HOUSE FLY, *Musca domestica* L. (DIPTERA: MUSCIDAE), INDICES AND INSECTICIDE RESISTANCE PROFILES IN POULTRY FARMS IN PENANG

BONG LEE JIN

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by

BONG LEE JIN

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LIST OF ABBREVIATIONS

a.i	active ingredient
°C	degree Celsius
LD	lethal dose
mm	millimeter
cm	centimeter
m	meter
km	kilometer
m^2	meter square
d	day
♂	male
♀	female

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INDEKS DAN PROFIL KERINTANGAN INSEKTISID LALAT RUMAH, *Musca domestica* L. (DIPTERA: MUSCIDAE) DI LADANG AYAM PULAU PINANG

ABSTRAK

Suatu kajian telah dijalankan untuk mengkaji perubahan bulanan indeks lalat dan profil kerintangannya terhadap insektisid dalam ladang ayam. Maklumat yang diperolehi boleh diguna untuk merangkakan suatu strategi kawalan lalat rumah yang berkesan.

Perubahan index lalat rumah, *Musca domestica*, telah dikaji sepanjang satu tahun di dua ladang ayam yang terletak di Pulau Pinang: satu di Balik Pulau pada kawasan pulau dan satu di Juru pada kawasan seberang. Persampelan lalat dijalankan dari Mac 2007 hingga April 2008 dengan menggunakan 'Scudder grill'.

Di Balik Pulau, puncak index lalat sebanyak 51.00 ± 11.51 dicatatkan pada bulan Februari 2008. Index lalat menunjukkan hubungan negatif terhadap kelembapan relatif dan jumlah taburan hujan. Namun, tiada korelasi signifikan di antara index lalat dengan factor cuaca tersebut. Di Juru, puncak index lalat mencatat jumlah sebanyak 41.17 ± 6.52 pada bulan Mac 2008. Index lalat di Juru berkorelasi rapat dengan kelembapan relatif ($r = 0.803$, $p < 0.05$) dan jumlah taburan hujan ($r = 0.731$, $p < 0.05$). Namun, suhu didapati tidak mempunyai pengaruh yang signifikan terhadap index lalat di kedua-dua lokasi tersebut disebabkan oleh perubahan suhu yang tidak ketara. Tahap sanitasi dianggap sederhana baik di sepanjang tahun.

Tahap kerintangan lalat terhadap insectisid DDT, malathion, propoxur, dan permethrin dinilai dengan menggunakan aplikasi topikal. Di kedua-dua lokasi, tahap kerintangan lalat terhadap insektisid menurun mengikut turutan propoxur > malathion > DDT > permethrin. Nisbah kerintangan (RR) strain Balik Pulau bagi propoxur,

malathion dan DDT masing-masing berada dalam lingkungan 10.28 hingga 99.00, 7.83 hingga 47.01 dan 6.05 hingga 31.10. Manakala, nisbah kerintangan (RR) strain Juru masing-masing berada dalam lingkungan 5.58 hingga 83.38, 15.19 hingga 27.82, dan 10.04 hingga 22.69. Permethrin merupakan insektisid yang paling berpotensi terhadap lalat di Balik Pulau (RR = 0.50 hingga 1.96) dan Juru (RR = 0.64 hingga 2.40). Perubahan aras kerintangan lalat terhadap insektisid dipengaruhi oleh profil applikasi insektisid.

Lalat rumah strain makmal jantan mepunyai jangka hayat pendek (19.83 ± 1.58 d) yang signifikan ($p < 0.05$) berbanding dengan kedua-dua lalat rumah strain Balik Pulau (30.95 ± 0.69 d) dan strain Juru (28.77 ± 0.52 d). Walaupun tiada perbezaan signifikan antara lalat rumah betina bagi ketiga-tiga strain, purata jangka hayat bagi lalat rumah betina strain Juru (37.79 ± 0.42 d) adalah tinggi sikit berbanding dengan strain Balik Pulau (33.99 ± 1.71 d) dan strain WHO (33.05 ± 1.43 d). Strain Juru menunjukkan tempoh oviposisi yang terpanjang (24.30 ± 0.52 d) dan berbeza dengan signifikan ($p < 0.05$) apabila berbanding dengan strain WHO dan Balik Pulau. Variasi ini mungkin disebabkan oleh jangka hayat yang berbeza antara strain. Tempoh oviposisi yang panjang dalam strain Juru mungkin mengganggu penghasilan telur (288.74 ± 49.30) yang banyak oleh strain Juru berbanding dengan strain Balik Pulau (283.52 ± 25.17) dan strain WHO(138.70 ± 57.65). Walaubagaimanapun, tiada perbezaan signikan terdapat antara strain.

HOUSE FLY, *Musca domestica* L. (DIPTERA: MUSCIDAE), INDICES AND INSECTICIDE RESISTANCE PROFILES IN POULTRY FARMS IN PENANG

ABSTRACT

The present research was conducted to study the monthly changes in fly index and the insecticide resistance profiles in poultry farms. The information gathered can be used to formulate an effective control strategy for house fly.

Changes in the index of the house fly, *Musca domestica*, was studied for a period of one year in two poultry farms in Penang, Malaysia: one in Balik Pulau, located on Penang Island, and the other in Juru, located on mainland Penang. The sampling of house flies was carried out from March 2007 to April 2008 by using the Scudder grill.

