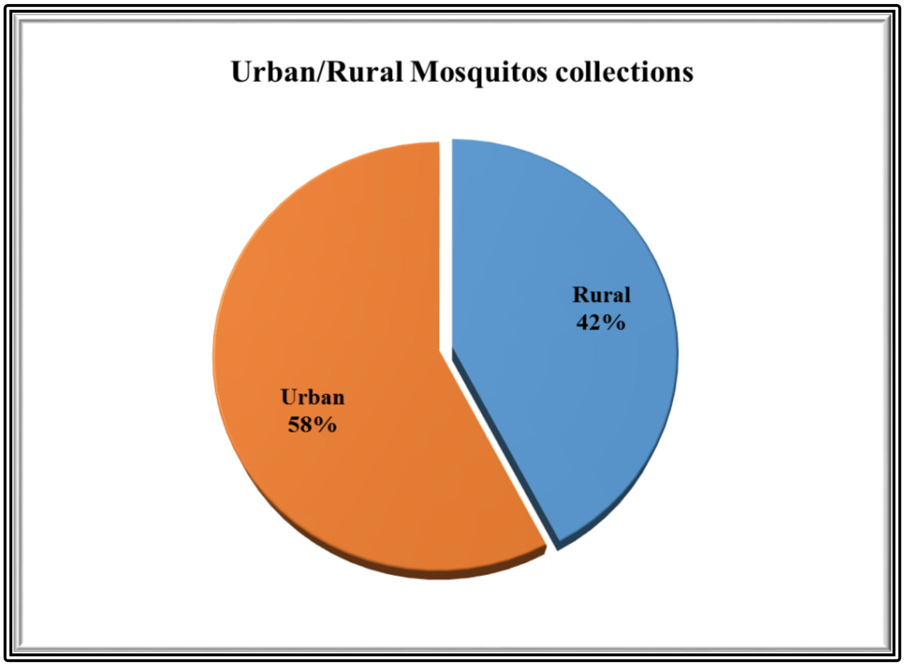


Figure 20: Rural Sites vs. urban sites

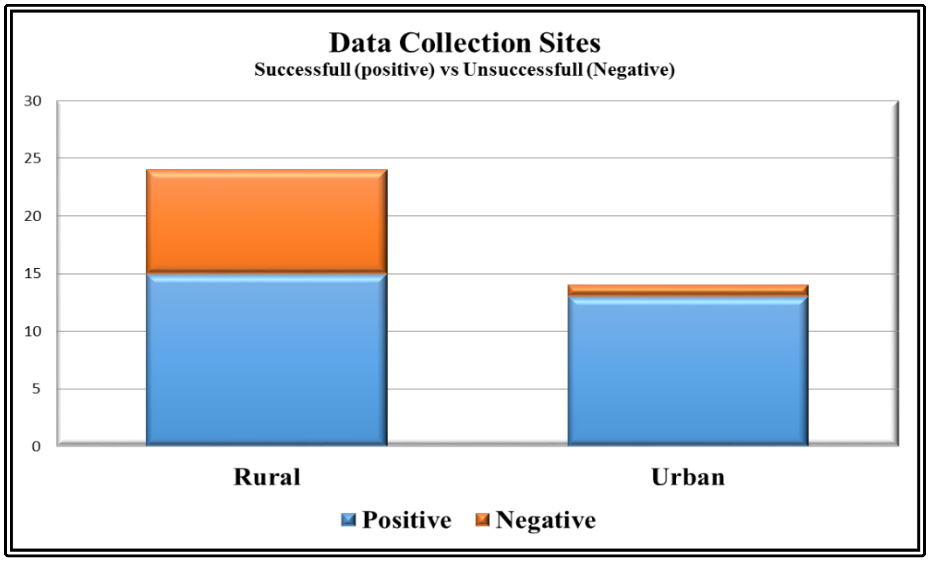


Figure 21: Sites with successful development Vs sites with unsuccessful development.

The Sampling sites has been categorized into six types, animal farms (A), plant farms (P), animal and plant farms (AP), residential (R), Shallow ground water (SGW), sewage waste water Ponds (Pn) and Car Tyre Tyre L (Figure 22).

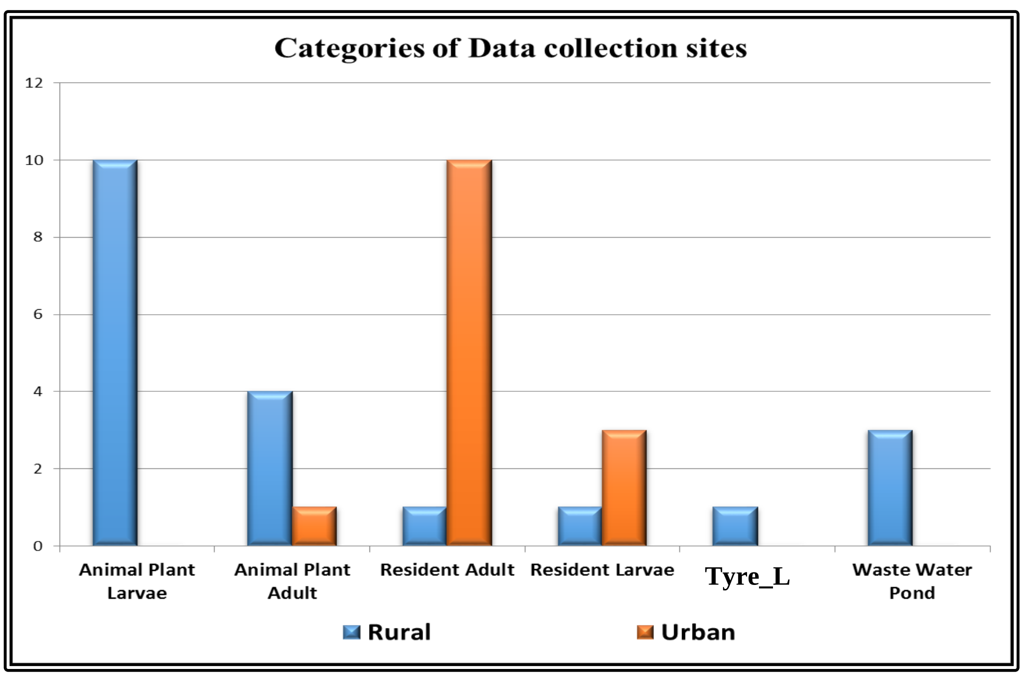


Figure 22: Samples sites category.

Types of mosquitoes genera encountered in these sites are displayed in (Figure 23).

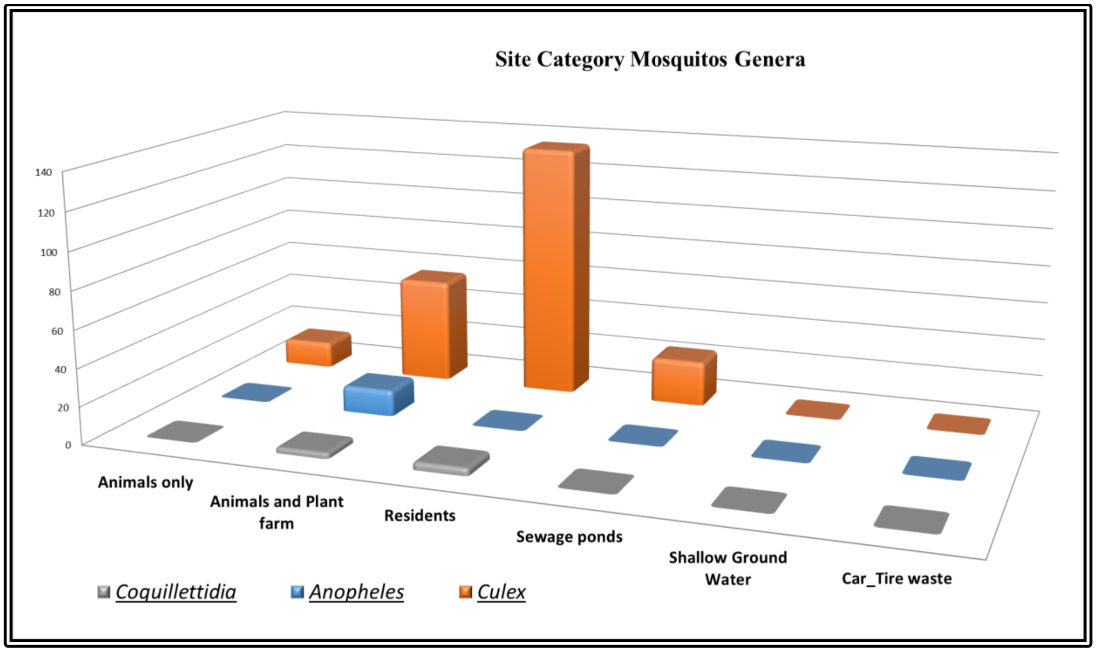


Figure 23: Site Category vs. Mosquitoes' Genera.

