# STUDY ON FLIES DIVERSITY AND EFFECT OF BIOTIC AND ABIOTIC FACTORS ON HOUSEFLIES AT DIFFERENT FARMING SYSTEMS

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by

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# LIST OF SYMBOLS

| | |
|---|---|
| °C | Celsius |
| % | Percentage |
| < | Less than |
| > | More than |
| ± | Plus minus |
| p | Probability of null hypothesis |
| t | The calculated difference represented in units of standard error |
| *r* | The correlation coefficient |
| n/N | The total number of individuals or observations in the sample |
| phi | phi correlation coefficient |

## LIST OF ABBREVIATIONS

| | |
|---|---|
| AMR | Anti-microbial resistance |
| DNA | Deoxyribonucleic acid |
| DVS | Department of Veterinary Services, veterinary authority |
| g | gram |
| $HS^-$ | Hydrosulfide |
| $H_2S$ | Hydrogen sulfide |
| IPM | Integrated pest management |
| KAP | Knowledge, attitude, and practices |
| Kg | Kilogram |
| KM | Kilometer |
| $mg/m^3$ | milligrams per cubic meter |
| $m^2$ | Square meter |
| mm | Milimeter |
| m/s | Meter/ second |
| $NH_3$ | Ammonia |
| ppb | Parts per billion |
| ppm | Parts per million |
| RH/ rH | Relative humidity |
| $S^{2-}$ | Sulfide |
| SD | Standard deviation |
| USM | Universiti Sains Malaysia |

## LIST OF APPENDICES

**KAJIAN KE ATAS KEPELBAGAIAN LALAT DAN KESAN FAKTOR BIOTIK SERTA ABIOTIK TERHADAP LALAT RUMAH PADA SISTEM PENTERNAKAN YANG BERBEZA**

## ABSTRAK

Kajian mengenai kesan sistem penternakan dan lantai bawah reban terhadap kelimpahan *Musca domestica* (lalat) dijalankan di daerah Kinta, Perak, Malaysia. Kajian ini dijalankan dengan bermatlamatkan untuk menentukan kepelbagaian spesis Diptera diukur di 14 buah ladang unggas komersial dengan sistem *"close house"* dan *"open house"*; kesan faktor biotik dan abiotik, kadar kelajuan angin terhadap kelimpahan lalat di ladang dengan jenis lantai tanah dan konkrit; perbezaan dalam indeks lalat berkaitan dengan kelembapan relatif, ketebalan najis yang termendak di lantai bawah reban, kandungan kelembapan semulajadi tinja dan tinja basah dikaji di enam ladang ayam pedaging. Sementara itu, tinjauan soal selidik *knowledge, attitude, and practices* (KAP) telah dibuat bagi menilai tahap pemahaman skop pencegahan dan kawalan lalat di ladang. Sebanyak 7,993 ekor Diptera yang ditangkap telah dibahagikan kepada sembilan famili iaitu Muscidae, Calliphoridae, Sacrophagidae, Ulidiidae, Phoridae, Stratiomyidae, Sciaridae, Fannidae dan Syrphidae. *Musca domestica* adalah spesies lalat yang dominan yang ditangkap ($p= 0.047$) dan ia mewakili 74.01% *(open house)* dan 68.26% *(close house)* sementara *Megaselia scalaris* adalah spesies kedua yang paling banyak (21.66% di *"open house"* dan 30.58% di *"close house"* daripada kesemua Diptera yang ditangkap. Penemuan pada sistem lantai tanah menunjukan bacaan indeks lalat sebanyak $7.67 \pm 0.23$ ekor/30 saat, $p= 0.000$; umur ayam ($12.29 \pm 0.48$ hari, $p= 0.057$); suhu ($29.14 \pm 0.07$ °C, $p= 0.000$); amonia ($0.85 \pm 0.04$ ppm, $p= 0.028$); hidrogen sulfida ($0.04 \pm 0.00$ ppm, $p=$

0.004); kelajuan angin (0.98 ± 0.03 m/s, p= 0.457); kelembapan relatif (56.79 ± 0.27%, p= 0.264); kelembapan semulajadi tinja (45.53 ± 1.09%, p= 0.107); kelembapan tinja basah (70.46 ± 1.62%, p= 0.707) dan ketebalan tinja termendak (26.13 ± 0.94 mm, p= 0.000). Manakala, penemuan pada lantai konkrit pula menunjukkan bacaan indeks lalat sebanyak 6.56 ± 0.22 ekor/30 saat, p= 0.000; umur ayam (11.04 ± 0.45 hari, p= 0.057); suhu (29.59 ± 0.07 ℃, p= 0.000); gas amonia (0.98 ± 0.04 ppm, p= 0.028); gas hidrogen sulfida (0.05 ± 0.00 ppm, p= 0.004); kelajuan angin (0.95 ± 0.03 m/s, p= 0.457); kelembapan relatif (55.83 ± 0.81%, p= 0.264); kelembapan semulajadi tinja (41.92 ± 2.14%, p= 0.107); kelembapan tinja basah (71.42 ± 2.01%, p= 0.707) and ketebalan tinja termendak (12.08 ± 0.50 mm, p= 0.000). Bagi kajian KAP, semua kumpulan pekerjaan menunjukkan tahap pengetahuan yang sederhana bagi biologi, tingkah laku dan reproduksi lalat, kesan infestasi lalat, dan tahap pemahaman dan kesedaran tentang betapa teruknya masalah lalat. Pengetahuan berkenaan pengurusan dan amalan kawalan lalat di kalangan semua responden juga didapati sangat lemah berdasarkan kelayakan akademik (p= 0.000) and kawasan perumahan (p= 0.000). Sebanyak 96.6% (85/88) daripada aduan gangguan lalat bulanan yang diterima adalah berkaitan dengan ladang ayam. Sehubungan dengan itu, kira-kira 55.2% responden telah mengesyorkan pematuhan pada Amalan Penternakan Baik (GAHP) untuk membentuk industri unggas yang mampan dengan kadar infestatsi lalat yang minimum dalam masa terdekat. Kesimpulannya, kelembapan tinja am dan tinja basah yang homogenus juga memberikan peluang yang sama bagi pembiakan lalat berlaku. Namun demikian, kelimpahan lalat sangat berkait rapat dengan pengurusan tinja dimana pematuhan GAHP dan kaedah kawalan kultur boleh dijadikan sebagai strategi kawalan asas bagi pengurusan lalat di ladang ayam pedaging di Malaysia.