In Balik Pulau, the fly index recorded a peak of 51.00 ± 11.51 flies in February 2008. The fly index showed an inverse relationship to relative humidity and total rainfall. However, no significant correlations were found between the fly index and the above mentioned climatic factors. In Juru, the fly index reached a peak of 41.17 ± 6.52 flies in March 2008. The fly index showed strong correlation with relative humidity ($r = 0.803$, $p < 0.05$) and total rainfall ($r = 0.731$, $p < 0.05$). However, temperature had no significant effect on the fly index in both poultry farms due to imperceptible changes in monthly temperature. The sanitation levels in both poultry farms were considered average throughout the year.

The resistance level of house flies was evaluated against DDT, malathion, propoxur, and permethrin using the topical application method. In both poultry farms, the house fly resistance to insecticide decreased in the order of propoxur > malathion > DDT > permethrin. The resistance ratio (RR) of Balik Pulau strain of house flies for

propoxur, malathion and DDT was in the range of 10.28 to 99.00, 7.83 to 47.01 and 6.05 to 31.10 respectively, whereas in the Juru strain, the RR was in the range of 5.58 to 83.38, 15.19 to 27.82, and 10.04 to 22.69 respectively. Permethrin appeared to be a highly potential insecticide against house fly in both the Balik Pulau (RR = 0.50 to 1.96) and Juru poultry farms (RR = 0.64 to 2.40). The fluctuations of house fly resistance level against insecticides were influenced by insecticide application profiles.

Laboratory strain of male house flies significantly ($p < 0.05$) possess a shorter life span (19.83 ± 1.58 d) compared with both Balik Pulau (30.95 ± 0.69 d) and Juru (28.77 ± 0.52 d) strain of house flies. Although no significant differences were found among the 3 strains of female house flies, mean longevity of the Juru strain female house flies (37.79 ± 0.42 d) was slightly higher than the Balik Pulau (33.99 ± 1.71 d) and WHO strains (33.05 ± 1.43 d). The Juru strain showed the longest oviposition period (24.30 ± 0.52 d) and significantly different ($p < 0.05$) when compared with the WHO and Balik Pulau strains. These variations may be attributed to the different female adult longevity among the strains. The longest oviposition period in the Juru strain may have influenced the substantial egg production by the Juru strain (288.74 ± 49.30) compared to Balik Pulau strain (283.52 ± 25.17) and WHO strain (138.70 ± 57.65). Nevertheless, no significant difference was shown between the strains.

CHAPTER ONE

GENERAL INTRODUCTION

Flies, belonging to the Order Diptera, are among the most familiar of insects and are recognized by the possession of only one pair of wings. The second pair of wings is reduced into halteres, which act as gyroscopic stabilizers during flight (Curran 1946). There are 150,000 different species of flies distributed throughout the world, and many more are being discovered every day (Wiegmann and Yeates 2007). All flies show complete metamorphosis, with the life cycle consisting of four phases: egg, larva, pupa and adult.

Filth flies refer to several species of true flies belonging primarily to the families Muscidae, Calliphoridae and Sarcophagidae (Pont and Paterson 1971). These flies are non-bloodsucking flies with mouthparts that are evolved for "sponging" up any liquid food (Oldroyd 1964). Filth flies are very strong fliers, with house fly capable of flying 8 kilometers per hour for several kilometers. Developmental sites can be a long distance away from areas where the adults are causing problems (Hogsette and Farkas 2000).

Filth flies are known as ‘synanthropic’ flies, which flourish particularly in association with man and attracted to foul substances, hence have frequent opportunity to transmit pathogenic organisms mechanically (West 1951). Graczyk et al. (2001) stated that synanthropic flies are major epidemiologic factors responsible for the spread of acute gastroenteritis, trachoma among infants and children in developing countries and transmission of multiple antibiotic-resistant nosocomial bacteria in hospital environment.

Filth flies have been of major medical concern during military exercises and operations conducted in warm weather. Fly problems may develop around field facilities that have inadequate screening, resulting in unsanitary conditions that make it difficult to protect against fly-borne diseases (AFPMB 2006). Moreover, filth flies are responsible for considerable financial losses due to agitation among livestocks. Fly breeding in accumulations of poultry excrement has become a great problem in modern egg and chicken industry, therefore reducing farmers' profit margins (WHO 1986c).

House fly, *Musca domestica*, is the best known and notorious of all filth flies. It is a cosmopolitan synanthropic insect of medical and veterinary importance (Greenberg 1971), which are capable of transmitting infectious organisms mechanically. It is also capable of transmitting pathogens by regurgitation of its vomitus and by passage through its alimentary tract (faecal-oral route) (West 1951). Ara and Marengo (1932) proved that 44% of the flies captured in rooms of typhoid patients were carrying typhoid bacillus, which remain alive in the alimentary tracts for as long as six days after capture. Moreover, further studies done by Holt et al. (2007) indicated that the entire house fly captured from a poultry farm, where *Salmonella* serovar Enteritidis-infected chickens are kept, were carrying *Salmonella enterica* serovar Enteritidis in the guts.

According to Esrey (1991) and Cohen et al (1991), diarrhoea cases apparently increased when high population of flies was recorded. Thus, information about the relative density of a species would suggest a particularly advantageous time to apply control. It is vital to study the temporal index of house fly and general pattern of population growth before developing any control strategies against house flies.