During the present survey four species were recorded and identified according to standard keys (Harbach, 1985; and Glick, 1992). The encountered species were belonging to 3 genera (Figure 24), namely; *Culex* 91% (3 species); *Anopheles* 6% (one species) and *Coquillettidia* 3% (no species has been identified samples are damaged).

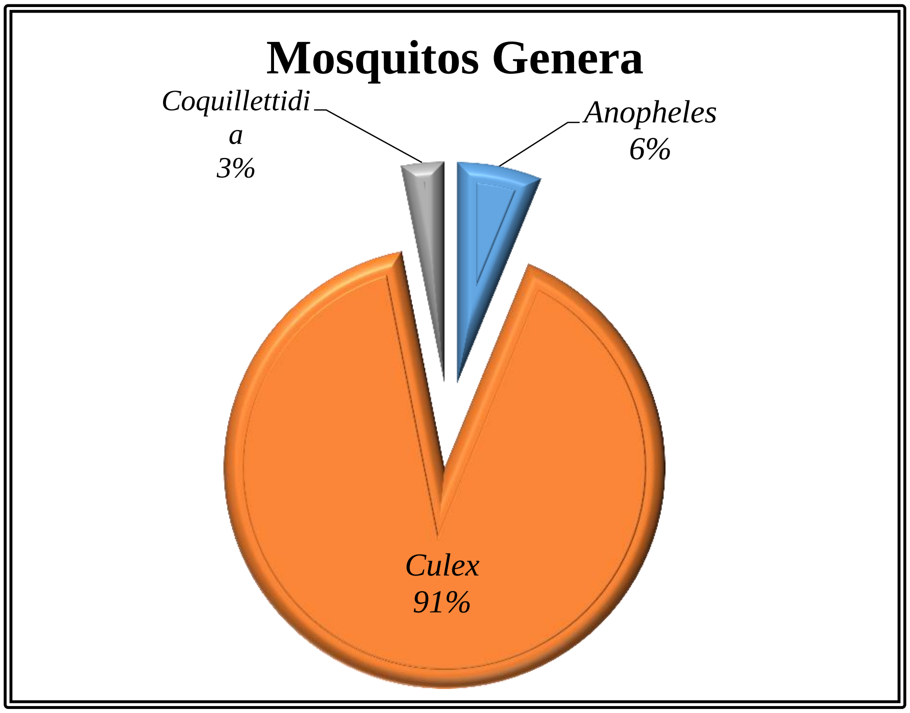


Figure 24: Types of genera

Sampling locations have been categorized into urban areas and rural areas. Two genera have been found in each area, however, in urban areas *Culex* and *Coquillettidia* were found while in rural areas *Culex* and *Anopheles* (Figure 25).

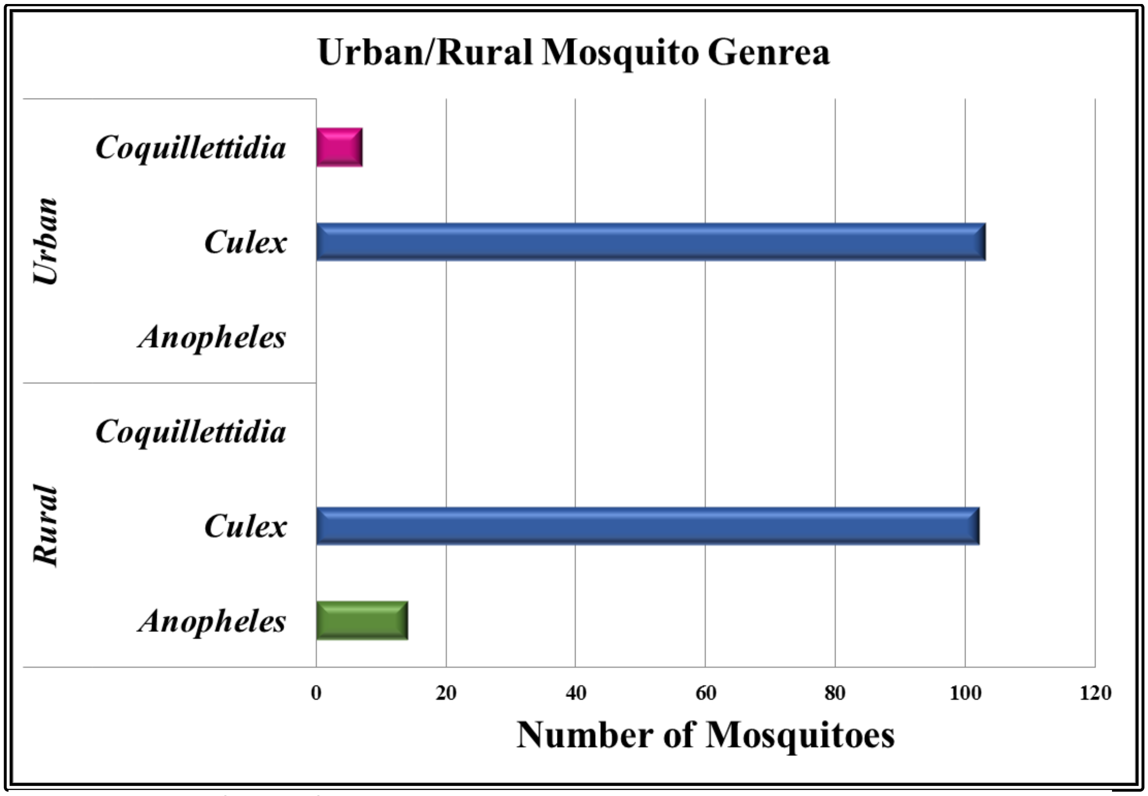


Figure 25: Types of genera found in urban and rural areas.

The number of adults' mosquitoes that reared from larvae was 115 mosquitoes out of this number, 9 individuals are *Anopheles* and the rest are Culex. Thus only two genera have been recorded regarding larvae samples (Figure 26) belonged to two genera, *Culex* and *Anopheles.*

