

Environmental effects on behavioral and physiological traits of the housefly *Musca domestica* in the transmission of pathogenic bacteria

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Abstract

This study was focused in the evaluation in the effects of temperature and relative humidity on behavioral and physiological characteristics of *Musca domestica*, as well as the role played by it on the mechanical transmission of pathogenic bacteria under controlled laboratory conditions. Adult houseflies caught in the urban area were reared in the laboratory and kept at different temperatures ($20 \pm 2^\circ\text{C}$, $25 \pm 2^\circ\text{C}$, and $30 \pm 2^\circ\text{C}$) and relative humidity level ($40 \pm 5\%$, $60 \pm 5\%$ and $80 \pm 5\%$)

Behavioral parameters such as locomotor activity, frequency of flying, number of landings and duration of contact with surfaces were recorded while physiological indicators such as survival rate, general activity and visible stress responses were monitored on day-to-day basis. Samples from external body surface, mouthparts and digestive system were checked for the presence of the bacteria using the standard microbiological method and the results of the analyses were analyzed by means of univariate analysis of variance with a level of significance of $P \leq 0.05$. The results showed that the behavioral activity increased with increasing temperature and moderate to high humidity when compared to rates of physiological stress and mortality which were higher under extreme environmental conditions. In addition, the percentage of bacteria positive samples improved significantly with the increase of the temperature and the humidity level, showing a positive correlation between the environmental condition, the fly activity and the potential of the mechanical transmission of pathogen bacteria, with all of the observed differences being statistically significant ($P \leq 0.05$).

Keywords: *Musca Domestica*; Temperature; Relative Humidity; Behavior; Physiology; Bacterial Transmission

1. Introduction

The housefly (*Musca domestica*) is one of the most common synanthropic insect as it lives very close to the human settlement in all parts of the world [1]. Due to its broad distribution in the urban environment, including resident habitat, markets, food processing and waste disposal, is a major public health concern [2]. Housefly are opportunist feeders, and can feed on decaying organic matter, animal waste and food of humans, which exposes them to the many pathogenic microorganisms [3]. As a consequence, they act as mechanical vectors, involved in the transfer of bacteria, viruses and protozoa from contaminated substrates to human and animals living environment [4].

Among the microorganisms that houseflies can transmit, bacteria like *Escherichia coli*, *Salmonella* spp. and *Staphylococcus aureus* and *Shigella* spp. are of special concern since of their importance for human health [5]. The transmission takes place mainly mechanical transmission by adhesion of pathogens on the outer surface of the fly or through regurgitation contamination, or defecation on food and surfaces [6]. Therefore, the housefly is an important vector in the epidemiology of foodborne and environmental diseases [7].

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Environmental factors which are important to determine the physiology and behavior of houseflies is the temperature, the relative humidity. It impacts on the survivability rates, flight behavior feeding rate, mating rate and fitness [8]. Environmental factors which are important to determine the physiology and behavior of houseflies is the temperature, the relative humidity [9]. It impacts on the survivability rates, flight behavior feeding rate, mating rate and fitness [10].

There is need of knowledge on how the environmental factors interact with the behavior of the housefly in disease control and intervention on the health of the populace [11]. The study of how temperature and humidity affect flies' activity and bacteria carriage could help in the control of pests in urban areas like congested communities or shortage of sanitation centers [12].