After 1944, with the development of chlorinated hydrocarbon insecticides, house fly was satisfactorily controlled (Chapman and Morgan 1992).The widespread use of DDT in the 1940's caused house fly to develop resistance to DDT. Since then, house fly has been found to develop resistance to other chlorinated hydrocarbon insecticides

(Busvine 1963). In the mid 1950s, organophosphorus and carbamate insecticides were used for fly control, followed by synthetic pyrethroid insecticides in 1973 (Chapman and Morgan 1992). Unfortunately, house flies greatly developed resistance to every insecticide used against them by encountering selection pressure from frequent treatments, particularly with single products.

Insecticide resistance is a major problem in controlling agriculturally and medically important insects especially house flies. Due to the high resistance of house flies to various insecticides, it is vital to assess the monthly resistance status of house flies to assist the authorities in selecting insecticides for successful fly control and implementing an Integrated Pest Management system to minimize insecticide resistance.

The first survey on house fly in Malaysia was carried out in 1952 by Pagden and Reid (1952). Studies on the temporal changes of insecticide resistance and index of house flies in Malaysia are scanty. To date, limited information is available on the resistance status and index of house fly in poultry farms of Northwestern Peninsular Malaysia.

Therefore, the objectives of this study are:

1. To study the temporal changes of *M. domestica* index in poultry farms in Penang.
2. To study the temporal changes of the resistance status towards chlorinated hydrocarbon, organophosphate, carbamate, and synthetic pyrethroid insecticides in *M. domestica*.
3. To study the longevity, fecundity and oviposition period of different strains of *M. domestica*.

CHAPTER TWO

LITERATURE REVIEW

2.1 House fly in general

House fly, *M. domestica*, is the most common cosmopolitan and notorious pest among filth flies in both farms and homes. It is believed to have originated in the southern Palearctic region, particularly in the Middle East (Skidmore 1985), and was introduced to the New World in colonial times (Legner and McCoy 1966). It is synanthropic and is found to colonize habitats wherever mankind and livestock exist.

In nature, house fly can readily establish colonies from the progeny of a single mating pair (Krafsur et al. 2005). House fly is a highly versatile insect which has constantly adapted to new environments. It is found to flourish in the tropics as well as in temperate countries (Oldroyd 1964). It is active all year round in Malaysia especially in poultry farms as the climate in this tropical country creates ideal conditions for it to breed.

Not only is house fly a nuisance, it is an insect of medical and veterinary importance due to its unpleasant feeding habits that include feces or other protein-containing filth, in addition to foodstuffs in the kitchen (Greenberg 1971). In Malaysia, house fly is found commonly foraging on or breeding in garbage, chicken droppings and other waste materials. It mechanically transmits pathogens causing human diseases when they visit clean food (Reid 1953; Greenberg 1973).

Esten and Mason (1908) as well as Schuler et al. (2005) documented that a single feeding house fly was capable of carrying 6×10^6 bacteria on the exterior surface while Harwood and James (1979) isolated more than 100 species of pathogens from the digestive tract of a single house fly, which may cause numerous diseases namely typhoid and paratyphoid fever, bacillary dysentery, cholera, bovine mastitis, conjunctivitis and poliomyelitis (Greenberg 1970; Greenberg 1973; Gough and Jorgenson 1983). In subtropical country, the yearly infant mortality in a village notably decreased from 227 to 115 per thousand live births, when the flies were controlled by chlordane (Peffly 1953).

House fly is a major pest in animal production facilities, especially poultry houses (West 1951; Axtell 1985; Williams et al. 1985), resulting in annoyance and indirect damage in livestock production and dairy housing systems which in turn causes contamination of agricultural and farming products, and impairs meat and milk production (IRAC 2007).

House fly carries microorganisms around its bodies and transfers the bacteria to cows' teats and milk containers, resulting in an elevated microbial count in the milk. In California, Freeborn (1925) reported that milk yield during a month with severe fly infestation particularly by house fly was reduced by 3.33%. In 1992, flies were recorded to cause the greatest economic loss to poultry industry especially egg industry in New York and Pennsylvania. Eggs were contaminated with fly excreta, and therefore declined in value which may reduce farmers' profit margins. The poultry producers were estimated to spend approximately 7,000 US dollar per year on fly control (PMEP 1993).

2.2 Basic biology of *Musca domestica*

2.2.1 Identifying characters

House fly, *M. domestica* belongs to the family Muscidae characterized by 4 dark longitudinal lines on the mesonotum of its thorax. The dull grey adult fly is usually 6 to 7 mm in length. The clear and transparent wings are somewhat yellowish at the base. When at rest, house fly shows a triangular appearance when viewed from above as the wings are directed posteriorly (West 1951). The male can be distinguished from its eyes that are much closer together (holoptic) than those of the female (dichoptic) (Fig 2.1).

The eggs that are deposited in masses in crevices of the development-medium (Colyer and Hammond 1968) are pearly white in color and measure approximately 1.1 to 1.2 mm in length (Hinton 1960; Hinton 1967). Both ends are bluntly rounded, but the anterior is always more tapered (West 1951).

The creamy whitish larva, also known as maggot, is legless, cylindrical in shape, tapered anteriorly and truncates posteriorly (Reid 1953). The posterior spiracles, a pair of D shaped brown dots on the hind end, with sinuous slits are applicable in distinguishing the maggots from those of other flies. The larva undergoes 3 instars with the earlier instar larva measuring 3 to 9 mm and full-grown larva measuring 7 to 12 mm in length (Hussein and John 1998).

