



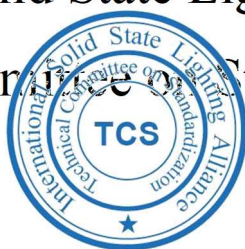
# ISA Technical Report

## LED Mosquito Light Trap and Related Issues

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Technical Committee on Standardization



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Working Group 21: “UV LED mosquito light trapping technology and application”

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## Foreword

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# Title of document

## 1 Introduction

Mosquitoes are one of the six calamities, it can spread zika, yellow fever, malaria, filariasis, dengue fever and other infectious diseases. The number of people threatened by mosquito borne diseases dengue fever reaches 2.5 billion in the world. So anti-mosquito has become extremely important.

Along with the extensive use of pesticides, the malpractice has gradually appeared: the pest propagation speed causes the easy resistant to pesticides, and the use of pesticide caused serious pollution to the environment. People began to seek for this new type of no pesticide control technology to solve the problem of insect, among which using the phototaxis of insect to control is an important attempt in this field. The black light source was firstly used to lure insects. Later, with the development of technology, there emerged some new insect trap light sources, such as high-pressure sodium lamp and LED, which made good use of the positive phototaxis of insects. As an important tool for vector monitoring and control, LED mosquito light trap is widely used. Because the LED has the characteristics of energy saving and environmentally friendly, small size, easy industrial design, low-voltage drive, easy to integrate solar panels, rich wavelength/customized on demand, easy to improve the benefit-harm ratio, strong environmental applicability, and long lifetime. In terms of mosquito control applications, replacing low-pressure mercury lamp is the main trend of market development.

## 2 Scope

This document introduces phototaxis/photophobia response of mosquitoes and requirement/methods for mosquitoes' control in public health, analyzes research of the photoreceptor of mosquitoes and evaluation method of mosquito light trapping and killing performance, and application scene of LED mosquito trap/repellent.

### 2.1 Study on the mosquito Phototaxis / photophobia response

#### 2.1.1 Mechanism of phototaxis of mosquitoes

The phototaxis of insects is mainly a kind of phototaxis reaction that insects produce tendency reaction to a specific spectrum through the photosensitive cells in their visual organs. The visual organs of insects include compound eye monocular: compound eyes are generally composed of many independent small eyes, and small eyes are composed of cornea, crystal cone, lesser omentum cell body, optic rod, cytochrome, and basal membrane. Compound eyes are the main visual organs of insects, located on both sides of the head. Most of them are oval or oblong and occupy a prominent position in the head of insects. The compound eyes of insects are composed of many small eyes, each small eye is an independent photosensitive unit. Monocular, divided into dorsal monocular and lateral monocular, only has a single small eye surface, has the function of photosensitive, but cannot adjust and imaging. Monocular's sensitivity to light is several times higher than compound eye, it can assist compound eye to judge the distance, color, etc., the color wavelength that insects can feel is obviously different from that of humans. Compound eye's sensitivity to light wavelength is wider than that of humans, and its resolution is also different from that of humans. The range of sensing wavelengths varies from 240 nm(ultraviolet) to 650 nm(orange), and most insects cannot feel long wavelengths of red. While humans can see directly

is the visible spectrum (390~700 nm). In other words, insects can not only recognize color, but also can see light waves that the human eye cannot directly see. The visual units of insects can perceive light of different wavelengths, and they can feel polarized light, have high spatial resolution, and can distinguish different shapes, colors, sizes, patterns, etc. Phototaxis is an instinctive reaction formed by insects in the process of long-term adaptation to the environment, and a specific range of light has an irresistible "attraction ". Most species of insects have phototaxis, which plays an important role in the search for food, heterosexual mating, and spawning sites. Different insects have positive and negative tendencies to specific spectrum. We can use the phototaxis of insects to achieve the effect of pest control, this technology is also called light booby-trap technology.

The essence of light is an electromagnetic wave. Because of the different wavelength of light, the characteristics of the corresponding light will be different. The wavelength of light recognized by the human eye is 380 nm~780 nm, and the light in this spectral range is also called visible light. In addition, light waves with wavelengths less than 380 nm are called ultraviolet rays, and light waves with wavelengths greater than 780 nm are called infrared rays. These two types of light wavelengths cannot be recognized by our eyes. Since the eye structure of insects is different from that of human beings, the spectral range of insect recognition is wider than that of human eye recognition. In addition to the visible range, insects can also feel 250 nm~380 nm of light. In addition, due to the different species of insects, there are some differences between their living habits and physiological characteristics, which leads to different insect sensitive light wavelengths.

### **2.2.2 Light source evolution**

The use of light trapping pests began in the 1930s, but because of the strong killing power of the existing insecticidal lamps to natural enemy insects, most agricultural developed countries prefer to use light trapping technology to monitor and forecast the active and omnivorous invasive organisms or nocturnal agricultural pests, while the direct use of light trapping technology as pest control measures in open environment is relatively few. However, with the further restriction of the use of highly toxic insecticides in various countries, the interest in the application of light trapping technology in developed countries has gradually increased in recent years, especially in the research and development of selective light sources based on LED in Japan. as the research progresses, new insect light traps are constantly being developed and improved, such as solar light trap, double wave light trap, LED light trap, etc. the main purpose is to improve the insect light trap from energy saving, the most important is to improve the light spectrum to make it more efficient to trap pests, while reducing the accidental injury to beneficial insect predators and protecting them. As the fourth generation of new light source, LED light trap has injected new power into the development of pest lighting guidance technology. LED light trap has a narrow wavelength range, single light color, high brightness, low energy consumption and long life, which makes it the best light source. The single wavelength LED light source cannot only solve the shortcomings of the traditional light source, such as wide spectral range and poor pest pertinence. Under the same energy consumption, its light intensity is stronger than other light sources, the light coverage area is broader, and the damage control range is wider.