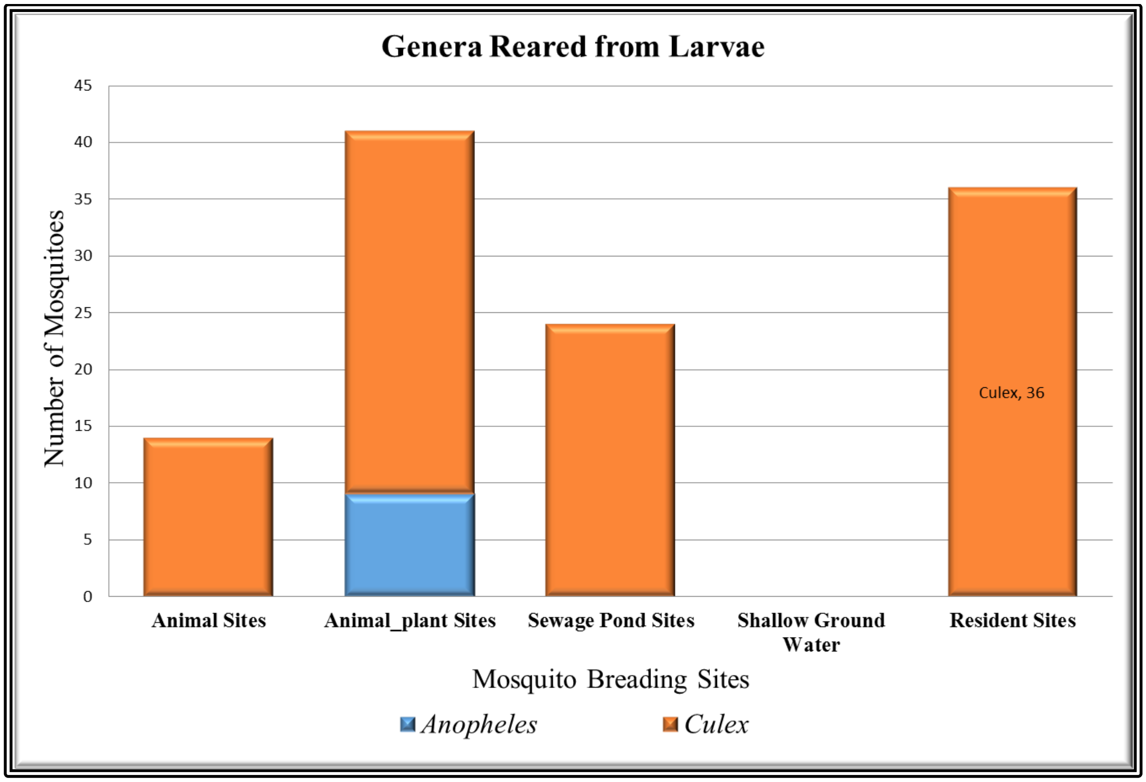


Figure 26: Mosquitoes genera identified from larvae

The range of pH taken from larvae breeding site was 8.46 – 6.2, the chart below display our finding (Figure 27).

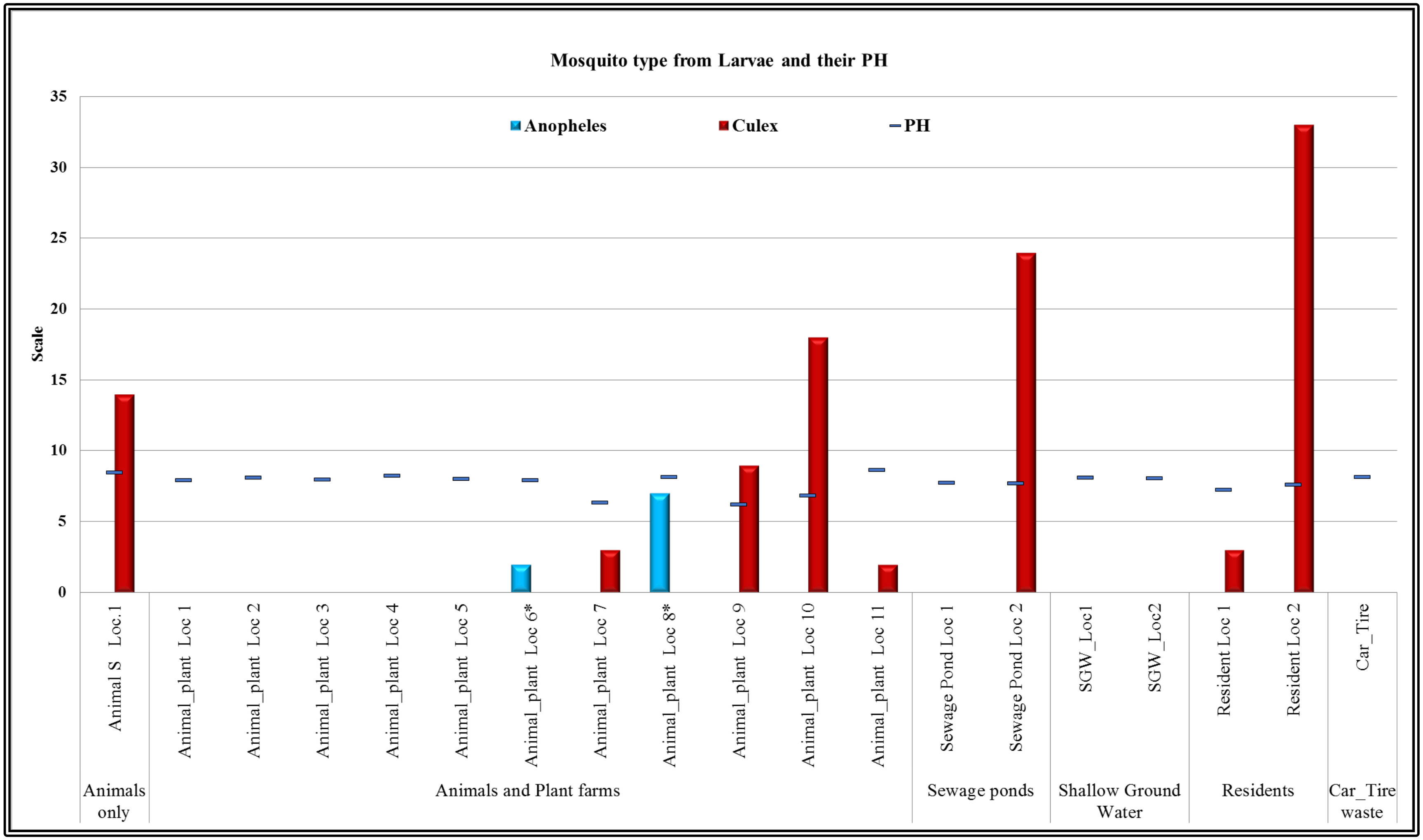


Figure 27: Mosquitoes Type Vs. pH

A one month survey was conducted to observe mosquitos' daily activity from March 10 – April 10. Three samples were collected every day (7 Am – 5 PM, 5 PM – 10 PM, 10 PM – 7 AM) temperature was recorded as well. A total of 63 mosquitoes have been collected throughout the period, only one species has been identified *Culex (Culex) vagans* (Figure 28).

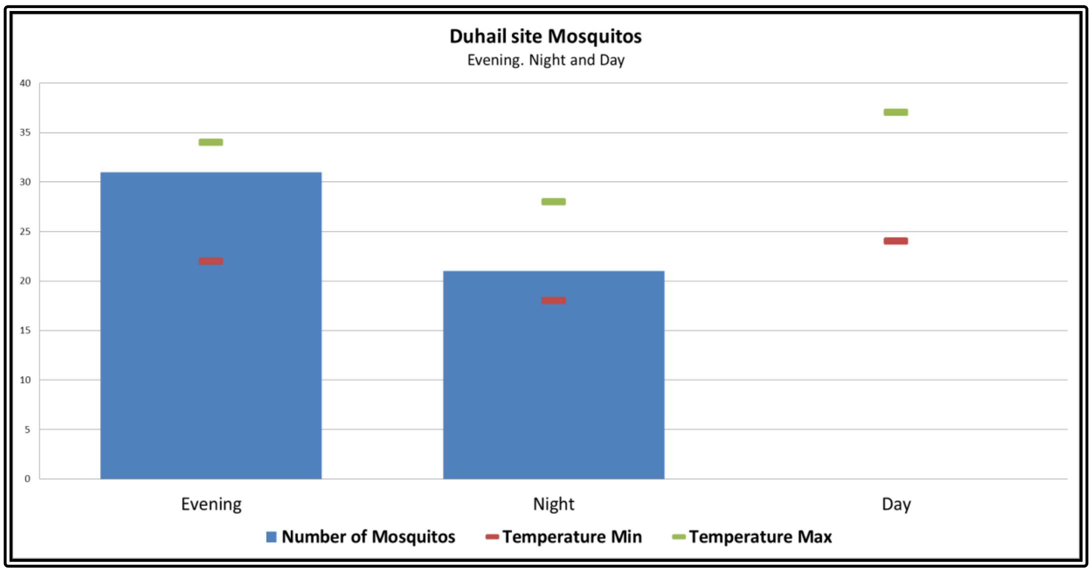


Figure 28: One Month Survey

When comparing the average temperature between evenings, night and day samples during the one month survey, night time samples recorded the least average among other sample followed by evening time and the highest temperature were recorded during day time. These averages reflected somewhat in the number of mosquitoes that have been collected. The highest number of mosquitoes has been collected during evening time (42 mosquitoes were captured) followed by night time samples (21 mosquitoes), however, no mosquitoes has been caught during day time (Figure 29).