The 8 mm long (Hussein and John 1998) pupa is a brown barrel-shaped seed-like object formed by the contraction and hardening of the larval skin (Reid 1953). It shows a slight gradual increase in diameter, but bluntly rounded at both ends (West 1951).

2.2.2 Life cycle

House fly has a complete metamorphosis with distinct egg, larva or maggot, pupa and adult stages. The length of time from egg to adult varies very much with temperature (Reid 1953). In the tropics, house fly can produce as many as 30 filial generations a year, whereas there might be only 10 filial generations in temperate countries (Lane and Crosskey 1993).

Copulation in house flies occurs within a day or two after emerging from the pupae. The gravid females, attracted to by-products like ammonia and carbon dioxide,

Figure 2.1: Holoptic eye in male (left) and dichoptic eye in female (right) house fly.

begin to deposit their first batch of eggs on decomposed matter 4 days after copulation (Reid 1953). Each female may lay up to 900 eggs throughout her life span of about a month, in batches of about 100 to 250 eggs (Colyer and Hammond 1968).

The eggs hatch within 8 to 24 hours into larvae, which feed and develop in the substract where the eggs are laid. Optimal temperature of 35 to 38°C, with high-moisture manure favours the survival of the larvae (Hussein and John 1998). The full-grown larvae will complete their development in 4 to 6 days and migrate to drier portions of the breeding medium and pupate (Koehler and Oi 1991).

When transformation has been completed in 2 to 6 days, the fly pushes off the anterior end of the puparium by expanding and contracting the inflated sac known as ptilinum (West 1951), and emerges with folded-wings, which will be expanded after one hour (Reid 1953). An adult house fly may live an average of 30 days (Koehler and Oi 1991). However, longevity is enhanced by the availability of suitable food, especially sugar (Hussein and John 1998).

2.3 Distribution of house fly

The density of house flies fluctuates in a given locality, even if no control measures are used. This depends on the conditions for breeding, especially the temperature and sunshine, and the breeding medium (WHO 1981).

According to Madwar and Zahar (1953), the density of house fly may decline during the hottest months with hot and dry conditions in tropical and subtropical regions. It is believed that the heat during the hottest months indirectly affects the development of the immature stages of house fly due to rapid desiccation of the breeding media. Furthermore, the fact that substantial rainfall influenced the high density of house flies in the poultry farm of Kundang, Malaysia (Nazni et al. 2003a) and the dairy farm of

Florida (Romero et al. 2003), indicating that moist manure provides a suitable breeding site for house fly to breed. However, total rainfall was found to have no influence on the density of house flies in Cameron Hinglands, Malaysia (Nazni et al. 2003a). This suggested that house fly density can be different between localities and environment although climatic factors are crucial in the seasonal changes of house fly density (Mullens and Meyer 1987; Lysyk 1993; Greene and Guo 1997).

Besides climatic factors, the seasonal density of house fly can also be affected by food abundance (Wolda 1988) because the availability of food and breeding sites are related to sanitation level (Quarterman et al. 1954a; Quarterman et al. 1954b; Morris and Hansen 1966; oda 1966; Pickens et al. 1967). Thus, poultry farms and dairy farms, which contain accumulated livestock manure, possess the highest relative density of house flies (Lysyk and Axtell 1986b).

2.4 Control of house fly

It is absolutely fundamental to improve environmental sanitation in order to obtain long-term control of flies. Insecticides also constitute effective immediate solution for reducing filth fly populations and must be considered when a disease threat exists (AFPMB 2006). However, insecticide application alone is not sustainable, being limited by both time and money. Thus, integration of control methods is essential in fly suppression programmes.

Sanitation is the mainstay of house fly control in and around the home or farm. Environmental control consists of clean garbage collection areas to reduce odor and fly breeding. Also, garbage should be collected twice weekly as a minimum recommended frequency for adequate fly prevention.

In poultry farms, fresh poultry manure with 70% to 80% moisture provides ideal conditions for house fly to breed and complete its life cycle in seven days (Stafford and Bay 1987). Thus, removal of wet manure at least twice a week is necessary to break the breeding cycle (Bennett 2003). Collected manure should be stored in cone-shaped piles to reduce the surface area for breeding as the outer surfaces dry rapidly and narrow the zone in which the larvae can develop, while the heat from fermentation makes the interior unsuitable for the flies. Dry manure not only reduces the suitability of larval development, but also provides a desirable habitat for beneficial predators and parasites (Axtell 1981).

Natural biological suppression of house fly results primarily from the actions of non-pestiferous predators and parasitoids. Proper management of poultry manure encourages the presence of beneficial predators and parasitoids. These biotic agents involve in fly reduction vary geographically (Legner and Olton 1970; Legner and Olton 1971). In Puerto Rico, Legner (1965) found certain Coleopterans and Dermapterans feed voraciously on eggs, larvae and pupae of house fly. *Spalangia endius* Walker, the parasitoid species on house fly, was discovered distributed around garbage dumping sites and poultry farms in Peninsular Malaysia (Sulaiman et al. 1990). The macrochelid mite preys upon eggs and first instars of the house fly, and was found to cause substantial reductions in house fly populations (Axtell 1961; Axtell 1963; Kinn 1966). In Denmark, *Spalangia cameroni* helped suppressing the population of house fly to below nuisance level in a dairy and pig farms, by parasitizing house fly pupae (Skovgard and Nachman 2004).