### **2.1.3 Effects of Different Spectra LED Light Sources on Mosquitoes**

In recent years, research on mosquito physical control has been in full swing, exploring that mosquitoes are more sensitive to 360 nm ~ 450 nm wavelengths of light, so at present, in the selection of light sources, the wavelength is mainly in the range of 360 nm~450 nm, but the application of other wavelength range is not excluded.

Michael [40] effects LED different wavelengths of light blue (470nm), green (502nm), red (660nm) and infrared IR (860nm) on pupa and spawning mosquitoes were tested. The results

showed that blue (470nm) and green (502nm) light mosquito habitat box caught the most mosquitoes.

Li [41] effects of CDC traps (365 nm), UV traps (325 nm), black lights (365 nm) and yellow lights (570 nm) on mosquitoes were tested under outdoor conditions. The results showed that the mosquito light trap (365 nm) had the largest number of mosquitoes and the highest proportion of mosquitoes to total insects.

Wilton [42] effect of CDC traps at different wavelengths (290nm, 340nm, 365nm, 390nm, 440nm, 490nm, 540nm, 590nm, 640nm, 690nm, 740nm) was tested, showing something similar to other insects and a significant tendency for 290nm、365nm、390nm lighting.

Mikel [43] effect of CDC traps (red 660 nm、green 570 nm、blue 430 nm and UV 390 nm) of different colors and wavelengths on *Culex pipiens pallens* was tested. The results showed that the trapping effect of 570 nm green light trap was like that of 390 nm ultraviolet light trap .570 nm green light trap has a great application prospect in the detection of blood sucking insects such as mosquitoes.

So far, the study of insect phototaxis has found that different insects have different trends for different light waves and light intensities. With the emergence of LED light source, people began to study the performance of LED light source and found that the light intensity and spectrum of LED light source can be adjusted freely. According to this characteristic, researchers have gradually favored the use of LED light source for insect phototaxis research experiments.

Li Siyu[37] results show, *Culex pipiens pallens* have obvious tendency towards ultraviolet and blue violet light sources, Particularly 365 nm、405nm、420nm and 450 nm, Of which 365 nm and 420 nm light source mosquito trap performance is the most prominent, And there is no obvious difference between them; The lab results show, In a room where people move, The monochromatic LED light source with a wavelength of 420 nm has the best mosquito trap effect, Unlike the results of the study of the unmanned darkroom, The trapping ability of 365 nm light source is obviously lower than 420 nm light source; In addition, This experiment also uses two groups of combined light sources, 405 nm 420nm and 420 nm 450nm, respectively The results obtained are as follows :420 nm >405 nm 420nm nm >405 nm >420 nm 450nm nm >450 Although the results showed that 420 nm light source was the most effective mosquito light trap, But since this paper does not mention whether to use the same power or not radiation power, and whether mosquito's phototaxis to 420 nm light source relate to power or not radiation intensity. These need further experimental studies. The effect on phototaxis is also relatively important.

Liu Pengpeng et al. showed that mosquitoes exhibited photophobia to the light source at 580nm~590nm. According to the review of Jiang Yueli et al., the application of yellow light source to insect control was reported in 2007, and much progress has been made so far. The study of Li Jianquan et al. showed that *Culex pipiens pallens* had an aversive reaction to the irradiation of commercially available yellow sodium light source. In addition, according to the experiment conducted by Ningbo Dayang Technology Co., Ltd., it is showed that *Culex* and *Aedes* exhibited a negative tendency to LED yellow light source around 560nm to 585nm based on the human-arm-in-cage (recommended by WHO2009) test result, the mosquito behavior experimental device test result and the 28m<sup>3</sup> room simulated field experiment test results.

**Table1 WHO human-arm -in -cage test result**

Test people No	Time (min)	Blank control (No of mosquito bite)	Yellow light source ( No of mosquito bite)	Repellent (%)	Notes
1	5	50	32	36.00	With lampshade

2	5	106	75	29.25	With lampshade
3	5	119	38	68.06	With lampshade
4	5	61	29	52.46	With lampshade
5	5	114	42	63.16	With lampshade
6	5	160	87	45.63	Without lampshade
7	5	75	17	75.71	With lampshade
8	5	178	60	66.30	With lampshade
9	2	52	10	80.77	With lampshade

**Table2 mosquito behavior experimental device test result**

Test sample	time (h)	No of mosquito released	Left side ( No of mosquito)	Right side ( No of mosquito)	repellent (%)
Blank control	16	20	3	4	
Yellow light source	16	50	10	7	30.00
Yellow light source	16	30	9	5	44.4
Yellow light source	48	30	11	1	90.90
Yellow light source	16	30	8	4	50.00

**Table3 28m<sup>3</sup> room simulated field test result**





time (min)	Blank control ( No of mosquito bite)	Yellow light source ( No of mosquito bite)	repellent (%)	No of mosquito released
10	53	19	62.96	30
10	18	7	61.11	30
10	65	30	53.85	50
10	25	13	48.00	50
10	16	5	68.75	50
10	92	51	44.57	50
10	53	28	47.17	50

Through preliminary test, QLEDs with different wavelengths provided by Ningbo Institute of Materials Technology and engineering have repellent effect on mosquitoes (Table 1). The specific repellent mechanism, wavelength range and other influencing factors need to be further studied. We also tested yellow light sources, the results showed that yellow light sources with different wavelengths and types have repellent effects on mosquitoes (Table 2). The specific factors such as wavelength and brightness need to be further studied.