# STUDY ON FLIES DIVERSITY AND EFFECT OF BIOTIC AND ABIOTIC FACTORS ON HOUSEFLIES AT DIFFERENT FARMING SYSTEMS

## ABSTRACT

A study on the effect of the rearing system and the manure settling floor on the abundance of *Musca domestica* (fly) was conducted in Kinta district, Perak, Malaysia. The objectives of this study were to determine the diversity of Diptera species in 14 commercial poultry farms with close house and open house systems; the effect of biotic and abiotic factors and wind speed on the abundance of flies in fields with soil and concrete floor types; differences in the housefly index related to relative humidity, the thickness of manure deposited on the floor of the house, the natural manure moisture content, and wet manure were studied on six broiler farms. Knowledge, attitude, and practices (KAP) survey was also carried out to assess the current housefly management and prevention measures in practice. A total of 7,993 flies captured belonged to nine families namely, Muscidae, Calliphoridae, Sacrophagidae, Ulidiidae, Phoridae, Stratiomyidae, Sciaridae, Fannidae and Syrphidae. The *Musca domestica* was the predominant fly species ($p= 0.047$) and it represented 74.01% (open house) and 68.26% (closed house) while *Megaselia scalaris* was the second most abundant species constitutes (21.66% in open house and 30.58% in closed house farming system). In earthen manure settling floor, mean housefly index was $7.67 \pm 0.23$ heads/30 seconds, $p= 0.000$ ; bird's age ($12.29 \pm 0.48$ days, $p= 0.057$); temperature ($29.14 \pm 0.07$ °C, $p= 0.000$); ammonia ($0.85 \pm 0.04$ ppm, $p= 0.028$); hydrogen sulfide ($0.04 \pm 0.00$ ppm, $p= 0.004$); wind speed ($0.98 \pm 0.03$ m/s, $p= 0.457$); relative humidity ($56.79 \pm 0.27$%, $p= 0.264$); general manure moisture (45.53 ± 1.09%, p= 0.107); wet spot moisture (70.46 ± 1.62%, p= 0.707) and thickness of manure settled (26.13 ± 0.94 mm, p= 0.000). While in concreted manure settling floor, the mean housefly index was 6.56 ± 0.22 heads/30 seconds (p= 0.000); bird's age (11.04 ± 0.45 days, p= 0.057); temperature (29.59 ± 0.07 °C, p= 0.000); ammonia (0.98 ± 0.04 ppm, p= 0.028); hydrogen sulfide (0.05 ± 0.00 ppm, p= 0.004); wind speed (0.95 ± 0.03 m/s, p= 0.457); relative humidity (55.83 ± 0.81%, p= 0.264); general manure moisture (41.92 ± 2.14%, p= 0.107); wet spot moisture (71.42 ± 2.01%, p= 0.707) and thickness of manure settled (12.08 ± 0.50 mm, p= 0.000). As for the KAP study, all occupational groups exhibited a moderate level of knowledge of houseflies biology, behaviour, and reproduction, the impact of houseflies infestations, and depth of understanding and awareness on the severity of housefly problems. Knowledge about management and fly control practices among all respondents was also very weak based on academic qualification (p= 0.000) and residential area (p= 0.000). A total of 96.6% (85/88) of the monthly fly nuisance complaints received were related to poultry farms. Subsequently, about 55.2% of respondents have proposed compliance with Good Animal Husbandry Practices (GAHP) to form a sustainable poultry industry with minimal housefly infestation in the near future. In conclusion, homogenous general and wet spot moisture content suggests an equal chance of housefly breeding. However, housefly abundance was highly related to the manure management system, where adherence to GAHP and cultural control methods can be considered the most basic fly management strategies on broiler chicken farms in Malaysia.

# CHAPTER 1

# INTRODUCTION

## 1.1 Poultry industry and farming system

Poultry refers to domesticated birds that are raised for the purpose of producing table eggs or feathers (Olsen et al., 2012). While the world boasts a record of around 10,000 bird species, only a limited selection of approximately 12 to 13 species fall under the classification of poultry. These include familiar birds like chickens, ducks, geese, turkeys, and capons, among others (Richa et al., 2020). Commercialization of poultry production is rapidly expanding worldwide for the past two decades. Poultry production can be categorized into seven groups, namely great grandparent stocks, grandparent stocks, broiler breeders, layer breeders, commercial broilers, commercial layers and village/native chicken farming. Through the advancement of industrialized production system, production cycle time for poultry became relatively short and profitable (Oduwaiye et al., 2017). The poultry industry in Malaysia, specifically broiler meat has achieved self-sufficiency level since 1984 (Bahri et.al., 2019). In Malaysia, the broiler production system is segmented into three distinct methods: open house, converted closed house, and fully enclosed closed house systems. Technology usage and practices for open and complete close house systems are quite distinctive. While, converted close house system is an upgraded version of open house farming with all elements of close house system except manure management. The emergence of this unique converted close house system is often a sequel of persistent urge, and enforcement activities of local veterinary authorities.

## 1.2 Flies and It's Impact

Ongoing residential development in previously agricultural or open areas near poultry facilities has led to an escalating frequency of conflicts over land use between farms, and their relatively new suburban neighbors. In general, the substantial presence of flies within poultry establishments becomes a source of irritation not just for the workers but also for the nearby inhabitants (Miller et al., 1993; Winpisinger et al., 2005; David et al., 2013), often resulting in violations of Poultry Enactments (Mullens et al., 2001). In the context of Malaysia, significant public health concerns stem from common synanthropic species within the Calliphoridae, Muscidae, and Sarchophagidae families (Nurita et al., 2008). Meanwhile muscoid flies, particularly those belonging to the Muscidae, Fanniidae, and Anthomyiidae families, are prominent pests on poultry farms (Zchori et al., 1992). These Dipterans are commonly associated with both human and animal production environments (Rezende et al., 2017). Other species coexisting with *Musca domestica* include *Stomoxys calcitrans* (Diptera: Muscidae), *Chrysomya sp.* (Diptera: Calliphoridae), and *Fannia sp.* (Diptera: Fanniidae) as reported by Axtell & Arends (1990), Lomnaco & Prado (1994), Axtell (1999), and Lopes et al. (2007).