Flytraps are used as a supplementary measure for fly control. The conical-designed traps have been found satisfactory for catching enormous numbers of house flies and blowflies when properly baited and placed in strategic locations outdoor. Ultraviolet light traps, the devices that are designed to electrocute the flies that enter the traps, can be used indoors in both agricultural and non-agricultural areas. They have been found especially effective against house flies.

Insecticides can play an important role in integrated fly management programmes if the fly populations are well-monitored on a regular basis, important as to decide on a particularly advantageous time to apply control. Treating building surfaces with residual sprays has been a very popular fly control strategy over the years. The flies that come into contact with the insecticide are killed when they rest on the sprayed surface. Unfortunately, the residual spray has led to exceptionally high levels of fly resistance to insecticides.

Space sprays containing synergized pyrethrins provide an excellent knockdown of adult flies. They are applied in and around livestock rearing facilities with mist, fog, or ultra low volume spraying apparatus to eliminate adult fly infestation. Both residual sprays and space sprays may be valuable measures, but should be regarded only as palliatives, not as real control measures.

Baits, the excellent selective adulticides, are usually used in conjunction with residual or space sprays to achieve an effective fly control. The baits are found to cause little or no resistance even after many years of widespread use. Fly suppression with Trichlorphon baits was first developed in Denmark in 1957. These paint-on sugar baits have been widely used on Danish farms since 1958 (Keiding and Jespersen 1986). In the United Kingdom, methomyl granular sugar bait (Golden Malrin®), containing the house fly sex attractant, Z-9-tricosene (Muscamone®), has been used extensively on farms for suppressing fly populations (Barson 1989). However, it is basically used in indoor livestock units. The use of Z-9-tricosene around the livestock farms is impractical (Hanley et al. 2004)

Larvicides are applied directly to the manure surface to kill maggots as well as predators and parasitoids associated with the manure. Thus, larvicides are not recommended for fly control program except for spot treatments. Cyromazine, the insect growth regulator, was found to control house fly effectively without killing predators and parasitoids (Hall and Foehse 1980; Miller and Corley 1980; William and Berry 1980). Neporex (a.i. 2% cyromazine) application in the dairies and pig farms caused 74-81% reduction on the house fly population (Kocisova et al. 2004). However, widespread use of cyromazine may lead to resistance in flies (Kristensen and Jespersen 2003).

2.5 Chlorinated hydrocarbons

Organochlorinated hydrocarbons, was introduced in 1940's as the first synthetic organic or 'second generation' insecticides to replace the 'first generation' insecticides (Casida 1980).

They are considered to have unfavorable persistence, environmental impact and toxic effects on higher animals including man. Therefore, organochlorinated hydrocarbons have been restricted or banned (Casida 1980; Sulaiman 1995).

DDT analogues, benzene hexachloride (BHC) isomers, and cyclodiene compounds are three major kinds of chlorinated hydrocarbon insecticides (Matsumura 1985).

2.5.1 DDT (dichlorodiphenyltrichlorethane)

DDT is a white and almost odourless crystalline solid at room temperature (Hayes and Laws 1991). It is derived from the reaction between trichloroacetaldehyde and the 2 chlorobenzene under optimal temperature and pressure conditions (Worthing and Walker 1987) (Figure 2.2).

DDT was first synthesized by Zeidler in 1874. However, its insecticidal properties were only discovered in 1939 by Paul Müller, who was awarded a Nobel Prize in 1948 in recognition of the humanitarian significance of his work on the control of vector-borne diseases (West 1951).

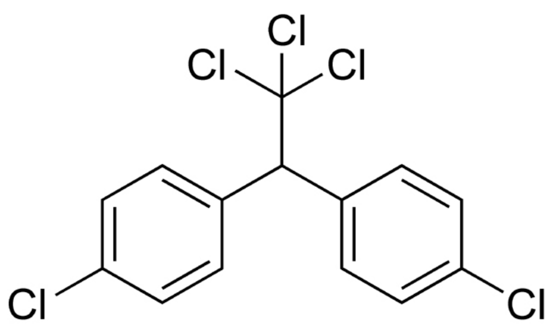

Figure 2.2: Structure of DDT.

In 1942, DDT was introduced by Geigy Chemical Company (West 1951; Davis 1971) as a broad-spectrum insecticide for the protection of the public from vector-borne diseases (Eriksson 1992). With growing desperation in seeking for a substitute for pyrethrum, DDT became a prominent military insecticide to control malaria, epidemic typhus, dysentery and typhoid fever during World War II (Davis 1971).

The mechanism of action on insects has not yet been determined. However it is believed to share the same mode of action as pyrethroids. It acts on the voltage-sensitive sodium channel of both sensory and motor nerves by altering the delicate balance of sodium and potassium ions within the axons of the neuron. Thus, prolonged depolarization occurs, leading to excitation, ataxia, convulsion, and death (Ramulu 1976).

The major drawback with the use of DDT is its resistance to breakdown under environmental conditions (persistence) and its solubility in fat (Eriksson 1992). In the 1960s, convincing evidence showed that DDT and its metabolites accumulate in food chains and are stored in living organisms (Dustman and Stickel 1969). This led to a ban on, or at least restrictions on the use of DDT in the developed countries during the 1970s and early 1980s (Coulston 1985, Eriksson 1992). With approval from World Health Organization, the use of DDT in South Africa started in 2000 when mosquitoes developed resistance to synthetic pyrethroid insecticides (Hecht 2004, Brown 2006). The World Health Organization has declared indoor use of DDT in malarious area regions where to control mosquitoes in regions where malaria is a major health problem.