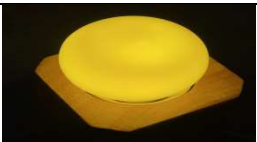
**Table 4 repellent of different wavelength(QLED)**

Sample NO	glow effect	Test method	repellent	repellent in average
YCC1978		arm-in-cage	75.83%	76.12%



				73.81%	
				78.72%	
YCC1983		arm-in-cage		18.20%	59.71%
				68.37%	
				61.90%	
				72.09%	
GCC1986		arm-in-cage		70.90%	55.05%
				38.78%	
				43.18%	
				67.65%	
GCC1980		arm-in-cage		61.29%	68.75%
				55.36%	
				83.33%	
				75.00%	

**Table 5 repellent of different light source and wavelength**

Sample NO	glow effect	Test method	repellent %	repellent in average%
A (1 LED) DP-1		arm-in-cage	43.90%	25.88%
			18.75%	
			32.76%	
			8.10%	
A (3 LED) DP-2		arm-in-cage	20.83%	29.01%
			37.18%	
B (3 LED) DP-3		arm-in-cage	42.86%	44.82%
			54.05%	
			26.67%	
			47.46%	
			52.24%	
			45.61%	
Yellow light source illuminant DP-5		arm-in-cage	47.06%	60.20%
			67.65%	
Yellow light source illuminant DP-6		arm-in-cage	87.50%	69.33%

#### 2.1.4 Advice on wavelength selection for indoor and outdoor mosquito control

The literature shows that the identifiable spectral range of mosquitoes is 250 nm~780 nm, covering ultraviolet and visible light, of which the sensitive spectral range of mosquitoes is 350

nm~450 nm [37-39]. The monochromatic light source of 360 nm~450 nm is more helpful to explore the influence of each wavelength on mosquitoes. In the meanwhile, LED light sources with its unique advantages improve the development of efficient and specific insect light trap. However, because the used environment of outdoor LED mosquito light trap is more complex than that of indoor, such as solar light, temperature, humidity, airflow and so on, it is easy to change, and the trapping effect may easily influence compared with the result of indoor. Therefore, in exploring the outdoor mosquito response to the spectrum, the different bands of spectra should be tested when the rated voltage of the LED light source is 220 V, such as 355nm~360nm, 360nm~365nm, 365nm~370nm, 370nm~380nm, 380nm~390nm, 390nm~400nm, 400nm~410nm, 420nm~440nm, 450nm~455nm. The full spectrum light source is mainly used as the experimental control, and the results of data analysis are collected in different outdoor experimental environments, to establish Whether the sensitive spectrum range of mosquitoes in outdoor environment is the same as that of indoor.

## **2.2 Evaluation of control effect for the LED mosquito light trap/repellent**

### **2.2.1 International standard**

The current international standards are only for the safety requirement of traditional light source, such as IEC 60335-1 Household and similar electrical appliances - Safety - Part 1: General requirements and IEC 60335-2-59 Household and similar electrical appliances - Safety - Part 2-59: Particular requirements for insect killers, not compatible with LED light source. The safety and performance requirement of the LED light source should be standardized.

### **2.2.2 Chinese standard**

The current standards in China related to LED mosquito light trap/repellent lamps are GB 4706.1—2005 Safety for household and similar electrical uses - Part 1: General Requirements(IEC 60335-1 IDT) and GB 4706.76—2008 Household and similar electrical appliances - Safety - Part 2-59: Particular requirements for insect killers (IEC 60335-2-59 IDT). Other standards consist of GB/T 24689.2—2017 plant protection mechanical insecticidal lamps, LY/T 1915—2010 trap lamps, general technical conditions for solar insecticidal lamps in the energy industry, and DB22/T 2281—2015 technical specifications for the use of trap lamps in vegetable fields, etc. These documents are also only for traditional light sources and are not compatible with LED light trap. they do not specify optical parameters (wavelength, irradiance), effect evaluation, benefit-to-harm ratio, and other parameters.

#### **2.2.2.1 Standard of LED mosquito light trap**

For the evaluation of indoor control effect, according to the Chinese national standard GB/T 27785-2011 Laboratory effect determination and Evaluation of Sanitary insecticidal Devices, one mosquito light trap was placed in a square room, around 28 m<sup>3</sup>, the trap is away from the ground 1.5 meters, and 100 female Culex mosquitoes are used. The mosquito killing rate within shall be greater than or equal to 70% in 18 hours. While this rate has poor applicability in practical application: (1) although the indoor dominant mosquito species is Culex pipiens pallens, Aedes mosquito also occupies a large proportion in the indoor and indoor mosquito light trap use scene species (Table 6); (2) although only female mosquitoes bite people, however, the presence of male mosquitoes in ordinary indoor scenes is also intrusive. Therefore, it is suggested that Aedes mosquito should also be regarded as the evaluation object in the evaluation of indoor LED mosquito light trap, and the number of mosquitoes should be half male and female (Table 7); (3) When evaluating the trapping rate, different sizes, trapping methods and different powers of LED mosquito trap have great differences in the trapping rate, so it is suggested to set different levels of trapping rate for evaluation.