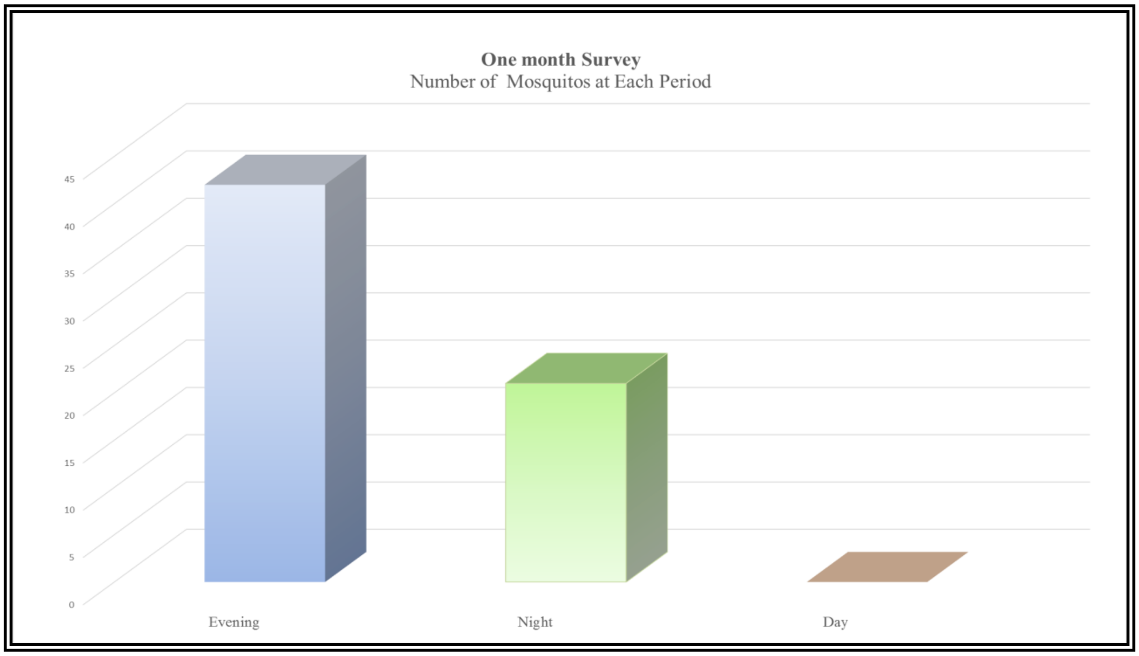


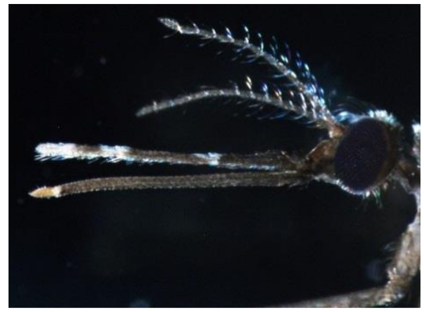
Figure 29: Total Mosquitos during day (Evening/Night/Day) for One Month Survey

4.1 Mosquitoes Genera

4.1.1 Anopheline Species:

One Anopheline species was collected from the surveyed areas, namely; *Anopheles stephensi* (Liston, 1901) (Figure 30). Distribution of this species is shown in (Figure 36).

1. *An. stephensi* Female



2. *An. stephensi* Head (palpi are as long as the proposcis. Palpomere 5 entirely pale

3.The wing: Wing with pale spots present on nearly all veins

.

Figure 30: *Anopheles stephensi*

4.1.2 Culicinae Species:

Three culicine species were encountered in the surveyed areas, namely; *Culex vagans* (Wiedemann 1828); *Culex mimeticus* (Noe 1899), and *Culex bitaeniorhynchus* (Giles 1901). The distribution of these three species is shown in. From the map it becomes clear that *Culex vagans* (Wiedemann 1828) is the most common culicine species in the study area. It was recorded in almost all types of breeding sites, with respect to *Culex mimeticus* (Noe 1899) which has been found only in Dukhan and captured as adults with light trap from residential area, and *Culex bitaeniorhynchus* (Giles 1901) which was found also in Dukhan, Al Ganim and Maazer.

4.1.3 *Coquillettidia* Species:

All sample with missing some part of their bodies or damaged, species identification was not possible.

4.2 Species and Their Habitats

4.2.1 *Anopheles stephensi* Liston

4.2.1.1 Distribution:

Two Larvae samples were collected from one location (Animal and plant farm). The farm is located in rural area Rodat Rashid (local streets, few houses or farms). The area is about 30 to 40 km from Doha (Figure 36). Three mosquitoes have been captured as adults from the same location using light trap figure in addition to another two *Anopheles* mosquitoes captured from another location using light trap.

4.2.1.2 Types of breeding sites:

Two small buckets (Figure 31) were filled with fresh water about half full. The pH was 7.88 and 8.14. The color of the water was greenish. Partially shaded with two trees beside them. Only very few larvae have been developed into adults.

Figure 31: *Anopheles* Breeding Sites

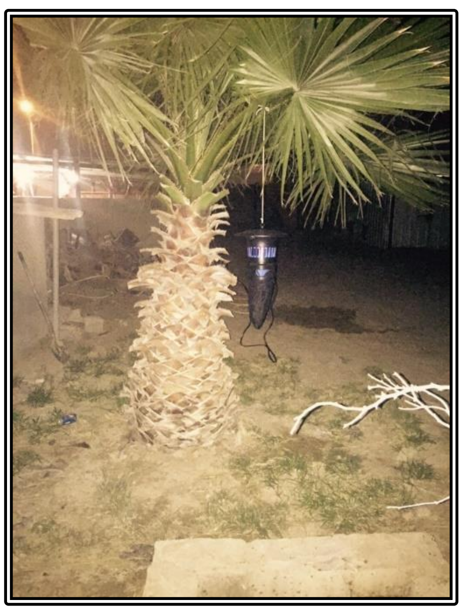


Figure 32: Using light trap

4.2.1.3 Seasons:

Larvae were collected in January- February, 2014 during winterseason

4.2.1.4 Description and Taxonomic of characters of Adults:

Maxillary palpus dark with at most 3 discrete pale bands; abdominal terga 11-VII with no dark scale-tufts; hindtarsomere 5 (Ta-Ills) Pale; Wing with pale spots existing on nearly all veins; Erect head scales broad, white on vertex (V) and dark brown laterally and posteriorly scutum with clear pale scales with setae and Palpomere 5 completely pale.

4.2.2 Culex mimeticus Noe

4.2.2.1 Distribution:

The species has been collected as adult using light traps from Dukhan (Fig. 37). Which is a relatively elevated area compared to other sampling location. However, this species has been found associated with *Culex bitaeniorhynchus* (caught in the same light trap).

4.2.2.2 Seasons:

Samples have been collected in March 2015.

4.2.2.3 Type of breeding site:

The type of breeding site is unknown, samples collected only as adults.

4.2.2.4 Description and Taxonomic Characters of female adults:

Scutellum trilobed, cell R, at least as long as vein R2,3; no spiracular setae, tarsomere 1 of fore- and midlegs no longer than tarsomeres 2-5 combined; hindungues small and inconspicuous; pulvilli present, no lower mesepimeral setae and wing with pattern of pale spots.

4.2.3 *Culex bitaeniorhynchus* Giles

4.2.3.1 Distributions:

This species was collected as adult using light traps from two locations, Dukhan and Al Ganim.

4.2.3.2 Types of Breeding Sites:

Types of breeding sites are unknown, since the samples have been collected as adult using light trap.

4.2.3.3 Season:

Samples have been collected during February- March, 2015 (wet season).

4.2.3.4 Discription and taxonomic Characters of Female Adults:

No lower mesepimeral setae; wing with variable amount of pale scaling along anterior veins; abdominal terga with apical pale bands; anterior surfaces of femora and tibiae speckled.

4.2.4 Culex vagans Wiedemann

4.2.4.1 Distribution:

Larvae of this species have been collected from Maazer (residential area), Al Shahaniya Animals Complex, Rodat Rashed and Alkarana sewage waste water pond. Also, adult samples have been collected from different locations, Al Ganim Alwaab, Al Garafah, Maazer and Alazyzia.