In a study conducted on layer poultry farms in Brazil, Lomnaco & Prado (1994) determined that *M. domestica* holds the highest population among dipterous species in this particular type of operation. They also confirmed the presence of *Fannia pusio* and *Fannia trimaculata* within the visited aviaries. Similarly, Bruno et al. (1993) identified *F. canicularis*, *F. trimaculata*, and *F. pusio* in poultry farms located in the Brazilian state of São Paulo. Further research by Lopes et al. (2007) in a layer poultry farm within the state of São Paulo in 2007 confirmed the existence of additional species alongside *Fannia* sp., including *M. domestica*, *Chrysomya megacephala*, *Hermetia illucens* (Diptera: Stratiomyidae), and Dipterans from the Sepsidae and Syrphidae families. Contrastingly, Avancini & Silveira (2000) investigated poultry farms in southeastern Brazil and observed a substantial presence of *Muscina stabulans* (Diptera: Muscidae), *M. domestica*, *Chrysomya putoria*, *C. megacephala*, and *S. calcitrans*. In another study, Monteiro & Prado (2000) recorded the prevalence of *C. putoria*, *M. stabulans*, *M. domestica*, and *F. pusio*. Borges (2006) found *Drosophila repleta* (Diptera: Drosophilidae), *M. domestica*, and *C. putoria* to be the dominant species within a poultry farm in Minas Gerais, Brazil.

On the other hand, in the United States, *M. domestica, M. stabulans,* and *S. calcitrans* are the most frequently reported adult species in poultry facilities (Coupland & Barker, 2004). Lopes et al. (2008) exclusively identified adult *D. repleta* stages on livestock farms, with no larval stages detected in manure. In another context, Fernandes et al. (1995) documented Dipterans from the Drosophilidae family in a poultry farm situated in Uberlândia, Minas Gerais, Brazil (Rezende et al., 2017).

The common housefly, *Musca domestica*, stands out as the predominant and troublesome fly species within poultry facilities, as evidenced by various studies (Rezende et al., 2017). In tropical conditions houseflies are active around 9.00 to 11.00 am (Sulaiman et al., 2000). Nevertheless, Elaiyabarathi & David (2009) recorded active housefly activity from 6.00 to 10.00 am and 4.00 to 6-00 pm (Bell et al., 2019). At night and any time when they are not eating or breeding, adults will be resting or roosting. They are often seen roosting on any stable surface adjacent to the breeding or feeding sites, such as floors, walls, ceilings, furniture, plants, fences, garbages, and etc. Although most adult houseflies stay close to their breeding sites, a proportion will disperse away and may cause problems to the public. Houseflies are capable of dispersing over distances of several kilometers, although problems seldom occur at distances greater than 2-3 km from the source. Significant fly attacks likely to occur within 500 m radius from the problematic farms (Wallace and Boase, 2018).

Where there are breeding sites nearby, residents or employees may experience hundreds of flies in their homes or workplace.

Adult houseflies possess the ability to solely consume liquids, necessitating the regurgitation of stomach fluids onto solid food to transform it into a consumable liquid state. This process, known as fly pecking, is marked by straw-colored regurgitation spots and dark fecal spots. Regrettably, this behavior accelerates the corrosion of metal equipment, deterioration of paint, compromises the quality of eggs, and leads to diminished illumination from poultry house lighting fixtures (David et al., 2013). Furthermore, houseflies are recognized carriers of over 65 intestinal diseases affecting humans and animals. They contribute to the spread of infections caused by protozoans (such as amoebic dysentery), bacteria (including shigellosis, salmonellosis, and cholera), helminths (comprising roundworms, hookworms, pinworms, and tapeworms), as well as viral and rickettsial agents (Malik, 2007). These flies exhibit a strong attraction to the odor of manure, often selecting it as a suitable site for laying their eggs (Axtell, 1999). Ammonia, a significant olfactory element in poultry farms, arises from the breakdown of nitrogen-containing compounds within manure and poultry litter (Malik et al., 2007). Coupled with instances of feed and water spillage, the accumulation of poultry manure fosters ideal conditions for housefly breeding. Extended grow-out periods or production cycles facilitate multiple fly breeding cycles. Stafford (2008) observed that housefly populations peaked when the moisture content of poultry manure ranged from 70% to 79%, with a notable reduction occurring when moisture levels fell below 60% (Mullens et al., 1996). Instances of leaky water nipples, inadequate ventilation, and external seepage contribute to the creation of damp and humid regions within barns, thereby promoting conditions conducive to housefly reproduction (Axtell, 1999).

The flies infestation issue has grown more pronounced within contemporary, high-density poultry facilities, particularly due to the tendency to amass manure beneath the birds. In these advanced setups, the accumulation of manure within the poultry house persists throughout the grow-out phase, spanning approximately 4 to 5 weeks for broiler chickens and up to 12 weeks for broiler ducks before they are sent to the market. This extended duration results in a larger volume of manure and spent litter being generated. Consequently, there is an augmented quantity of material suitable for fly breeding, leading to the expansion of fly populations (Axtell, 1999). Adding to that, the volume of manure production is directly proportional to fly harbourage in poultry facilities (Achiano & Giliomee, 2005).