**Table 6 Mosquito Species Trapping In Different Site**

Date	site	sample	No of mosquito trapping			other insects
			Culex No(female/male)	Aedes No(female/male)	total No of mosquito	
8.21	corridor (Semi-Outdoor)	Y8	2(1/1)	1(1/0)	3	36
8.21	underground parking(indoor)	16S	4(2/2)	0	4	2
8.21	toilet(indoor)	Y8RK	0	0	0	0
8.21	bedroom(indoor)	16	0	1(1/0)	1	6
8.21	bedroom(indoor)	16	0	1(0/1)	1	2
8.21	bedroom(indoor)	16	0	0	0	0
8.21	living room(indoor)	16S	0	0	0	5
8.21	bedroom(indoor)	Y8	0	0	0	2
8.21	bedroom(indoor)	16S	0	0	0	8
8.21	bedroom(indoor)	Y8E	0	0	0	38
8.21	kitchen(indoor)	16S	3(0/3)	0	3	5
8.21	kitchen(indoor)	16S	3(0/3)	0	3	5
8.22	underground parking (Semi-Outdoor)	16S	9(3/6)	0	9	2
8.22	laundry (Outdoor)	Y8	0	1	1	25
8.22	living room(indoor)	16S	1(1/0)	0	1	5
8.22	living room(indoor)	16S	0	0	0	3
8.22	corridor (Semi-Outdoor)	Y8	1(1/0)	12(6/6)	13	16
8.22	corridor (Semi-Outdoor)	16S	12(4/8)	5(3/2)	12	85
8.22	warehouse(indoor/public)	16S	3(1/2)	0	3	
8.22	bedroom(indoor)	16S	7(3/4)	0	7	25
8.24 ( 36h )	laundry(Outdoor)	Y8	0	2(1/1)	2	20
8.24 ( 36h )	underground parking(indoor/public)	16s	11(3/8)	2(2/0)	13	7
8.24 ( 36h )	office(indoor/public)	Y8				26
8.24 ( 24h )	living room(indoor)	16S	1(1/0)	0	1	10
8.24 ( 72h )	office(indoor/public)	16S	5(5/0)	0	5	12
8.24 ( 36h )	balcony(Semi-Outdoor)	Y8	1(0/1)	0	1	12
8.24 ( 36h )	corridor(Semi-Outdoor)	Y8	1(1/0)	1	2	72
8.24 ( 36h )	warehouse(indoor/public)	16+	1(1/0)	10(8/2)	10	1
8.24 ( 36h )	corridor (Semi-Outdoor)	16S	51(12/39)	(18/6)	75	183
8.24 ( 24h )	yard(outdoor)		1(1/0)	0	1	33
8.24 ( 36h )	bedroom(indoor)		1(1/0)	1	2	
8.24 ( 5h )	kitchen(indoor)		0	2(1/1)	2	2

8.24 ( 24h )	balcony(Semi-Outdoor)	Y8RK	1(1/0)	0	1	5
8.25	outdoor	Y8RK	1(1/0)	0	1	10
8.25	outdoor	16	0	0	0	20
8.25	bedroom(indoor)		0	0	0	4
8.25	outdoor	16s	0	0	7	4
8.25	outdoor	Y8E	0	0	0	31
8.25	outdoor	Y8E	0	1(1/0)	1	106
8.25	outdoor	16S	13	0	13	86

**Table 7 No of Trapping With Different Species And Gender of Mosquitoes**

Date	Test sample	Time (h)	Mosquito species	No of mosquito relesed	No of mosquito trapping	No of mosquito trapping in average
6.10	DYT-09	16h	Culex	30	19	14.6
6.11		16h	Culex	30	13	
6.12		16h	Culex	30	20	
7.7		16h	Culex	30	12	
7.8		16h	Culex	30	9	
6.10		16h	Aedes	30	9	8.2
6.11		16h	Aedes	30	12	
6.12		16h	Aedes	30	12	
7.7		16h	Aedes	30	4	
7.8		16h	Aedes	30	4	
7.3		16h	Female Aedes	30	22	14
7.9		16h	Female Aedes	30	8	
7.10		40h	Female Aedes	30	15	
7.13		16h	Female Aedes	30	11	
7.3		16h	Aedes (half female)	30	20 (11female:9male)	13 ( 8.25female : 4.75male)
7.9		16h	Aedes (half female)	30	9 ( 4female : 5male)	
7.10		16h	Aedes (half female)	30	13 (10female:3male)	
7.13		16h	Aedes (half female)	30	10 ( 8female : 2male)	
7.16		16h	Culex (half female)	30	0	
7.20		16h	Culex (half female)	30	9 ( 4female : 5male)	13 ( 6.5female : 6.5male)
7.21		16h	Culex (half female)	30	17 ( 9female : 8male)	
7.16		16h	Female Culex	30	19	15.6
7.20		16h	Female Culex	30	18	
7.21		16h	Female Culex	30	10	

For the evaluation of outdoor control effect, according to 2.1.4 in the GB/T 23797-2009 Surveillance methods for vector density - Mosquito, we use mosquito density to evaluate the effectiveness of the trap; Or collect the insects caught by the mosquito trap at each point, classify and identify them, and calculate the ratio of mosquitoes to other insects. When the ratio is greater than S, it means that the mosquito trap can effectively trap the target mosquitoes. However, due to the complexity of outdoor application scenarios, the specific evaluation criteria need to be further discussed.