2.6 Organophosphorus insecticides

Organophosphorus, an important class of insecticides, contains carbon, hydrogen, oxygen and phosphorus (Story 1990). The compounds of the insecticides were discovered during World War II in 1937 by Gerhard Schrader, a German scientist (Ramulu 1976). They are much less persistent than the organochlorine insecticides and do not accumulate in fatty tissues (Meyer 2003). In seeking substitutes for nicotine, the first organophosphorus insecticide, Bladan, was discovered and introduced in the German market in 1944 (Hayes and Laws 1991).

Organophosphorus insecticides possess the mode of action that phosphorylates the active site of the nervous system enzyme, cholinesterase (ChE), thus preventing the hydrolytic action of the enzyme upon its natural substrate, acetylcholine, which is essential in transmitting impulses between nerves. The inhibition is irreversible which results in consequent accumulation of acetylcholine in the nerve cells, giving rise to typical cholinergic symptoms associated in insects with organophosphorus poisoning, like hypersensitivity, hyperactivity, tremors, convulsions, paralysis, and death (Ramulu 1976).

At least 100 organophosphorus insecticides have been reviewed by WHO (1986a) for consideration as agents for the control of diseases. Since the removal of organochlorine insecticides from use, organophosphorus insecticides have become the most widely used insecticides for crop protection since 1970, particularly parathion and malathion (WHO 1986a).

2.6.1 Malathion

Malathion with the composition of dimethoxythiophosphorylthio (Newhart 2006) (Figure 2.3) is one of the earliest organophosphorus insecticide introduced by American Cynamid Company in 1950 (Hayes and Laws 1991). It is one of the safest organophosphorus compounds, because of its low mammalian toxicity (Hartley and West 1969; Buchel 1983; Matsumura 1985).

Malathion kills insects by contact or vapor action (Matsumura 1985). Its toxicity is increased by its break-down product, malaoxon, which is 40 times more toxic than malathion (Borwn et al. 1993).

Malathion is a nonsystemic and wide spectrum insecticide which was often used to control mosquitoes and flies (Hayes and Laws 1991). It is chemically unstable and non persistent in the environment (Lee et al. 2003). Therefore, it is used as an excellent substitute for DDT in agricultural and horticultural pests control (Rettich 1980; Matsumura 1985; Chambers 1992; Barlas 1996).

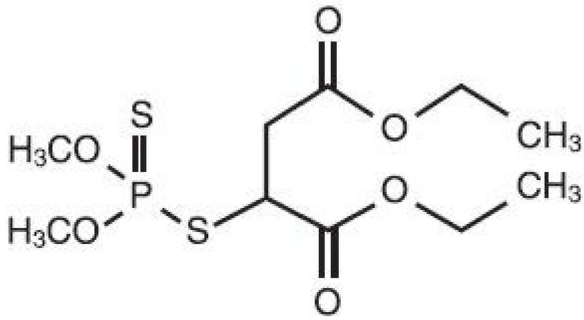

Figure 2.3: Structure of malathion.

2.7 Carbamate insecticides

Investigations of chemicals that exert an anticholinesterase action on the nervous system similar to organophosphates led in the 1950s to the development of the carbamate insecticides (Fishel 2005). Carbamates have mostly been used in toxic baits (WHO 1980), but otherwise have not played an important role in house fly control (Keiding 1977).

The first carbamate, carbaryl, was introduced by Rhone Poulenc in 1956 (Sulaiman 1995).

2.7.1 Propoxur

Propoxur, 2-(1-methylethoxy)phenol methyl carbamate (Bowman and Sans 1983a; Bowman and Sans 1983b) (Figure 2.4), is a non-systemic carbamate insecticide of moderate mammalian toxicity (WHO 1976) which was introduced in 1959 (Hayes and Laws 1991). It is developed by Bayer AG, Germany, and first registered as a pesticide in the United States (EPA 1997).

Propoxur is a broad spectrum insecticide. It is a white to cream colored crystalline compound which is highly soluble in organic solvents and slightly soluble in water (Ramulu 1976).

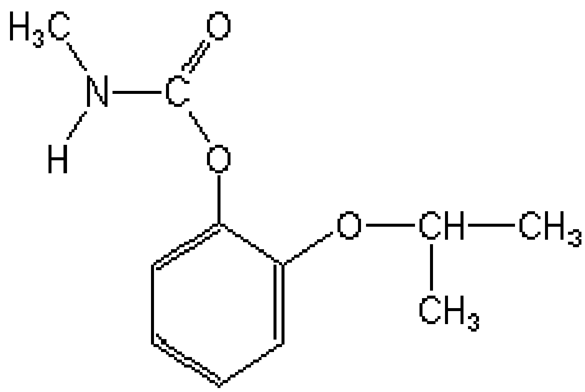

Figure 2.4: Structure of propoxur.

2.8 Synthetic pyrethroids

Pyrethroids are derivatives of natural pyrethrins isolated from *Chrysanthemum cinerariifolium*. Allethrin, the first commercial pyrethroid produced in 1949, is highly insecticidal but not persistent enough for agricultural use (Casida 1980). Photostable pyrethroids were later discovered (Elliott et al. 1973; Elliott et al. 1974) and commercialized as agriculture and home formulations since 1976 (Shafer et al. 2005).