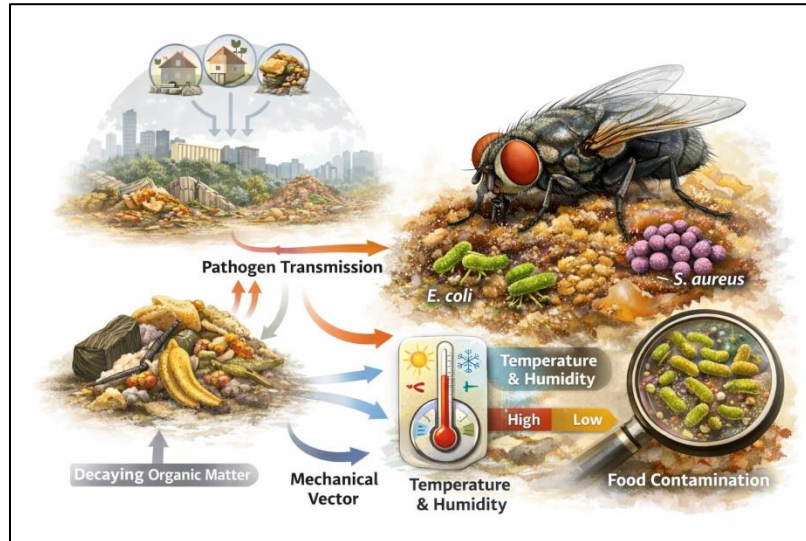


Figure 1 Housefly (*Musca domestica*) as a mechanical carrier of bacteria, which has the temperature and humidity impact

2. Materials and Methods

2.1. Study Design

The experimental design used in this study was the factorial one in order to examine what kind of impact two significant environmental factors, temperature and relative humidity have on the behavioral and physiological aspects of housefly (*Musca domestica*) and whether it is efficient or not a mechanical carrier of pathogenic bacteria.

The experiments were conducted under controlled laboratory conditions and other factors were held constant in order to allow the potential of reducing the confounding effects.

2.2. Collection and Identification of Houseflies

In the various cities, adult houseflies were collected using clean net traps in various locations of high or human activities (e.g. waste dumps and local markets) [13].

The specimens were forwarded to the laboratory in capped specimen containers and taxonomically characterized using standard entomological taxonomic keys on morphological features (shape of head, wings, mouthparts) of the specimen [14].

To reduce the variation, the experiments were carried out only on the visually healthy adults and of the same age group.

2.3. Laboratory Rearing of Houseflies

The flies utilized were maintained in insect cages within a particular laboratory and were allowed to mature after an acclimatization phase of 12 hours under normal conditions of the environment which comprised average temperature ($25 \pm 2^\circ\text{C}$) and relative humidity ($60 \pm 5\%$) and 12 hours of light: 12 hours dark photoperiod so that they could be as close as possible to actual conditions.

Fly was provided with good food and a source of fresh water. Status of health was monitored on a daily basis and, dead or inactive persons were not included at the start of experiments [15].

2.4. Environmental Treatments (Temperature and Humidity)

After the acclimatization, flies were randomly introduced to experimental groups where low ($20 \pm 2^\circ\text{C}$), moderate ($25 \pm 2^\circ\text{C}$) and high ($30 \pm 2^\circ\text{C}$) temperatures and low ($40 \pm 5\%$), medium ($60 \pm 5\%$) and high ($80 \pm 5\%$) relative humidity were employed [16].

These conditions were kept using environmental incubators with an absolute control mechanism and actual values were noted in between to ensure similar experimental conditions throughout the study [17].

2.5. Behavioral Assessment

The housefly behavioral parameters were tested to each of the environmental treatments and the locomotor activity as well as the frequency of flight, number of landings and the duration of contact with various surfaces were measured [18].

The regular and fixed daily intervals observed were -3 times a day and the direct observation and the use of visual documentation which was done where necessary to make the observations accurate [19].

Behavioral data were quoted in the form of mean values which were taken of each of the environmental treatments.

2.6. Physiological Assessment

Physiological measurements were also conducted to show how the houseflies responded to the environmental stressor of the house flies namely; the survival rate, the overall level of activity and signs of visible stresses [20].

These responses were checked and recorded on a daily-basis at the period of exposure to various environment treatments and the outcomes were presented as means of each group [21].

2.7. Evaluation of Housefly Role in Mechanical Bacterial Transmission

In order to determine the potential of mechanical transmission of house flies (*Musca domestica*), the samples were taken by means of sterile methods to refer to the external surface of the house flies, mouthparts, and digestive tract.

Standard microbiological analysis of samples was done to identify the existence of pathogenic bacteria and preliminary characterization of bacterial isolates was done on morphological basis and general diagnostic tests, without conducting any elaborate molecular studies.