#### 2.2.2.2 Standard of LED mosquito light repellent

As a new attempt in mosquito control method, many kinds of LED mosquito light repellent have come to the market. But for the evaluation of LED mosquito light repellent, there is still on the sketch stage.

The method “human-arm-in-cage” (recommended by WHO2009) is used to test mosquito repellent such as pesticide and essential oils. Mosquito behavior experimental device is widely used in the study of insect’s phototaxis/photophobia response. The room simulated field experiment test is conducted in a 28m<sup>3</sup> room, and release 50 mosquitoes, people sit at the middle of this room and expose two lower legs. Then people count the number of mosquito bite when the yellow light source is turn on/turn off.

## **2.3 Application scene analysis of mosquito LED trap/repellent**

### **2.3.1 Indoor LED mosquito trap/repellent application scene**

#### **2.3.1.1 Bedroom/living room**

In the bedroom, living room, bathroom, balcony and other closed or semi-closed places in the family, it is necessary to consider the safety of mosquito-killing lamps, such as the irritation of light brightness to the human eye, etc. When setting up mosquito-killing lamps, it is also necessary to select mosquito-killing lamps of different sizes and trapping methods according to the actual area, the number of mosquitoes, the location and other factors.

#### **2.3.1.2 Hotel / Public Toilet**

Compared with other indoor scenes, hotels will have relatively dense flow of people, large area, most of the semi-closed places, should use larger LED mosquito trap lights, or according to the layout of multiple mosquito-killing lights, and should pay attention to safety.

### **2.3.2 Application of outdoor LED mosquito trap**

#### **2.3.2.1 Natural villages**

The village which more than 3 people to live in concentrated, at the same time raise pigs, horses, cattle, sheep and other livestock, around the planting of crops, villages or villages with groves (concentrated planting more than 20 trees) and other representative natural villages should be selected for evaluation. It is of great significance for epidemiological risk assessment to investigate the species of mosquito vectors that can be close contact with the population and obtain their density and population composition.

#### **2.3.2.2 Urban public areas**

In urban public areas, mosquito breeding sites are mainly concentrated in green belts, garbage bins, parks, residential areas, etc. In order to prevent mosquitoes and control mosquitoes more pertinently, the correlation of specific environmental factors should be explored, especially in parks and other mosquito breeding areas in urban hot spots.

#### **2.3.2.3 Orchard farms and production areas**

Agricultural pests monitor and control with light trap is an important measure for comprehensive pest management. The use object of light trap is not only for mosquitoes, but also for other agricultural and forestry pests. The light trap can not only trap a large number of pests, reduce their mating probability and fall eggs in the field, but also monitor the population dynamics of pests and provide the application of insect condition prediction and prediction (monitoring, killing, driving, etc). Therefore, when the LED light is induced in this field, the species of pests monitored simultaneously by light trapping and field investigation should be counted, and the annual variation law and correlation of the number of light trapping and field investigation should be analyzed.

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## Appendix A

### Overview of the Physiology and Hazards of Mosquitoes

#### A.1 General

Mosquitoes belong to the family insects, Diptera, mosquito idae. Small insects, 0.5~1.5cm long and slender antennae. The mouthpart forms a long beak, and the beak of a female mosquito is generally suitable for pierce and sucking blood. Wing veins are special, namely the leading-edge veins around the whole wings, the fourth longitudinal vein (midvein) two branches; both the wing veins and wing edge have scales. Mosquitoes are closely related to people's life and health of health pests, but also one of the "four pests ", with a variety of species, wide distribution, rapid reproduction characteristics [1].On the one hand, they affect people's lives through bites, harassment and so on; on the other hand, they are vectors of a class of important transmission diseases, which can spread many diseases, including dengue fever, malaria, typhoid fever, plague, filariasis and so on, and are of the factors that seriously threaten people's lives and health in the world at present [2,3] .At present, there are more than 3500 species of mosquitoes and their subspecies in 37 genera ,119 subgenus, and common of Culex, Anopheles and Aedes[4].

#### A.2 Physiological overview of the Mosquito

##### A.2.1 Classification of mosquitoes

The morphological characteristics of the three genera anopheles (Anopheles), culex (Culex) and aedes (Aedes) are shown in table 1 .At present, the mosquitoes that need to be controlled are such as Anopheles sinensis(*Anopheles sinensis*), *Anopheles lesteri*, *Anopheles dirus*, *Anopheles minimus*, *Culex pipiens pallens*, *Culex quinquefasciatus*, *Culex tritaeniorhynchus*, *Aedes albopictu*), *Aedes aegypti*[5].