4.2.4.2 Types of Breeding Sites:

Breeding sites encountered were construction water tank, leaking sewer water, kitchen/sink sewer water and Al Karaanah swage waste water pond. The pH was 7.2, 7.6, 6.2, 6.8 and 7.88 respectively. The construction water tank was full and the water was leaking from the top since the water tab was slightly opened directly on the tank but the water was kind of calm. In the second location, water was leaking from underneath some house due to mistakes in the sewer system which results in the production of several small ponds. The third breeding site was a very big waste water pond (it is used as a point for sewer water collection)

4.2.4.3 Season:

Larvae have been collected throughout the study from December to April

4.2.4.4 Description of Taxonomic characters:

Lower mesepimeral setae present, tarsi all dark; no postspiracular scales; no prealar scales; anterior surfaces of fore- and midfemora and all tibiae with pale stripes.

4.2.5 GIS Maps

Numbers of maps have been produced using Geographical Information System (GIS) tools. Results have been displayed as layers on Qatar map to show the actual location for each layer of information. Results are shown in the below figures.

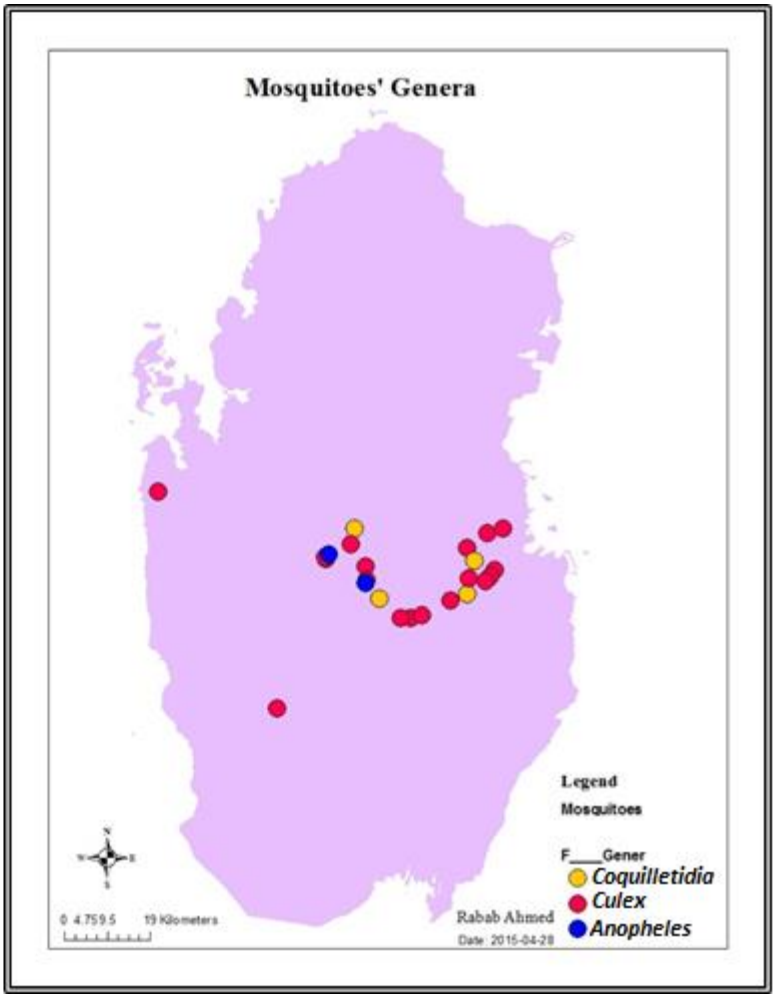


Figure 33: Distribution of genera in Al Rayyan Municipality

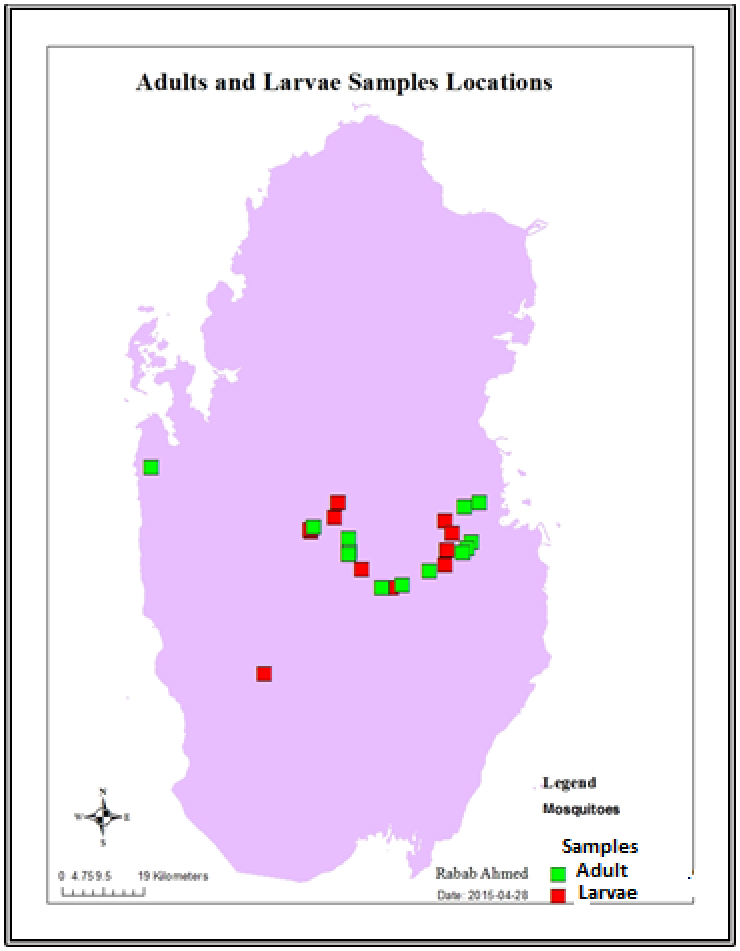


Figure 34: Larvae and Adults Samples Locations

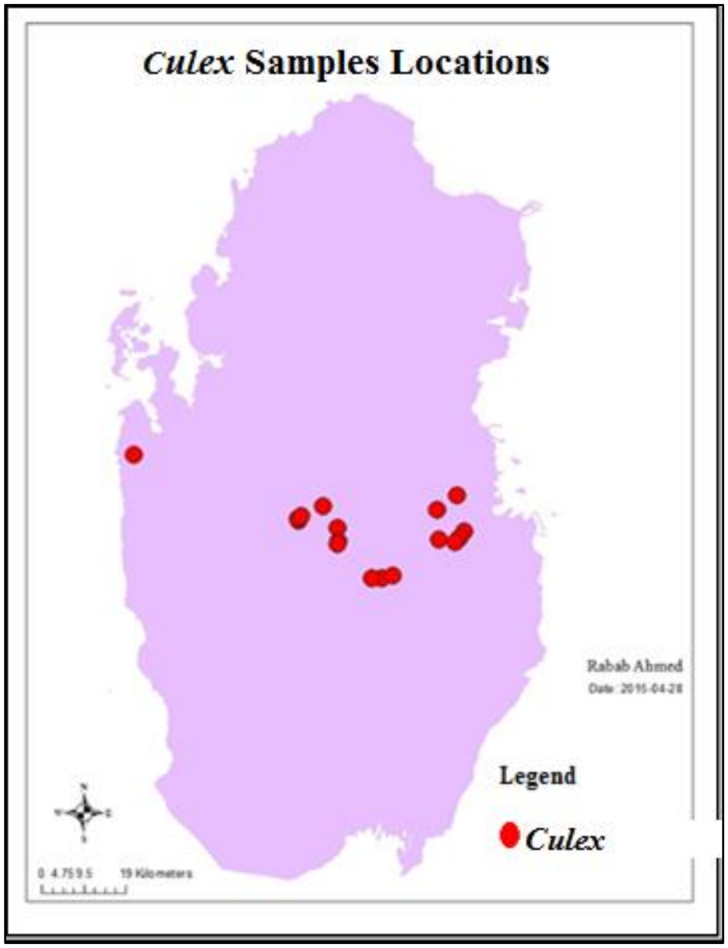


Figure 35: Sampling locations

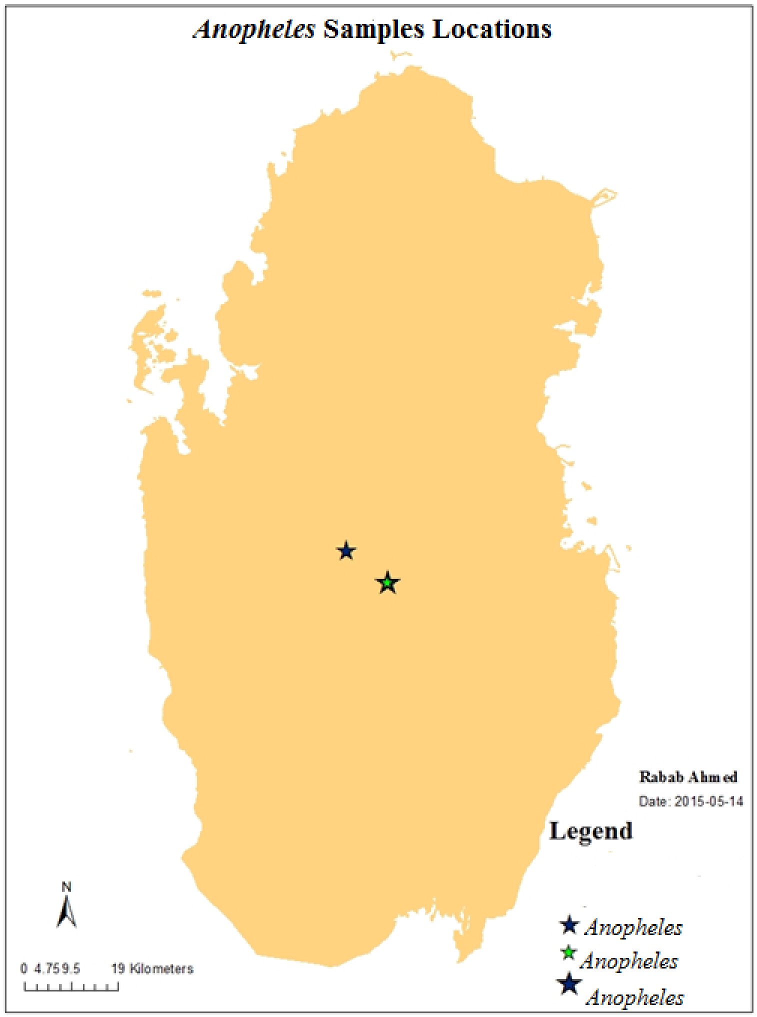


Figure 36: *Anopheles* Distribution in Al Rayyan Municipality

4.2.6 Statistical Analysis

A t- test result is perrformed to determine if there is any relation between types of mosquitoes and the pH value of breeding sites waters (Table 3).

T-Test

Table 3: T-Tes; types of mosquitoes and the pH (tables 1-6)

Group Statistics-1

	Mosquito_Type	N	Mean	Std. Deviation	Std. Error Mean
PH	Culex	49	7.3814	.72699	.10386
	Anopheles	6	8.0533	.13426	.05481

Independent Samples Test-2

		Levene's Test for Equality of Variances		t-test for Equality of Means	
		F	Sig.	t	df
PH	Equal variances assumed	6.790	.012	-2.241	53
	Equal variances not assumed			-5.722	44.969

Independent Samples Test-3

		t-test for Equality of Means			
					95% Confidence Interval of the Difference
		Sig. (2-tailed)	Mean Difference	Std. Error Difference	Lower
PH	Equal variances assumed	.029	-.67190	.29977	-1.27317
	Equal variances not assumed	.000	-.67190	.11743	-.90843

Independent Samples Test-4

		t-test for Equality of Means
		95% Confidence Interval of the Difference
		Upper
PH	Equal variances assumed	-.07064
	Equal variances not assumed	-.43538

Correlations-5

		Qty	PH
Qty	Pearson Correlation	1	-.004
	Sig. (2-tailed)		.975
	N	55	55