Since major fly breeding occurs in poultry farms, fly control is the responsibility of the farmers. Therefore, understanding the extent of fly mobility within and around poultry farms holds significance in formulating effective strategies for fly control initiatives (Avicola et al., 2022). Fundamentally, farmers, veterinary authorities, and the policy makers must accept the fact that flies cannot be eliminated. However, houseflies population can be kept at acceptable low levels by proper management practices (Geden et al., 2021). Basically, Integrated fly control procedures have been established in earlier days, most anti-fly measures are pesticide-centered, resulting in insecticide resistance in flies and environmental pollution (Keiding, 1999; Shono et al., 2004; David et.al., 2013). Apart from that, insecticide based fly control strategies incurs high cost. Likewise, poultry farmers were estimated to allocate around RM 32,815.99 annually for fly management (PMEP, 1993). Primarily, cultural control techniques represent the most viable and achievable approach, achieved through the alteration of the conducive breeding environment by manipulating abiotic elements like temperature, breeding habitat moisture, and relative humidity. This serves to diminish the prevalence of houseflies within poultry establishments (Walker & Stachecki, 1996; WHO, 1991).

## 1.3 Objectives

The present study was concentrated on broiler chicken farms with open house systems, as there are a huge number of these farms to assure the continuous supply of chicken meat at local markets. So far, not many studies has been carried out to evaluate the efficiency of these two types of poultry housing systems in controlling housefly abundance. Thus, findings on the effect of manure floor type on manure moisture content, breeding site humidity, and air speed can contribute to the optimization of a management plan in order to reduce flies population and improve the quality of life of the communities neighbouring poultry farms. Hence, the objectives of this study were:

1. To determine fly species diversity between closed and open house commercial poultry farms.
2. To investigate the effect of biotic and abiotic factors (temperature, wind speed, concentration of hydrogen sulfide and ammonia gasses) on the housefly abundance in earthen and concreted manure settling floors in open house broiler chicken farm.
3. To investigate the effect of manure settling floor type on housefly abundance based on humidity, general manure moisture content, wet spot manure moisture content, and thickness of manure settled in open house system of broiler chicken farms.

4. To investigate the Knowledge, Attitude and Practices (KAP), and awareness of the poultry farmers, veterinary authorities and public towards housefly control management in poultry farms in Malaysia.

# CHAPTER 2

# LITERATURE REVIEW

## 2.1 Introduction

### 2.1.1 Poultry Industry

The term "commercial poultry raising" pertains to any location where domesticated birds like chickens, ducks, geese, or turkeys are nurtured or bred with the intent of selling them or their eggs. It also encompasses any site where more than twenty such birds, aged three months or older, are housed for any designated objective (FAO, 2022). Commercialization of poultry production is rapidly expanding worldwide for the past two decades. In Malaysia, the poultry sector plays a leading role in addressing the needs of the nation's populace. Chicken meat and eggs, in particular, are widely favored due to their affordability and widespread appeal across diverse ethnic groups, catering to preferences without encountering religious limitations (Faiza et al., 2013). The commercial broiler chicken farming is the most common poultry industry in Malaysia.

### 2.1.2 Commercial Broiler

Broilers, which are rapidly growing specialized chickens raised for their meat, attain weights ranging from 1.4 to 1.8 kg (Asaniyan et al., 2007). Typically, both male and female broilers are raised together and are processed before reaching sexual maturity. In Malaysia, the broiler industry has experienced significant advantages through the implementation of major structural changes, transitioning to large-scale, vertically integrated operations that subcontract the growing phase to smaller farmers. Close housing systems and modern technologies are embraced by around 60% of broiler operators in Malaysia. The closed house has two floors with a low ceiling, utilizes mechanical tunnel ventilation and automated systems for feeding, watering, ventilation, heating, feed storage and feed distribution, lights and control system. Additionally, it employs semi-closed side walls with plastic curtains and sealed floors with litter, as described by Carmelo (2020). Temperature, moisture, and odors, particularly ammonia, are regulated through a combination of exhaust fans (in the closed house system) and natural airflow (in the open house system).

Besides that, factors like animal health management, feed management, and biosecurity measures are central concerns for farm operators due to their direct impact on profitability. Among the technological practices adopted by broiler operators are feeding systems, disinfection methods, vaccination protocols, as well as strategies to control fly and odor pollution, manage farm waste, and handle the disposal of broiler carcasses. The average production cycle for broiler chickens in Malaysia is approximately 5.33 cycles per year, with only a few operators, primarily multinational companies, achieving six cycles annually. Harvesting takes place when the birds attain a minimum weight of 2.2 kg. Generally, broiler chickens are ready for sale around 30 to 33 days after being raised (Kaur & Arshad, 2008).

### 2.1.3 Broiler industry in Malaysia

The poultry industry in Malaysia has achieved self-sufficiency. By 2019, the ex-farm value of livestock products within Malaysia had surpassed RM 24 billion. Poultry meat contributed approximately RM 12.4 billion, while eggs held an ex-farm value of RM 5.8 billion (Carmelo, 2020). These accomplishments have been realized through a series of structural transformations in the sector. This includes the rise and expansion of "land-independent" (industrial) farming establishments, along with the intensification and concentration of poultry operations. These changes have unfolded over the past years, driven by the persistent drive to reduce production costs and elevate supply. The response has been the adoption of more efficient methods, facilitated by transitioning to larger, specialized, and integrated facilities. This is supported by advancements in animal genetics, optimized nutrition, and innovative production technologies (Gerber et al., 2007; Carmelo, 2020). A typical broiler farm encompasses multiple broiler houses, each designed to house 20,000 to 25,000 birds. In essence, a broiler shed employing a closed-house system incorporates automated equipment for essential functions including feeding lines, drinking lines, ventilation, heating, feed storage and distribution, lighting, and a control system (Carmelo, 2020).

## 2.2 Close house and open house poultry farming system

The selection of housing style and management approaches is contingent upon factors such as the specific poultry variety being raised, economic considerations, as well as regional and climatic preferences. Regardless of the specifics, confined poultry housing entails a higher degree of environmental regulation compared to outdoor settings (Carmelo, 2020). Presently, the broiler production system in Malaysia is categorized into three distinct systems: complete closed house, converted closed house, and open house systems. Each of these systems employs differing technological applications and practices. Notably, the complete closed house system exhibits superior efficiency and effectiveness in comparison to the open house system (Smith et al., 2023). While, the converted closed house system is an upgraded version of open house farming with all elements of the closed house system; except manure management. The emergence of this unique converted closed house system is often a sequela of persistent urge and enforcement activities of local veterinary authorities. However, the closed house system is more efficient and effective than the converted closed house and open house systems. Technically, both complete closed house and converted closed house farming are referred to closed house system. Closed house systems are employed by nearly 60% of the entire broiler farms in Malaysia, as reported by DVS in 2011.