**Table A.1 Morphological characteristics of different species of mosquitoes**

characteristic		Anopheles	Culex	Aedes
adult mosquito	wing	Black and white spots	Generally spot - free	immaculate

	antennae	Male mosquitoes are longer than beaks, extremity rod-like, female mosquitoes are as long as beaks	Male mosquitoes are as long or longer as beaks, extremity are tip and thin, female mosquitoes very short, beak 1/6-1/4	Same as Culex pipiens
	body colour	Most are grey and spot - free	Mostly brownish yellow or tan	Mostly black, with white spots or stripes
	Posture of stay	The body and the beak form a straight line, with the stay flat into an angle	The beak is angled to the abdomen, and the body is parallel to the surface	Same as Culex pipiens
	hematophagia	More at night	More at night	More daytime
ovum	Shape and arrangement	Spread out, floating on the surface of the water, boat-shaped, both sides have float	Gathering like rafts, floating on the surface of the water, individual ovum are conical	Single, often submerged under water, olive shaped
larva	Breathing tube	No, but a pair of breathing holes	Thin and long	Rough and short
	Abinal brown hair	yes	no	no
	statics	The body is parallel to the water surface	The tail is near the surface of the water, the head droops, and the body forms a angle to the water surface	Same as Culex pipiens
	habitat	More in clear water, such as ponds, rice fields, streams, etc	More in sewage and field water, such as sewage pits, tanks, ponds, etc	Same as Culex pipiens
aurelia		Thick, short but mouth wide	Slender and small mouth	Same as Culex pipiens

## A.2.2 Ecological ecological habits of of mosquitoes

### A.2.2.1 Growth cycle of mosquitoes

The life of mosquitoes passes through four periods: adult mosquitoes, ovum, larvae, and pupae, which belong to complete metamorphosis development. Larvae and pupae live in water, but adult mosquitoes live out of the water environment.

Mosquitoes belong to complete metamorphosis. There are four insect states: ovum, larva, pupa, and adult. The first three species live in water and adult mosquitoes live on land. Female and male mosquitoes lay eggs after mating and female mosquito blood sucking, and the morphology, mode, place and number of fertilized eggs vary according to species, mosquito eggs are boat-shaped, single in large clean water body, floating on the water surface; Culex eggs are conical and produced in sewage water body, cluster lump of egg and float in water surface; Aedes eggs are olive shape, single in small clean water, sink bottom; a few species of ovum sticky under the

leaves of aquatic plants. Most kinds of egg embryos mature, hatching larvae, egg development depends on mosquito species and water temperature. Larvae are usually hatched in 2 days in summer, but some *Anopheles* eggs cannot hatch for more than 10 days or even 1 month under low temperature and wet conditions.

The larvae pupate after 4 times of molting. Mosquito pupae do not eat, often floating on the water or diving into the water. Summer pupa usually breaks from the back after 2 ~ 3 days and feathering into adult mosquitoes. Mosquitoes are 1 ~ 2 days after feathering and often mate before not sucking blood. In general, female mosquitoes only suck blood to lay eggs and reproduce offspring. From laying eggs to developing into adult mosquitoes, the time required to complete the life history varies according to the living conditions of mosquito species, temperature food and so on. The *Culex pipiens pallens* about 2 weeks at 23~26°C, *Anopheles sinensis* at 28 ~ 30°C about two weeks, and *Aedes albopictus* at 28°C about two weeks. Under suitable conditions, mosquitoes can reproduce 7~8 generations a year [5,6].

#### **A.2.2.2 Mosquito habitat**

The mosquito larvae all live in water bodies. Different water quality produces different species of mosquitoes. When female mosquitoes lay eggs, there is a certain selectivity for water accumulation. According to the perspective of control, media mosquitoes can be divided into 5 types [5];

- a) Rice field type is mainly born in paddy fields, marshes and other large water, *Anopheles sinensis*, *Anopheles anthropophagus*, *Culex there-banded* mosquito;
- b) Slow-flow type is mainly born in streams, ditches and other slow-flow, *Anopheles minus*;
- c) Jungle type is mainly born in the jungle shady stone cave and other small water, *Anopheles dirus*;
- d) The sewage type is mainly found in polluted containers or small water, causing *Culex pipiens pipiens* and *Culex pipiens pallens*;
- e) The container type is mainly found in household containers or plant containers, *Aedes albopictus* and *Aedes aegypti*;
- f) Adult mosquito habitat, due to different species and environment, generally divided into three categories;
- g) Domestic habitat habits mostly in the room, livestock house shelter during the day. After blood absorption still stay indoors, for stomach blood digestion, ovary mature, fly outside to find a suitable place for spawning. Such as *Culex pipiens pipiens* and *Culex pipiens pallens*;
- h) Half - dwelling mosquito species have both the habit of internal and external habitat, after blood absorption immediately or slightly stay indoors, fly out of the outdoor habitat. Such as *Anopheles anthropophagus*, *Culex tristrata*;
- i) Wild habitat types bloodsucking and spawning are in the wild, such as *Aedes albopictus*. But this classification is not absolute, and even the same mosquito will change its habitat type with different seasons, regions, or environments. For example, *Anopheles minus* is recognized as a domestic mosquito species, but it has been found in some uninhabited mountain forests in Taiwan Province and Hainan Province, while in Guangxi, Yunnan, Guizhou and other places.

### A.2.3 An overview of mosquito infestation

Mosquito vector control is an important task of public health. About 8.3 million people die from mosquito-borne diseases each year as a result of mosquito bites, making them the number one killer of all animals. The study found that mosquitoes carry more than 530 viruses, of which more than 100 can cause disease. In recent years, the virus found in mosquitoes in China has such as the Gaeta virus, Chikungunya virus, (CHIK) 、 Sindbis virus, (SINV) 、 Liaoning virus, (LNV) 、 Western equine encephalitis virus, (WEEV) 、 Ross River Virus, (RRV) 、 Epimodovirus、 Batai virus, (BATV) 、 Banna virus, (BAV) 、 Kadipiro (KDV) 、 Yunnan ring virus、 Colti、 Desonucleosis virus, (DNV) .Among the major mosquito-borne diseases are malaria, dengue fever, Zika virus, yellow fever, Japanese encephalitis and filariasis [8,9].