2.8. Statistical Analysis

All data was analyzed in a standard statistical software package. A mix of analysis of variance (ANOVA) was used to investigate the effect of the temperature, humidity, and their interaction on the various measured variables followed by post hoc comparisons if required. Results were significant at a level of $P \leq 0.05$.

3. Results

The results from the studies showed that the effects of temperature and relative humidity were significant on behavior and physiological features of housefly (*Musca domestica*), and their efficiency as mechanical vector of pathogenic bacteria.

Microbiological samples analysis was performed in standard manner to determine the presence of pathogenic bacteria and rudimentary characterization of bacterial isolates was done on morphological basis and general diagnostic tests, but no elaborate molecular studies were performed. It was found to be the highest at high temperature ($30 \pm 2^\circ\text{C}$) and the lowest at low temperature ($20 \pm 2^\circ\text{C}$) [22]. Statistical finding proved that there were significant differences in the other the temperature treatments ($P \leq 0.05$). These behavioral indicator graphical results are represented in figure 2 of the temperature treatments.

Table 1 Effect of temperature on behavioral traits of houseflies (*Musca domestica*)

Temperature (°C)	Locomotor Activity	Flight (flights/h)	Frequency	Number Landings	of	Contact Duration (s)
20 ± 2	2.1 ± 0.3	6.4 ± 1.1		4.2 ± 0.8		12.5 ± 2.1
25 ± 2	3.6 ± 0.4	12.8 ± 1.6		9.5 ± 1.2		21.3 ± 3.0
30 ± 2	4.2 ± 0.5	16.3 ± 2.0		12.7 ± 1.5		28.6 ± 3.8

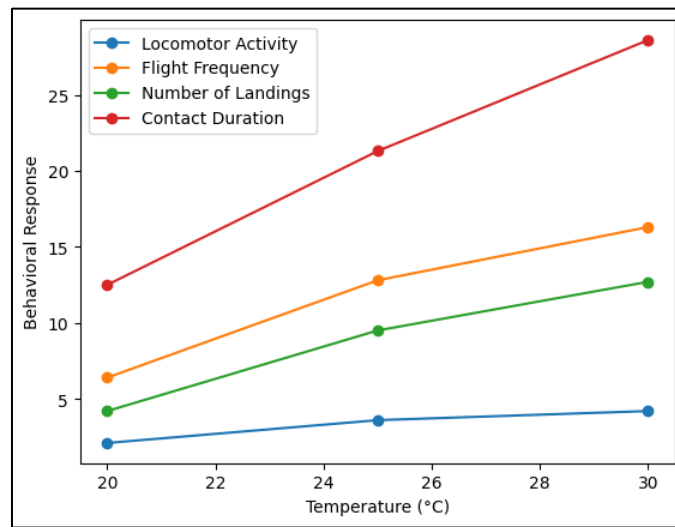
**Figure 2** Influence of temperature on selected behavioral traits of the housefly (*Musca domestica*)

Table 2 shows the impact of the relative humidity on behavioral characteristics. The presence of moderate and high relative humidity (60-80%) significantly contributed to the growth of all behavioral parameters as compared to the low level (40%) condition. [23]. Statistical analysis results revealed that there are significant differences among different degrees of humidity ($P \leq 0.05$). Figure 3 Graphical relationship between relative humidity and house fly (behavioral activity).

Table 2 Effect of relative humidity on behavioral traits of houseflies (*Musca domestica*)

Relative Humidity (%)	Locomotor Activity	Flight (flights/h)	Frequency	Number Landings	of	Contact Duration (s)
40 ± 5	2.3 ± 0.4	7.2 ± 1.0		5.1 ± 0.9		14.8 ± 2.4
60 ± 5	3.7 ± 0.5	13.4 ± 1.7		10.3 ± 1.3		23.5 ± 3.2
80 ± 5	4.0 ± 0.6	15.1 ± 1.9		11.6 ± 1.4		26.9 ± 3.6