In a closed house system, the indoor microclimate can be tailored as required, whereas in an open house, the microenvironment within a cage is contingent upon the prevailing natural conditions in the surroundings of the cage setting (Hameed et al., 2012). Elements of microclimates within poultry housing encompass factors like humidity, temperature, air velocity and movement, air composition, and lighting (Kalio & Okafor, 2012). Broilers possess specific microclimate requisites aligned with their physiological growth and production stages (Bonnet et al., 1997; Muharlien et al., 2020). Conversely, the open poultry housing system is recognized for its straightforwardness, cost-effectiveness, and the ease with which heat regulation can be managed through natural ventilation within the structure (Muharlien et al., 2020). Nevertheless, this approach is susceptible to infiltration by insects, rodents, birds, and other minor predators that can disrupt the well-being, productivity, and performance of broilers.

Apart from that, the manure management in a complete closed house system is either by manure belt extraction or litter-based excreta maintenance. Basically, manure belts are cleared every one to three days once while litter materials are only removed at the end of the production cycle. In a converted closed house system, manure will be accumulated on a curtain-covered, exhaust fan-ventilated manure settling floor. Meanwhile, manure will be accumulated on the open aired manure settling floor in the open house farming system. Manure removal in both converted closed house and open house systems are usually performed once a cycle. Essentially, as the poultry industry undergoes rapid expansion, the confinement of birds in high-density housing facilities results in the generation of greater volumes of manure and used litter. This situation provides optimal circumstances for the proliferation of fly populations (Axtell, 1999).

### 2.3 Biology of flies

Flies are pervasive insects that inhabit a wide range of ecological niches, adapting to environments with extreme temperatures, whether cold or hot. They thrive in various places, from decaying matter and manure to fungi, plants, water, and even as parasites of mammals and other creatures. Some fly species are attracted to sweet substances (Syrphidae), while others are drawn to decomposing materials (Phoridae, Muscidae, Calliphoridae), and certain flies serve as predators of other insects (Asilidae, Tabanidae). However, the flies with the most significant global impact are those that feed on blood, like mosquitoes (Culicidae), blackflies (Simuliidae), and sandflies, which can transmit diseases such as malaria, dengue fever, and leishmaniasis (Psychodidae). The diseases borne by these flies afflict millions of people worldwide, resulting in thousands of deaths annually (Hedges & Moreland, 2020).

A shared characteristic among many of these pest species is their substantial need for moisture-rich breeding sites. Conversely, specific species like mosquitos (Culicidae), crane flies (Tipulidae), midges (Chironomidae, Ceratopogonidae), and horse/deer flies (Tabanidae) have evolved specialized adaptations for breeding within aquatic environments. The cluster fly, *Pollenia sp.* is an earthworm parasite and a nuisance pest. Fungus gnats (Sciaridae, Mycetophilidae) lay their eggs in moulds and fungi. The larval developmental behaviors of hover flies and flower flies (Syrphidae) diverge due to their varied dietary habits; certain species feed on aphids, while others thrive in decaying organic matter or stagnant aquatic settings. All flies follow a holometabolous life cycle, undergoing complete metamorphosis with distinct life stages including egg, larva, pupa, and adult. A significant deviation occurs with numerous flesh flies (Sarcophagidae), as they completely skip the exposed egg stage. Instead, they directly deposit their internally hatched larvae from their abdomen onto the breeding substrate, often animal carcasses (Hedges & Moreland, 2020).

### 2.3.1 Major species of flies related to the poultry industry

Axtell (1990) reported a relative abundance of *Musca domestica, Fannia ltucoslicla, Sepsis lateralis,* species of Cecidomyiidae, Desmometopa sp., Sphaeralcea sp., *Chrysomya putoria, Stomoxys caldtrans, Muscina stabulans, Rhinia apicalis, Fannia albitanus,* and *Ophyra capensis* in manure accumulations below caged hens in poultry farms. Additionally, Machtinger et al. (2021) reviewed those filth fly pests of livestock animal industry, including the house fly (*Musca domestica*), horn fly (*Haematobia irritans*), stable fly (*Stomoxys calcitrans*), little or lesser house fly (*Fannia canicularis),* and face fly (*Musca autumnalis*). Similarly, Rachita et al. (2018) encountered the highest abundance of *Musca* sp. followed by *Aedes* sp*., Culex* sp, *Anopheles* sp., and *Stomoxys* sp. in a study conducted in poultry houses in Bhubaneswar, Odisha, India.

Another study carried out by Zuha & Disney (2020) recorded the presence of *Megaselia scalaris* (Diptera: Phoridae) and *Megaselia spiracularis* on marketed table chicken eggs in their study. On the other hand, Baharudin et al.(2003) discovered that families Calliphoridae, Muscidae, and Sarchophagidae are the common synanthropic species of significant public health importance in Malaysia. Synanthropic flies commonly inhabit urban and agricultural areas characterized by inadequate sanitation. Their presence tends to diminish significantly when stringent sanitary measures are enforced (Olsen, 1998). A study conducted by Nurita et al. (2008) revealed that the prevailing species in their surveys, carried out in Kedah and Penang, were predominantly *Musca domestica*. The occurrences of *Musca sorbens, Chrysoma megacephala,* and *Lucillia cuprina* were comparatively lower than those of *M. domestica.* Conversely, *Sarcophaga* sp.*, Megaselia* sp.*, Psycoda* sp.*, Piophila* sp.*,* and *Fannia* sp. were scarce in their captures. Ultimately, the housefly, *Musca domestica* (Diptera: Muscidae), has been identified as the most prevalent fly species within poultry establishments, posing both nuisance and health concerns for the poultry, barn workers, and nearby residents (Axtell, 1990).