Malaria, a disease transmitted by Anopheles mosquito, is infected by the bite of Anopheles mosquito. The main clinical symptoms are paroxysmal shivering, high fever, sweating, splenomegaly, blood spray and other [10,11]. Malaria is one of the leading causes of human death worldwide. Infants and young children are the main parasitic [11] of malaria. In the 1930s, a World Health Department survey showed that in areas where malaria is endemic around the world, half of the residents have malaria parasites in their blood, and 72 percent of these residents have malignant malaria. Approximately 198 million malaria patients and 584,000 deaths were worldwide in 2013[11,12].

Dengue fever, a endemic of acute infectious diseases caused mainly by the transmission of dengue virus by Aedes aegypti or Aedes albopictus [13] .Patients infected with dengue fever develop high fever, severe pains, muscle and joint pain, hemorrhagic shock, thrombocytopenia, blood concentration, and death with severe infection [14].During the period 1978-2010 in China, a total of 734637 cases of dengue fever and 541 deaths were reported [15,16] .The number of dengue infections in Singapore reached 11300 between the beginning of 2016 and September.

Epidemic encephalitis B is a zoonotic acute viral infectious disease transmitted by mosquitoes. The mortality rate is high, mainly through the Culex tritaenata to spread [17]. A virus can damage the patient's central nervous system, but also high fever, alertness, coma and other symptoms, severe spastic paralysis or even death [18]. As high as 20 per 100,000 in the 1950s, the incidence was effectively controlled after the 21st century, basically under 1 per 100,000, and showed a continuous downward trend [19,20].

Yellow fever is an acute infectious disease caused by yellow fever virus.spread by Aedes Mainly popular in Africa and Central and South America, clinical features are fever, shivering, headache, bleeding, proteinuria.

Filariasis is caused by Wuchereria bancrofti or Brugia malayi that they parasitize in human bodys [21]. Children with microfilariae are the main source of infection of filariasis, the Bain filariasis virus is mainly transmitted by Culex pipiens, and the Malay filariasis virus is mainly transmitted [22] Anopheles. Before prevention and treatment, there were as many as 30.994 million filariasis

patients in China, including 21.962 million in Bain filariasis patients and 9.032 million in Malay filariasis patients [23].

The Zika virus is transmitted mainly through Aedes mosquitoes. Its clinical symptoms are fever, spotted papules, joint pain, nonsuppurative conjunctivitis, etc. We found that the first record of Zika virus disease occurred in 185 cases in the Micronesian Islands in the Western Pacific Ocean in 2007. French Polynesia has had tens of thousands of infected people since October 2013. In May 2015, Brazil found the first local infection case, after which the outbreak continued to ferment and had a massive outbreak of [24]. Specifically, pregnant women have a certain chance of congenital defects such as [25].

### **A.3 Requirements and Methods of Mosquito Control in Public Health**

#### **A.3.1 Public health mosquito prevention and control requirements**

"Healthy China 2030" program outline, for the next 15 years of China's health development pointed out the direction. The third chapter-the seventh chapter-the first section of the prevention and control of major diseases: strengthen the prevention and control of major infectious diseases; the fifth chapter-the thirteenth chapter-the second section: vector biological control level is an important index of healthy cities and healthy villages and towns. The program highlights the importance of monitoring and control of vector organisms such as mosquitoes, and effectively responds to the epidemic situation of key infectious diseases such as influenza, hand, foot and mouth disease, dengue fever, measles and so on. Continue to adhere to the source of infection control of schistosomiasis comprehensive control strategy. Continue to consolidate the national achievements against malaria. Strengthen the source control of major animal-derived infectious diseases.

At present, mosquito vector control advocates comprehensive control strategies, including environmental control, chemical control, biological control, physical control, genetic control, regulatory control and other methods. Among them, environmental control is to eliminate mosquito breeding environment through environmental control; chemical control is to eliminate mosquitoes through the use of chemical insecticides; biological control is to control mosquito density through the use of biological insecticides or natural enemies of mosquitoes; physical control is to control mosquitoes by using physical methods such as mosquito nets, electric mosquito traps, Dquito mosquito lamp; genetic control is to reduce mosquito density by changing the genetic characteristics of mosquitoes; regulatory control is to control mosquitoes through a series of rules and regulations, multi-sectoral joint action strategies; other control measures include health education to reduce mosquito hazards. In the implementation of mosquito control, the key problem is how to scientifically choose the method of mosquito control to achieve the ideal mosquito control effect.

#### **A.3.2 Methods of mosquito control**

##### **A.3.2.1 comprehensive ecological improvement**

Reducing mosquito breeding environment through environmental control is very important for mosquito control. Consumers have insufficient understanding of the importance of mosquito

prevention and mosquito control and related popular science knowledge. Health education and popular science propaganda should be popularized. In addition to personal protection, the breeding environment of mosquitoes in life should also be eliminated. For example, the water storage along the vegetable altar is regularly replaced or salted or covered, the water accumulation problem of the pot pad plate, the water accumulation problem of the waste bottle and tank, the water accumulation problem of the rubber bowl in the rubber forest during the idle period, the landfill of the small useless water body, etc. Environmental governance is not only a matter of relevant functional departments, but also should mobilize the masses and the whole people to participate to achieve the desired results.