PH	Pearson Correlation	-.004	1
	Sig. (2-tailed)	.975	
	N	55	55

Case Processing Summary-6

	Cases					
	Valid		Missing		Total	
	N	Percent	N	Percent	N	Percent
Location * Type of Mosqutoes	31	100.0%	0	0.0%	31	100.0%

4.2.7 Chi-square (X2) Test

Chi-square (X2) test is a nonparametric statistical test to define if the two or more classifications of the samples are independent or not. Furthermore; chi-square tests is one of the most used statistical analyses for replying questions about the relationship or difference between categorical variables. In our case we are trying to examine whether or not the types of mosquitos collected is associated with the areas where they were found.

Table 4 : Chi Square Test (1,2

Location * Type of Mosquitoes Crosstabulation - 1

			Type of Mosquitoes			Total
			1	2	3	
Location	Rural	Count	4	13	0	17
		% within Location	23.5%	76.5%	0.0%	100.0%
		% within Type of Mosquitoes	100.0%	54.2%	0.0%	54.8%
		% of Total	12.9%	41.9%	0.0%	54.8%
	Urban	Count	0	11	3	14
		% within Location	0.0%	78.6%	21.4%	100.0%
		% within Type of Mosquitoes	0.0%	45.8%	100.0 %	45.2%
		% of Total	0.0%	35.5%	9.7%	45.2%
Total		Count	4	24	3	31
		% within Location	12.9%	77.4%	9.7%	100.0%
		% within Type of Mosquitoes	100.0%	100.0%	100.0 %	100.0%
		% of Total	12.9%	77.4%	9.7%	100.0%

Chi-Square Tests - 2

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	6.941[a]	2	.031
Likelihood Ratio	9.580	2	.008
N of Valid Cases	31		

a. 4 cells (66.7%) have expected count less than 5. The minimum expected count is 1.3

5 Discussion

Results obtained from this study, showed that the percentage of adult emerged from larvae samples was lower than that of adult samples even though 18 larvae samples have been collected while 19 samples were collected as adults, this can be explained by the number of larvae which did not developed to adults during the metamorphoses. We are not sure about the exact reasons for that, but cold temperature were observed inside the lab several times in addition to the small size of breeding sites. Temperature is known as an important factor that affect the duration of the mosquitoes development specially the aquatic stages (Norbert *et al.* 2010). Therefore, since mosquito immature stages are cold blooded, their activity depends on the temperature of the water they inhabit. Besides food, temperature is the leading factor that affects the development and growth of mosquito larvae (White 1974 and Norbert *et al*. 2010). Generally, an increase in water temperature will consequence accelerate development of aquatic stages, but will decrease the size of the emerging adults (Bayoh & Lindsay 2003). On the other hand, at higher temperatures fewer adults are produced due to increased mortality (Bayoh & Lindsay 2004).