Permethrin, decamethrin, cypermethrin and fenvalerate are the photostable pyrethroids that are used for crop protection. Due to their relatively low mammalian toxicity and outstanding potency to rapidly immobilize and kill insects (Elliott 1976; Elliott et al. 1978), these pyrethroids have replaced other insecticides and achieved worldwide use. Of the four pyrethroids, permethrin was the first pyrethroid with properties appropriate for crop protection. It was produced in 1973 (Casida 1980).

Pyrethroids are commonly classified as Type I pyrethroids and Type II pyrethroids based on their different behavioral, neuropsychological and biochemical profiles (Gammon et al. 1981; Scott and Matsumura 1983a; Ray and Fry 2006). The mode of action of Type I pyrethroid involves interference with the nerve-membrane sodium channels, leading to prolonged depolarization and induction of repetitive activity (Wouter and van den Berken 1978; Narahashi 1985; van den Berken and Vijverberg 1988). This kind of neurotoxic action and the development of neurotoxic symptoms are identical to that of DDT (van den Berken et al. 1973; Narahashi 1982; Lund and Narahashi 1983). The most prominent reason cited for this conclusion is the cross-resistance exhibited by *kdr*-type DDT-resistant insects to Type I pyrethroids (Matsumura 1985).

2.8.1 Permethrin

Permethrin is an ester of the dichloro analogue of chrysanthemic acid, 3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-carb-oxylic acid, and 3-phenoxybenzyl alcohol (IPCS 1990) (Figure 2.5). Permethrin is a non-systemic and moderately persistent insecticide that has proved to be effective against a wide range of insects (Hassall 1990). It was discovered in 1973 as the first photostable pyrethroid (Elliott et al. 1973; Casida 1980). It was marketed in 1977 and has become an integral component of crop protection management (Sibley and kaushik 1991).

Permethrin in formulations of emulsifiable concentrate, ultra-low-volume concentrate, wettable and dustable powder is nonselective in pest control programmes, resulting in widespread impact on non-target communities, particularly in aquatic environments (Mulla et al. 1978; Muurhead-Thomson 1978).

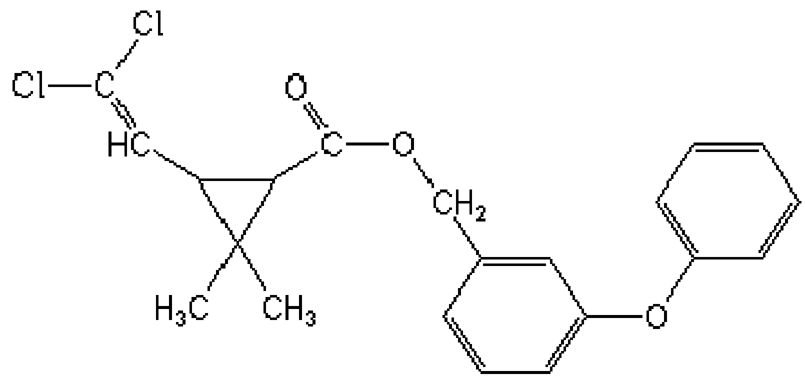

Figure 2.5: Structure of permethrin.

2.9 Resistance status of house fly

Resistance is due to inherent characteristics that allow an insect to survive an insecticide dosage which is proven to be fatal (Hemingway 1992). Pesticide resistance is found in many living organisms ranging from bacteria to mammals (Georghiou and Mellon 1983). Of the pesticide resistance, insecticide resistance is the immense problem the public concern about because of the ability of insects to develop resistance to insecticides physiologically or behaviorally (Sawicki and Denholm 1984).

Lime sulphur resistance in San Jose scale *Quandrispidiotus perniciosus* (Comstock) on apple in the state of Washington, USA was the first documented case of insecticide resistance reported by Melander (1914). Thus, insecticide resistance not only causes problems for agricultural production and control of vector-borne diseases (Akiner and Caglar 2006), but also increases the cost of control programme to total control failures by chemical tools (Moberg 1990).

Insecticides constitute the most effective immediate solution for reducing filth fly populations and must be considered when a disease threat exists. But, house flies have shown a remarkable ability to develop resistance to virtually every insecticide used against them (Brown and Pal 1971; Keiding 1977) and insecticide resistance in the house fly is a global problem (Georghiou and Mellon 1983; Scott et al. 1989). In addition, house fly has shown a remarkable ability to cross resistant to new insecticides. (MacDonald et al. 1983; Scott et al. 1989; Shen and Plapp 1990; Scott and Wen 1997).

As early as 1946, house fly was found to develop resistance to organochlorinated hydrocarbon insecticides. This was followed by the development of resistance to other groups of insecticides, namely organophosphorus, carbamates and synthetic pyrethroids (Chapman and Morgan 1992).

DDT resistance seems to occur everywhere, in spite of the fact that DDT has not been used for fly control for many years in most places. The widespread use of DDT in the 1940's has led to appearance in many countries of resistant strains of house flies (Harrison 1952). The first report that house flies did not respond as expected to DDT was discovered in 1946 in northern Sweden (Brown 1971; Wiesmann 1947). Since then, several hundred similar instances have been noted involving many of the newer insecticides, starting with resistance to BHC and dieldrin which was first reported in 1948 (Busvine 1963). In Florida, King (1950) found that DDT-resistant house flies can be effectively controlled by using lindane. However in New York, house flies were found to develop resistance to lindane and other insecticides used as residual sprays in dairy barns 2 years after exposure (Goodwin and Schwardt 1953).