### 2.3.2 *Musca domestica*

*Musca domestica* belongs to the Insecta Class and falls under the Order Diptera, which encompasses the majority of two-winged insects. Within the suborder Cyclorrhapha, they stand as members. The *Musca* genus, part of the Muscidae family, houses various other synanthropic flies as well (Alikhan et al., 2018). This genus, Musca, comprises approximately 26 species, most of which are considered "wild" and of negligible public health significance. The *Musca* sp. typically exhibit a medium-sized stature, featuring non-metallic black-and-grey striped thoraxes and a distinctive angle in the fourth longitudinal wing vein. Taxonomies concerning the domestic (and endophilic) variations of *Musca* sp. remain somewhat unclear. While it's widely accepted that they all belong to a single species, *M. domestica*, for a considerable time, four distinct subspecies were recognized:1) *M. domestica* Linnaeus, found across temperate zones globally, extending into subarctic and subtropical regions. 2) *M. domestica* vicina Macqvart, prevalent in subtropical and tropical zones across diverse regions, such as the Mediterranean, Asia, Africa, South and Central America, the Pacific, and Australia. 3) *M. domestica* nebulo Fabricius, exclusively found in tropical Asia. 4) *M. domestica* curviforceps Sacca and Rivosecchi, restricted to Africa, where it's the prevailing housefly south of the Sahara (Hulley, 2008).

Recent inquiries have indicated that all *Musca domestica* populations found outside of Africa should be classified under one subspecies, namely *M. domestica domestica*. This consolidation encompasses the previous three subspecies: domestica, vicina, and nebulo. These distinctions were primarily based on factors such as male compound eye size and abdominal pigmentation. However, these attributes exhibit overlapping traits and variation, not only across latitudinal gradients but also within specific climatic contexts, including altitude fluctuations. The behaviors and ecological patterns of housefly populations may diverge across global regions, adapted to local climatic and environmental factors. Nevertheless, it remains uncertain which, if any, of these biological variations are linked to morphological characteristics (WHO, 1991).

### 2.3.3 Morphology, feeding, and reproduction of houseflies

The housefly, typically measuring 4 to 9 mm in length, displays variations in size based on available resources. Its abdomen presents a color spectrum from grey to yellow, featuring a dark midline and sporadic dark markings (Welch, 2006). Houseflies are medium-sized insects, with a predominantly subdued grey hue. They showcase four black stripes on their thorax (Chapman, 1998; Moon, 2002). When at rest, their wings are positioned posteriorly, while a distinct bend appears in the fourth longitudinal wing vein. The thorax boasts four parallel dark stripes running vertically. Their legs bear sensory hairs (setae), with the last segment of each leg equipped with a pair of claws and a cushiony, adhesive pad named the pulvillus. This pad secretes a sticky substance that facilitates walking on smooth, vertical surfaces (Hedges, 1990; Sukontason, 2006). Males possess an abdomen with eight segments only, while females, usually slightly larger, are readily identifiable by observing the gap between their eyes, which is almost double in comparison to males (Welch, 2006). The female abdomen is composed of nine segments, with only the first five segments externally visible, and the remaining four typically retracted within the body, extending outward during oviposition (Chapman, 1998; Acharya, 2015).

Both male and female houseflies exhibit strong survival rates when nourished with a combination of water and sugar. However, in the case of females, the presence of protein or protein components (amino acids) is essential for the development of their eggs, while fat and other lipoids are not necessary for this purpose (Hussein et al., 2017). Houseflies display a versatile dietary range, encompassing human food, refuse, excrement (including sweat), and animal dung. Their olfactory receptors on the antennae are relatively limited, especially when compared to blow-flies. Consequently, long-distance attraction through odor plays a minor role in their feeding behavior. Instead, the flies locate food primarily through extensive random movements combined with visual cues, such as retraction to darker spots, and responses to humidity and smell at close proximity. This short-range approach is particularly effective, as many scents elicit a reaction. Houseflies are responsive to the scents of fermenting and putrefying substances, alcohols, lower aliphatic acids (like acetic acid), aldehydes, and esters. It is worth noting that these components may or may not be encompassed within the term "sweet smell" (Duistermars et al., 2009). However, they do not react to toxic substances like chloroform, formaldehyde, and certain organophosphorus insecticides (Abobakr et al., 2022).

Gustatory hairs present on their legs enable adult houseflies to perceive the taste of their food, leading them to often traverse over food items while at rest. To consume their meals, they typically need to liquefy and/or partially digest the food using salivary gland secretions that they regurgitate (Graczyk et al., 2001). For this purpose, they possess specialized sponging-type mouthparts designed to efficiently absorb liquid nourishment. Larvae, commonly known as maggots, employ mouth-hooks to filter-feed on bacterial masses (WHO, 2004), displaying a versatile capacity to consume various human and animal foods as well as waste. Houseflies are known for their high reproductive rates and can exploit a wide range of animal and plant-derived foods and waste materials. Mating in houseflies generally commences one to three days after emerging from the puparium. Males are drawn by a female sex pheromone called Z-9-tricosene (Acharya, 2015). These insects follow a polygamous mating system, although females primarily seek to mate with just one male as that is sufficient to fertilize all the eggs they will lay during their lifetime. Nonetheless, instances of females mating with more than one male are rare (ter Haar et al., 2023).

Gravid female houseflies lay small, elongated eggs that appear creamy white. These eggs are typically placed on damp, decaying organic matter. The most suitable substrates for both oviposition and maggot development generally have a moisture content ranging from 60% to 75%. Moreover, the act of oviposition in females is stimulated by volatile compounds emanating from decomposing organic materials (Cosse & Baker, 1996), as well as semio-chemicals released by symbiotic bacteria associated with the eggs (Lam et al., 2007). These bacteria, which proliferate on the egg surface, not only serve as cues for oviposition in other gravid flies but also enhance the quality of the substrate for larval growth (Lam et al., 2007; Acharya, 2015). In an insightful calculation of an unchecked housefly pair initiated reproduction in April, their population by August could escalate to a staggering 1.91 X $10^{20}$ flies (Welch, 2006). This estimate is equivalent to covering the earth's surface to a depth of 47 feet. Typically, a female fly deposits egg batches numbering from 75 to 159 eggs each, presented in distinct clusters. Reports indicate that houseflies tend to produce between two and twenty-one such egg batches, with intervals of eight to fourteen days. However, the norm is usually around two to four batches (Welch, 2006).