The number of mosquito specimens collected during the study period was relatively low; this can be justified in many ways. For example, with respect to the temperature effect, the size of breeding sites in the area could be accounted for that, most of larvae samples have been collected from relatively small non-natural breeding sites (Figure 16). Only two locations can be identified as natural sites (shallow ground water), however, samples collected from these locations did not developed into adults. Some of the breeding sites (tanks that store animals drinking water), were filled regularly with water once or twice a week, therefor the aquatic stages of the mosquitoes will be disturbed. In addition, sometimes these tanks are over filled with water which results in the dropping of these aquatic stages outside of the tanks, hence dry out by the heat of the sun (Figure 16). Other breeding sites, were just small containers field with small amount of water (Figure 31), in these types of breeding sites only a few number of larvae has been found and even fewer number has been successfully reared to adult. These encountered habitats have been categorized in attempt to observe and investigate types of mosquitos' habitat available in Qatar. We can divide these habitats into two groups, the first group was small in size and easy to avoid and control, but the public need to be aware and informed about the risk of these breeding sites they are offering. During this survey we had the chance to meet with so many people who know little about mosquitoes habitats. They did not realize the risk of offering mosquitoes a suitable breeding site by leaving only small amount of water sitting for days. The other group of mosquitos' habitat is large sewer ponds used by the government as an area to dump waste water. These habitats are suitable for a range of mosquitos' species, specially *Culex* (Norbert et al. 2010). Only few studies have been carried out on mosquitoes of Qatar. According to these studies (Harbach 1985; Khan *et al*. 2009 and Mikhail *et al*. 2009), eight species have been found in Qatar. However, no data records were provided by the Supreme Council of Health. The outcomes of the present study revealed the presence of one Anopheline and three culicine in addition to genus *Coquillettidia*. From the distribution map (Figure ***33***), it could be assumed that *Culex* is the most common mosquitoes in the study area. No previous records are available regarding the encountered species or their breeding sites as well as their distribution in the surveyed region. However, according to mentioned studies, *Culex* has been found as the most dominant mosquitoes in the country, but different species have been identified, *Culex pipiens* (Linnaeus 1758), *Culex univittatus* (Theobald 1901), and *Culex quinquefasciatus* (Say 1823) (Harbach 1985; and Khan *et al*. 2009). *Culex pipiens* (Linnaeus 1758) has been found as the most dominant mosquitoes. Mattingly and Knight (1956) reported the presence of *Culex laticinctus* (Edwards 1913), *Culex tritaeniorhynchus* (Giles 1901), *Culex sitiens*, *Culex sinaiticus* (Kirkpatrick 1924), *Culex (Lutzia) tigripes* (Grandpre and Charmoy 1900), *Culex pipiens* (Linnaeus 1758) in Saudi Arabia. Wills *et al.* (1985) also recorded *Culex pipiens* (Linnaeus 1758) and *Culex tritaeniorhynchus* (Giles 1901) from the same region. Other studies mentioned that *Culex pipiens* (Linnaeus 1758) complex is highly distributed not only in Qatar but also worldwide where it breeds in pools, canals swamps, water containers and even in any water collection (Mikhail *et al.* 2009). In the present study, the three identified culicine species were *Culex vagans* (Wiedemann 1828), *Culex mimeticus* (Noe 1899) and *Culex Bitaeniorhynchus* (Giles 1901). *Culex vagans* (Wiedemann 1828) have been found as the most dominant mosquitoes in the study area. *Culex vagans* (Wiedemann 1828) inhabit all types of habitats we have encountered, large sewage waste water bonds, construction water tank small leaking sewer water etc. We can assume that, their wide distribution and dominance among other encountered species may be due to its wide range suitability of breeding sites (availability of pools sewage) and high tolerance of variable extremes of temperature. *Culex vagans* (Wiedemann 1828) has been reported in Pakistan, Egypt and Afghanistan (Harback 1985). *Culex mimeticus* (Noe 1899), has been reported in Afghanistan, Egypt, Iran, Irag, Jordan, Labanon, Pakistan, Saudi Arabia, Syria and Turkey while *Culex bitaeniorhynchus* (Giles 1901) has been reported in Iran, Pakistan and Yamen (Harbach 1985).

Identification of *Anopheles stephensi* (Liston 1901), in this survey come with agreement with some of the previous surveys in Qatar, although it has been recorded in different location (Mikhail *et al.* 2009), (was reported in Ain Khalid, in the present survey it was encountered in Rodat Rashid in two different locations). This may give an indication of the distribution of this species as well as its tolerance to Qatar climate. *Anopheles stephensi* (Liston, 1901) has been collected as larvae and adults. Larvae were collected from two small buckets found in the same location. Only nine mosquitoes were emerged from both buckets. The low density of the mosquitoes can be assumed due to the size of breeding sites or lack of food, but in the same time choosing this type of breeding sites indicate the insect behavioral mechanism to overcome difficulties they might encounter in other types of breeding site (avoiding predators and competition for food). Only *Anopheles* has been found in both buckets however, it has been captured associated with *Culex* when using light traps. *Anopheles stephensi* (Liston 1901) has been reported in Afganistan, Bahrain, Iran, Irag, Kuwait, Oman, Pakistan, Saudi Arabia and United Arab Emarat. Although mlaria is not prevalent in Qatar, but it is one of the most regularly imported communicable diseases The presence of such vector put the country at risk of such diseases, especially, since *Anopheles stephensi* (Liston 1901) is well known as primary vectors in the Southwest Asian region (White 1989, Zahar 1974).

Sample locations where divided into urban and rural area, a chi square test were performed to determine if there is a significant difference in the association between mosquitoes types found and the area where they have been captured. Due to the high percentage of the cells that have expected count less than five. The minimum expected count is 1.35. So we checked beside the chi square test the likelihood ratio statistics to compare our results which show a great probability to reject the null hypotheses which stated no association between the two different areas and the type of mosquitoes encountered with p-value 0.008. Which mean we can accept the alternative hypotheses. Precisely, the presence of *Anopheles* species is dependent on the area. This can be explained or supported by the complete absence of *Anopheles* in urban area. On the other hand the t–test was conducted to test if there is no significant difference in the pH concentration between types of mosquitoes (*Culex* and *Anopheles*). According to the results we reject the null hypotheses. However, a correlation coefficient test was conducted as well, test the relationship between number of mosquitoes captured and the pH concentration and we reject the null hypotheses because the p- value was greater than 0.05. But the descriptive measures show that *Anopheles* species have pH measurement higher than that of the other species. In contrast, one study has investigated the pH directing abilities of two members of the mosquito tribe Aedini, which recognized to have intensely diverse saline tolerances. Both species complete larval development in waters fluctuating between pH 4 to pH 11, but naïve larvae always die in water of pH 3 or 12. Acclimation of *Aedes aegypti* to pH 4 or 11 increases survival times in pH 3 or 12, respectively, and allows a small proportion of larvae to pupate positively at these extreme pH values (Clark *et al*. 2004).

Results from the one- month survey reveal that mosquitoes were more present during the evening hours, followed by night time, no mosquitoes have been collected during daytime. According to study in United States spring species flight activity remains high throughout the day compared to other species but peaks at least 1-2 hours before sunset. *Coquillettidia* and *Anopheles walkeri* (Theobald 1901) flight activity appears to reach a plateau of activity from 10 pm to 2 am. Early morning appears to be a significant time during periods of peak *Aedes vexans* (Meigen 1830) activity (Crisp and Knepper 2002). These mosquitoes were collected only in the beginning of the survey period. Regarding the rest of this survey, as stated in the results, no mosquitoes were captured. However, several confounded factors were present at the time of sampling. The samples have been collected during the wet season, thus, several rainfalls and dust storms occurred throughout the period. In addition, one of the pest control companies sprayed the area with some insecticide. It is not regular for such companies to spray the area (residential complex), in the same time this may explain the high density of mosquitoes that have been captured (63 mosquitoes), during only ten days. Sometimes, the complex experience problems in the sewer system, may be the survey has been carried out during one of these times (during a peak in mosquitoes population due to availability of temporally breeding sites). All mosquitoes, from this location have been identified as

Culex vagans (Wiedemann 1828), which as mentioned known to inhabit this type of breeding sites.

Several limitations were encountered during this survey, although all necessary papers and permissions have been prepared. At our request two maps were provided by the Ministery of Environment. These maps contain potential locations for mosquitoes breeding, but no streets or any kind of landmarks were included in the maps which make it impossible to reach the selected areas. Al Rayyan Municipality offer to contact me whenever, a complain is made by one of the companies or resident, but some times we go to the mentioned location and we find that it is already being sprayed. Arrangement has been done with the municipality in order to find a suitable time. In the other hand, these locations were only potential areas, each location was searched completely. Although we have collected only 37 samples, always in each visit, several locations were checked in order to get one sample. During adult's collection, the main obstacle was getting electricity source for the light trap, even though extended wire were used still the sampling were limited to certain areas. Because this type of research never been conducted by government sector before, not even the health sectors, no record regarding mosquitoes types or even no identification key were available.

Information presented in this study would be of a great value towards managing and controlling mosquito's population to minimize their negative impact on human health and economic.