Organophosphorus compounds have received major attention to overcome the resistance to organochlorinated hydrocarbon compounds. However, as had happened with DDT, organophosphorus-resistance in house fly has been shown by a number of surveys and found to be increasing (Scott et al. 2000; Kristensen et al. 2001). Resistance to the newly introduced organophosphorus insecticides was first noticed in Denmark and

USA in 1955, and later in many other countries (Brown and Pal 1971; Pal and Wharton 1974). In California, Georghiou and Bowen (1966) reported that house flies were resistant to various organophosphorus and carbamate compounds. In Denmark, organophosphorus-multiresistant strains of house flies showed high resistance to carbamates (Keiding 1977).

Pyrethroid insecticides have achieved great commercial success in control of many insect pests. However, potential resistance problems exist, particularly where insects were previously selected by DDT and are cross-resistant to pyrethroids (Busvine 1953; Whitehead 1959; Plapp et al. 1968; Scott and Matsumura 1981). House fly resistance to pyrethroids has been documented in several countries (Keiding 1976; Kunast 1979; Sawicki et al. 1981; MacDonald et al. 1983). In 1957, house fly resistance to pyrethroid insecticides was first reported in Sweden (Davies et al. 1958). Since then, a few other cases of resistance have been reported. The first permethrin-resistance in house fly was reported in 1979 in Canada by Surgeoner (Solomon et al. 1990).

House fly is the major pest of public health importance in Malaysia and its control depends mainly on the use of insecticides (Sulaiman and Omar 1992; Nazni et al. 1997). In Malaysia, Wharton et al. (1962) revealed that house flies from Cameron Highlands and lowlands around Kuala Lumpur were resistance to DDT and BHC (benzene hexa-chloride). Nevertheless, Cheong et al. (1970) reported that lowland flies possessed higher degree of resistance to hydrocarbon compounds and organophosphates compared to highland flies. A few years later, house fly was found to possess a considerable degree of resistance to malathion, toxaphene, aldrin, dieldrin and pyreiithrin (Singh 1971; Singh 1973). In the 1990's, flies from Cameron Highlands have shown to develop resistance towards malathion and permethrin (Nazni et al. 1997).

CHAPTER THREE

MONTHLY INDEX OF *Musca domestica*, THE HOUSE FLY, IN POULTRY FARMS IN PENANG, MALAYSIA

3.1 INTRODUCTION

The number of flies varies in a given locality, depending on climatic factors (WHO 1981) and duration of daytime (WHO 1986b; WHO 1986c), both influence the rate of mating, the preoviposition period, oviposition and feeding of adults.

In temperate regions, fly density starts increasing in spring and reaches its peak during the summer season and then decreases in autumn. Freezing conditions during winter may preclude all possibility of species survival (Oldroyd 1964). According to Hewitt (1915) and Somme (1961), adult flies are the important overwintering stage. They overwinter in part by continuously breeding in suitable places, or as hibernating females. This contributes to the high density of flies in the next spring and summer. However, Skinner (1913) and Kobayashi (1921) observed that pupae appear to tolerate lower temperatures.

In tropical and subtropical areas, fly density may decline during the hottest months (Madwar and Zahar 1953; WHO 1986b; WHO 1986c). However, flies are active all year round in tropical rainforest countries. Breeding of flies in the tropics rarely terminates completely as a result of seasonal change. In unfavorable conditions, a mere reduction in the intensity of reproductive activity results in a reduction of the population. With the return of favorable conditions, flies increase in numbers, and may reach a seasonal peak (Oldroyd 1964).

Apart from that, fly abundance is also found to be associated with poor sanitation where manure is abundant (Hughes and Walker 1969). Livestock manure that is constantly being changed by numerous abiotic and biotic factors, affects the population of house flies (Stafford and Bay 1987). The foul-smelling manure gives off by-products like ammonia and carbon dioxide which attracts a large number of flies to breed in the moist manure. Concentrated organic manures, used in the vegetable gardens are also principal breeding places for house flies (Pagden and Reid 1952).

Insecticide applications to reduce dense populations to lower levels have been attributed to house fly resistance towards insecticides. Hence, the development of new technologies for control has raised the interests of researchers to investigate fly density over an extended period of time as the information might suggest a particularly advantageous period to apply control.

In Malaysia, seasonal abundance of flies has been reported by Singh (1971) and Nazni et al. (2003a) in the central region of western Peninsular Malaysia. However, limited information is available on the temporal index of flies in northwestern Peninsular Malaysia.

3.2 MATERIALS AND METHODS

3.2.1 Site selection

The 14-month study was conducted from March 2007 to April 2008 in two poultry farms that are 21.68km apart, one in Balik Pulau and the other in Juru.

Balik Pulau, a suburban area on the island of Penang, is famous for its fruit orchards and spice gardens. It is located approximately 8km (altitude 2.8m) from the capital, Georgetown. The egg-production poultry farm consists of one narrow-style (4.32m by 23.87m) poultry shed (Figure 3.1), with two rows of three-tiered cages (91 cages per tier with 2-3 birds per cage) suspended 0.80m above the ground (Figure 3.2).

Figure 3.1: Balik Pulau poultry farm.

Figure 3.2: The poultry shed in the Balik Pulau poultry farm.