### 2.3.4 Biology and life cycle of houseflies

The eggs, resembling a banana in shape, measure approximately 1-1.2 mm in length and exhibit opal-white to cream hues. The female fly deposits clusters of these eggs within decaying, fermenting, or putrefying organic matter, provided that the environment maintains a suitable level of moisture, although it should not be overly liquid. The typical incubation period ranges from eight to 20 hours. Eggs that become excessively dry fail to hatch (WHO, 2004). The larvae undergo three distinct developmental stages referred to as instars (I, II, and III), each separated by molting processes. Instar I witnesses growth from 1 to 3 mm, instar II from 3 to 5 mm, and instar III from 5 to 12 mm. The larva's body shape is elongated and cylindrical, featuring a conical and tapering anterior end, while the posterior is more rounded, devoid of any appendages. Instars I, II, and the initial part of III display translucency, but as they approach the pupation stage, the larvae transition to a white or yellowish color. The later third-instar maggots, known as pre-pupae, are the ones most commonly observed during examinations of fly breeding sites. These larvae possess a pair of mouth hooks, one larger and one smaller, serving both feeding and locomotion purposes.

During instars I, II, and the initial part of III, the larvae are primarily engaged in feeding activities. In their natural environments, their diet primarily consists of bacteria or yeasts and the resulting decomposition products. These substances offer the essential protein (amino acids), B vitamins, and sterols required for their development. Throughout this feeding phase, the larvae are highly responsive to odors associated with their breeding materials. Upon ceasing their feeding activities during the third-instar phase, they transition into pre-pupae, migrating to cooler and drier locations. These locations can include the surface or sides of dung or garbage heaps, or even the surrounding soil. Pupation, which often takes place in large groups numbering hundreds or thousands, occurs in these selected spots (Justine et al., 2015).

The duration of larval development, from egg hatching to pupation, varies based on factors such as nutrition, moisture, and temperature. Under the most favorable conditions, notably at around 35°C, this process takes a minimum of 3-3.5 days. The pupal stage begins when the movements of the contracted larvae come to a halt, and they remain a cream color. This phase concludes with the emergence of the adult fly (WHO, 2004). As the puparium progresses, its color transforms from cream to a reddish-brown or black hue. The puparium generally measures about 6 to 7 mm in length and weighs approximately 21 mg. Remarkably, the pupal stage constitutes over half of the fly's immature development period. The duration of this stage spans from 3.75 to 28 days, contingent upon the larval stage's diet and the prevailing temperature conditions (Welch, 2006).

The emergence of the adult occurs as it breaks free from the puparium by utilizing the ptilinum located on the frontal region of its head. Employing the ptilinum, the fly contracts and subsequently exits the puparium, thus escaping the pupal medium, whether it be manure, debris and sand. An overview of the housefly's life cycle is depicted in Figure 2.1. For maturation, males necessitate a minimum of 16 hours, whereas females require at least 24 hours. However, the time for male maturation is no less than 20 hours, and female maturation demands at least 40 hours (Welch, 2006). The lifespan of houseflies is directly linked to temperature and humidity conditions. For instance, WHO (2004) observed that at an average temperature of 23.9°C, males live for an average of 33 days, while females survive for approximately 43 days, with a range spanning from one to 99 days. In contrast, at a temperature of 26.7°C and a relative humidity of 45%, male houseflies exhibited lifespans varying from 15 to 40 days, whereas female houseflies lived from 20 to 60 days (Justine et al., 2015).

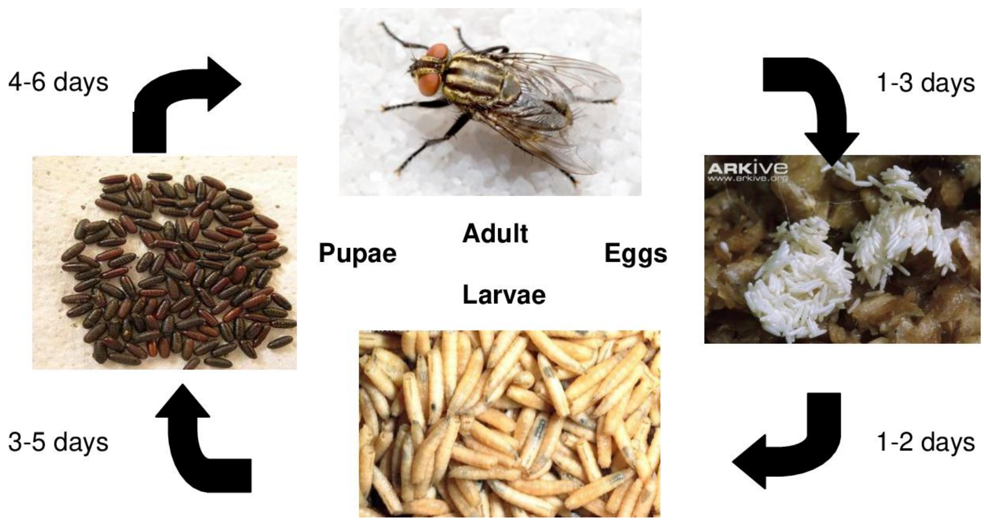


Figure 2.1 Life cycle of *Musca domestica* [Source: Justine et al. (2015)].

Poultry farms assume a significant role in the dispersion of flies, as noted by Gržinić et al. (2023). They observed that in the morning, houseflies tends to congregate in aisles and to a lesser extent in lateral vegetation. As the day progresses, their presence becomes more prominent in areas like manure, electric wiring, and lateral vegetation. By late afternoon, a majority of the flies settle on electric wiring, with only a small portion remaining in the aisles. During the evening, a large population of flies can be found on walls, cage stanchions, outer surfaces of cage feeders, and cage wires. At night, the flies tend to aggregate in greater numbers near the ends of houses in overhead resting areas rather than the central region. Tucci (2011) has highlighted that several studies indicate that *M. domestica* possesses a substantial flight range of 10 to 20 km and can travel at speeds of six to eight km per hour. However, observations reveal that these flies tend to explore a wide area before settling. Once they discover suitable food and shelter, they exhibit a tendency to remain within a range of 100 to 500 m from their breeding grounds.