6 Conclusion

This survey provides information on the most dominant mosquitoes in Al Rayyan Municipality, Qatar, only few earlier studies have been carried out regarding this issue. The survey was conducted at Al Rayyan Municipality, Qatar, for a duration of five months (December, 2014 to April, 2015). A total of 37 collection sites were visited throughout the study period (samples include larvae as well as adults), revealing a total of 312 mosquitoes. Four species were identified belonging to three genera namely; *Culex* 91% (3 species); *Anopheles* 6% (one species) and *Coquillettidia* 3% (no species have been identified). The collected species were, *Anopheles stephensi* (Liston, 1901), *Culex vagans* (Wiedemann 1828), *Culex mimeticus* (Noe 1899), and *Culex bitaeniorhynchus* (Giles 1901). The most dominant mosquito genus in the study area was *Culex* which represents 91% of the total samples.

According to the one- month survey, mosquitoes were more present during the evening hours, followed by night time. However, no mosquitoes have been collected during daytime. In addition, temperature has a significant effect on the quantity of mosquitoes collected throughout the day.

The pH was not significantly associated with the types of mosquitos which have been found at the breeding sites. It appears that, the area from which the mosquitoes have been collected have some effect on the presence of certain types of mosquitoes (i.e. *Anopheles* and *Coquillettidia*). The encountered breeding habitats were large swage waste water ponds, construction water tank, animal drinking water tanks, leaking sewer water and small containers all of which are not natural habitats. Most of the breeding sites are easy to avoid or eliminate (except for the swage waste water ponds). Lack of knowledge and awareness are probably the leading reasons of the existence of these kind of breeding habitats.

Since imported malaria has been relatively common in Qatar, due to increasing travel and immigration from malaria-endemic countries, the presence of malaria vector borne diseases (*Anopheles stephensi*), availability of breeding sites, and the presence of individuals who are infected with malaria protozoans (*Plasmodium* spp.), states the risk of Qatar to a reintroduction of malaria.

7 Recommendations

- Incorporation of methods for monitoring of adult mosquito populations into vector surveillance programs for more trustworthy predictions of malaria outbreak in an area.
- Take action to control breeding of mosquitoes in containers or likewise breeding sites at different locations in particularly, home owners
- Inform owners about malaria transmission risk and malaria prevention, and encourage people to take responsibility in order to eliminate breeding sites.
- Improve communication / cooperation between departments of the health sector so that different departments can easily work together.
- Establish an inventory and records data change focusing on people to take responsibility to eliminate breeding sites.
- Set special programs focusing on educating people to cover all water containers.
- Adaptation of legislation is necessary in order to take action against those who refuse to participate in vector control activities and whose behavior can lead to establish mosquito breeding sites.
- Encourage further research and surveys to be carried out throughout the year with the government involvement.

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9 Appendix

9.1 Samples Data

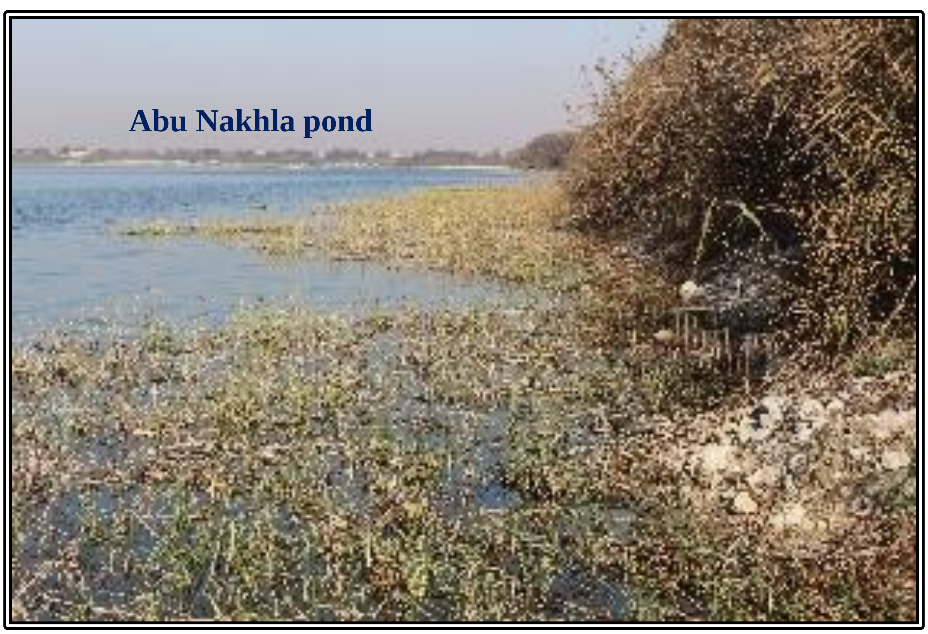
					No Mosquitoes				Mosquitoe types			Culex species		
Sample No	Rural/ Urban	Larvae(L)/ Adults(At)	PH	Positive	Male	Female	Total	# of Genera	Anopheles	Culex	coquillettidia	1	2	Vegans
1	R	L	7.89	N										
2	R	L	8.07	N										
3	R	L	7.95	N										
4	R	L	8.2	N										
5	R	L	6.33	y		3	3	1		3				3
6	R	L	8.6	Y	1	1	2	1		2				
7	R	L	6.2	Y	3	6	9	1		9				
8	R	L	6.8	Y	10	8	18	1		18				8
9	R	L	8.46	Y	5	9	14	1		14				
10	R	L	7.88	Y		2	2	1	2					
11	R	L	8.14	Y	2	5	7	1	7					
12	R	L	7.98	N										
13	R	At	7.21	Y	4		4	1		4				
14	R	At	N/A	N	10		10	1		10				
15	R	At	N/A	Y	2	2	4	2	3	1				
16	R	At	N/A	Y	2	2	4	1		4				
17	R	At	N/A	Y	18	2	20	1		20				
18	R	At	N/A	Y	7	1	8	1		8				1
19	U	At	7.6	Y		1	1	1		1				
20	U	At	N/A	Y	5	1	6	1		6				
21	U	At	N/A	Y	2	5	7	2		5	2			
22	U	L	N/A	Y	20	13	33	1		33			1	12
23	U	At	N/A	Y	2	2	4	2		2	2			
24	U	At	N/A	Y	5	8	13	1		13				13
25	U	At	N/A	Y	6	2	8	1		8				1
26	U	At	N/A	Y	1	3	4	1		4				
27	R	L	N/A	N										
28	R	L	N/A	N										
29	U	L	7.2	Y		3	3	1		3				3
30	U	L	8.05	N										
31	R	At		Y	10	1	11	2	2	9				
32	R	L	8.1	N										
33	U	At	N/A	Y	7	11	18	1		19				11
34	U	At	N/A	Y	1	8	9	1		9				
35	U	At	N/A	Y	25	38	63	1						
36	U	At	N/A	Y	1	2	3	1			3			
37	R	L	7.68	Y	8	16	24	1						
38	R	At	N/A	N										

9.2 One month data

Date	Rainy? Y/N	Evening		Night		Day	
		Qty	Temp	Qty	Temp	Qty	Temp
Mar-10	N	4	26	3	19		28
Mar-11	N	10	27	2	20		30
Mar-12	N	2	22	1	18		25
Mar-13	N	4	24	4	20		26
Mar-14	N	9	22	6	20		26
Mar-15	N	1	22	3	21		25
Mar-16	N	5	24	1	22		28
Mar-17	Y		22		21		25
Mar-18	Y		24		24		27
Mar-19	N		26		24		28
Mar-20	N		26		23		28
Mar-21	N	3	29	1	25		33
Mar-22	N	4	23		21		25
Mar-23	y		23		20		25
Mar-24	Y		22		19		25
Mar-25	N		24		20		27
Mar-26	N		27		22		30
Mar-27	N		25		23		28
Mar-28	N		25		22		28
Mar-29	N		26		20		29
Mar-30	N		27		22		30
Mar-31	N		32		27		37
Apr-01	N		28		25		32
Apr-02	N		25		24		25
Apr-03	N		28		25		30
Apr-04	N		23		21		24
Apr-05	N		22		21		24
Apr-06	N		34		25		37
Apr-07	N		30		26		34
Apr-08	N		30		26		34
Apr-09	N		33		28		36
Apr-10	N		33		27		36
		42		21		0	

9.3 Sewage waste water breading sites

Residential area close to the pond




Al Karaana pond


Al Karaana pond

9.4 Shallow ground water