#### **A.3.2.2 chemical pest control**

Chemical insecticides are easy to use, effective, strong, and lasting, especially when the disease epidemic can play a rapid role in mosquito control, disease spread. In addition to the traditional four-generation insecticides (organochlorine, organophosphorus, carbamate and pyrethroids), organofluorine insecticides, nicotinamide insecticides, pyrrole insecticides and trioxyclohexane insecticides, insect growth regulators, insect sterility agents, insect attractants, phytogenic insecticides (such as nicotinic, neem, etc.) and synthetic insecticides based on the chemical structure of natural substances or similar derivative structures have received extensive attention and improved the effectiveness of chemical control. The application of new epidemic prevention vehicle, motorcycle type and UAV type spraying equipment have greatly improved the spraying efficiency of chemical insecticides and solved the problem of drug spraying in special environment. However, due to the long-term spread of chemical insecticides, mosquito resistance to chemical insecticides is becoming more and more serious and widely [26], coupled with the harmfulness of chemical insecticides to humans and animals and the pollution of the environment, people should think about new insecticidal strategies.

#### **A.3.2.3 biological pest control**

Biological control is the direct or indirect application of natural enemies that produce or do not produce metabolites to control pests, including human disease vectors. At present, the main pathogens (bacteria, fungi, viruses, etc.) and predators are used in mosquito vector biological control. Because biological control has many advantages, such as strong action specificity, harmless to non-target and beneficial organisms, no pollution of the environment, not easy to produce resistance, some biological control substances can be recycled themselves, have a long effective period, can save a lot of manpower and material resources.

Biological prevention system includes the use of biological pesticides and the rational application of mosquito natural enemies. The most widely used biopesticides are B. Bti (Bs) and Bacillus (Bs), The application of biopesticides in the United States has accounted for 50% of the market share, the application of biopesticides in China is relatively few, but it has also shown an increasing trend in recent years. Relevant research institutions such as the United States and India are using slow - release technology to develop long - term biological pesticides, which can improve the mosquito control effect of biological pesticides and prolong their efficacy time.

Applications of mosquito natural enemies include nematodes (such as insectivorous *Rosso* nematodes), fish, insects (such as dragon lice, pine algae, dragonflies, giant mosquitoes, turboworms, etc.), domestic ducks, shrimp, mosquito removal plants, etc. The latest research has found that transgenic technology lets fungi carry toxic genes that can effectively kill mosquitoes and reduce the mosquito density [27].

#### **A.3.2.4 Mosquito vector transformation**

At present, the transformation of mosquito vector mainly focuses on four hot areas: transgenic mosquito, innate immune mechanism of mosquito, development of blocking vaccine, and study of intestinal flora of mosquito.

With regard to the study of transgenic mosquitoes, insect sterility (sterile insect technique, SIT) can be used to make transgenic male mosquitoes mate with wild female mosquitoes to produce only male offspring, or to improve the ability of mosquito pathogens (such as the expression of antimicrobial peptides, pathogen resistance factors, etc.) by introducing exogenous genes, thus reducing the of mosquito transmission ability[28,29].

Studies on the innate immune mechanism of mosquitoes have found that mosquitoes can inhibit the development of related pathogens in mosquitoes through Toll, Imd, JAKSTAT and JNK signaling pathways and melanization mechanisms [30-33]; Understanding the innate immune mechanism of mosquito against pathogens can help to develop the effect of transmission blocking by changing the immune ability of mosquitoes to pathogens.

In the development of blocking vaccine, can be the way of immunization make people or livestock for mosquito digestive enzymes (such as trypsin) or pathogens in a development stage of antibodies, after mosquito bite blood, such antibodies can block the mosquito digestion enzymes to affect the nutritional absorption and inhibit the development of the pathogen in the mosquito, achieve the mosquito effect and block the effect of disease transmission.

On the study of mosquito intestinal flora, the composition of mosquito intestinal flora was grasped by high-throughput sequencing technology, and then the symbiotic bacteria were used to express proteins that blocked the development of related pathogens, thus reducing the transmission ability of mosquitoes. Or by feeding *Wolbachia* and other bacteria to block the transmission of certain diseases [34].

#### **A.3.2.5 Physical control**

Physical control is the use of mosquito nets, electric mosquito nets, mosquito light trap and other physical methods to control mosquitoes, because of its environmental pollution and harmless to people and animals and other advantages, but also widely used in people's lives. The wing vibration of female mosquitoes attracts male mosquitoes to mate with them. Although this method does not directly reduce the number of mosquitoes, it significantly reduces the mating rate of adult mosquitoes, which can indirectly lead to a decrease in the number of mosquitoes. In addition, because the female mosquito will escape quickly when it hears the wing vibration of the same male mosquito again after mating, it can imitate the wing vibration of the male mosquito to interfere with the female mosquito after mating to find the host. The use of mosquito nets is

an important measure in malaria control strategies in epidemic areas. With the integration of more and more high-tech and Internet technologies in mosquito control, we look forward to the successful development of new and efficient physical anti-mosquito devices for alternative applications (such as infrared detection and laser anti-mosquito devices funded by the Bill Gates Foundation).

Mosquitoes react differently to different wavelengths of light. Ultraviolet light, especially near ultraviolet light, has strong attraction to mosquitoes, while yellow light has repellent effect. According to these special physiological reactions of mosquitoes, many mosquito repellents and mosquito light traps have been developed. Physical control is directly aimed at the physiological habits of mosquitoes and will not cause damage to the environment and other organisms, so it is worth popularizing [35].