## 2.4 Environmental factors associated with houseflies breeding in poultry farm

Accumulated poultry manure serves as an ideal breeding ground for houseflies, particularly in situations of inadequate overall cleanliness and high levels of moisture. While houseflies exhibit a preference for utilizing manure as their primary breeding substrate, instances have been documented where they engage in breeding within damp spilled feeds and other warm, decaying organic materials (Walker & Stachecki, 1996) The following are factors that determine successful housefly breeding and its population maintenance and sustainability in poultry farms.

### 2.4.1 Temperature

Houseflies behavior is influenced by shifts in environmental factors such as temperature, humidity, light levels, and wind velocity (Schou et al., 2013; Zahn, 2020). Optimal temperatures have been observed to enhance metabolic processes, impact developmental pace, lifespan, body dimensions, copulation, and reproductive yield in houseflies (Fletcher et al., 1990; Berger et al., 2008; Francuski et al., 2020). Furthermore, Schou et al. (2013) discovered that the locomotor activity of houseflies escalates as temperatures rise up to 35°C, after which the physiological functions of the houseflies begin to deteriorate, leading to a decline in activity levels. Among houseflies, males exhibit greater activity than females within the temperature range of >10°C and <40°C; however, the optimal temperature for houseflies appears to be around 30°C (as noted by Schou et al., 2013 and Zahn, 2020). Likewise, research has indicated that houseflies display a shortened period of sexual maturation, a reduced preoviposition period, a decreased interval between successive egg batches, and a notable increase in reproductive output at temperatures above 30°C (Francuski et al., 2020).

### 2.4.2 Wind speed

The airflow within closed-house farming systems significantly influences the spatial arrangement of flies (Geden et al., 1999). In closed environments, the behavior of houseflies in relation to wind patterns has been documented to vary: they have been observed dispersing against the wind at angles relative to the wind's direction, or even without any discernible correlation to wind direction (Zahn, 2020). In general, the flight activity of most flying insects tends to diminish when wind speeds exceed their maximum flight capabilities. In the case of adult houseflies, their documented maximum flight speed is approximately 2 m/s (Dahlem, 2009). Hence, housefly flight activity would not begin to decrease till wind speeds increased to some level above 2 m/s. Furthermore, when the wind speed exceeds 2 m/s flight can no longer be self-controlled and it may be restricted to near ground level (Zahn, 2020). Additionally, appropriate ventilation within the poultry houses also aids in reducing moisture and subsequently, decreasing flies reproduction (Axtell, 1990; Justine et al., 2015).

### 2.4.3 Ammonia and hydrogen sulfide

Temperature and pH are the two most important factors that affect ammonia release from poultry houses with accumulated manure and poultry litter (Elliot & Collins, 1982; Moore et al., 1995; Li & Xin, 2010; Wheeler et al., 2006). Pastor et al. (2011) and Berta et al. (2011) reported that ovipositing houseflies are notably attracted to ammonia, which naturally occurs in minor quantities within poultry manure. Moreover, Berta et al. (2011) reported that houseflies would travel a brief distance away from the precise ammonia source to deposit their eggs in a more suitable material.

Hydrogen sulfide has garnered significant attention as one of the volatile sulfur compounds linked to livestock farming emissions due to its notably low odor threshold ($H_2S$=10 ppb) and its adverse effects on human, animal health, and the environment. Microorganisms decompose sulfur-containing amino acids, like cysteine and methionine, under anaerobic conditions, resulting in intermediate sulfur-containing compounds that ultimately give rise to Hydrogen sulfide and other volatile sulfur compounds (Saksrithai & King, 2018). According to a study by Park et al. (2020), poultry farms primarily exhibited detectable concentrations of ammonia, with Hydrogen sulfide being absent in broiler chicken farms. However, its presence was measured within the range of 0.7 to 3.4 ppm at layer chicken farms.

### 2.4.4 Humidity

Hansson (1991) and Zahn (2020) hinted that many insects are sensitive to the changes in the weather. Especially, humidity has been noticed to have strong impact on houseflies behavior in the tropic. With reference to that, significant rise in housefly mortality rate was seen, when relative humidity exceeds 80% while longevity was extended when the temperatures are over 20°C and humidity levels around 50%. Housefly diurnal flight activity is negatively correlated with humidity. Besides that, cuticular hydrocarbons production by both males and females of *M. domestica* under very wet conditions (90% relative humidity) was rapid while a delay by 3 days in the production was documented at 50% and 20% relative humidity after emergence from the pupa (Zahn, 2020).

### 2.4.5 Manure

Malone (1992) provided an estimate for the average rate of broiler litter production, indicating 1.0 dry metric ton per 1,000 broilers per flock, with a variability spanning from 0.7 to 2.0 metric tons. Conversely, Chamblee & Todd (2002) put forth an estimation for broiler litter production in Mississippi, proposing 1.6 tons per 1,000 broilers when houses underwent complete cleaning on an annual basis. In cases where houses were entirely cleaned after two years, the rate was suggested to be 1 ton per 1,000 broilers (as cited by Coufal et al., 2005). The poultry farm is an ideal reproductive environment for housefly populations. Conditions favorable for fly breeding persist year-round owing to the buildup of manure and regulated temperatures within poultry houses, as described by Kaufman et al., (2000). The larger the production capacity of a farm, the higher the volume of manure and used litters are generated. Thus, the higher the chances for houseflies breed on these materials and consequently results in the expansion of fly populations (Axtell, 1999; Justine et al., 2015).

While the expansions of poultry farms cater a suitable environment for fly breeding, the poultry manure itself may serve as the best choice of breeding substrate for houseflies. In a comparative investigation undertaken by Khan et al. (2012), it was observed that housefly larvae reared on poultry manure exhibited accelerated development compared to those raised on alternative host manures (including buffalo,