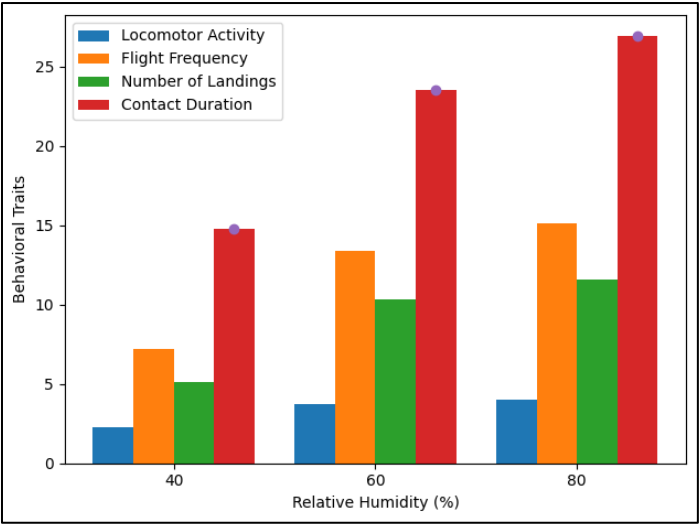


Figure 3 Influence of relative humidity on selected behavioral traits of the housefly (*Musca domestica*)

When it came to physiological measures, Table 3 below showed that the maximum survival rates were achieved under moderate temperature (25 ± 2 °C) as compared to high temperature (30 ± 2 °C) with a clear demonstration of stress and reduced overall activity.

It was statistically analyzed that temperature treatments differed significantly ($P \leq 0.05$). Figure 3 represents graphical influence of the temperature to the physiological parameters.

Table 3 Physiological effects under different temperature treatments

Temperature (°C)	Survival Rate (%)	General Activity	Stress Signs
20 ± 2	92 ± 3	Medium	Low
25 ± 2	96 ± 2	High	Low
30 ± 2	78 ± 4	Low	High

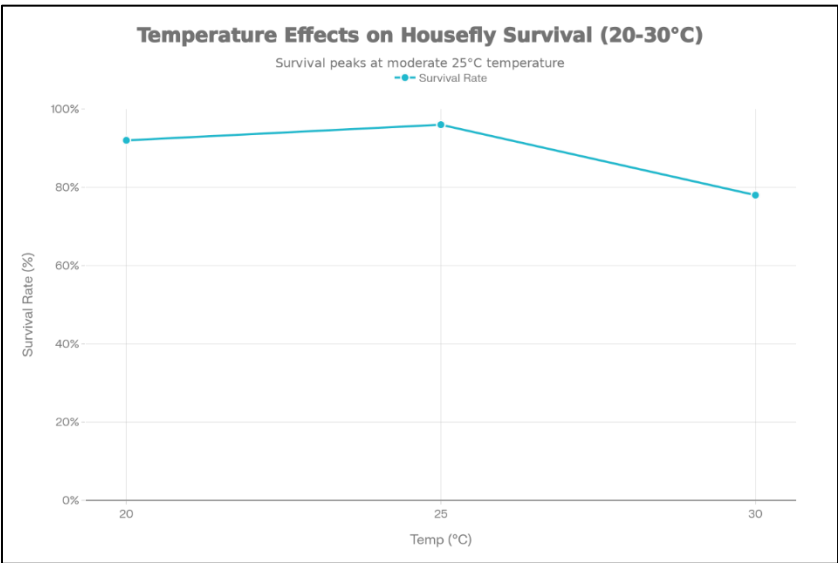


Figure 4 Temperature effects on housefly survival

Table 4 reveals the effect on physiological indicators of the relative humidity that produces the maximum survival and high activity at the middle level of relative humidity ($60 \pm 5\%$), and the high humidity leads to the lower survival and

complicated expression of stress. The results of statistical analysis revealed significant differences in humidity treatments ($P \leq 0.05$). Figure 4 shows the graphical report of the physiological parameters of different humidity levels.

Table 4 Physiological effects under different relative humidity treatments

Relative Humidity (%)	Survival Rate (%)	General Activity	Stress Signs
40 ± 5	88 ± 3	Medium	Moderate
60 ± 5	95 ± 2	High	Low
80 ± 5	81 ± 4	Low	High

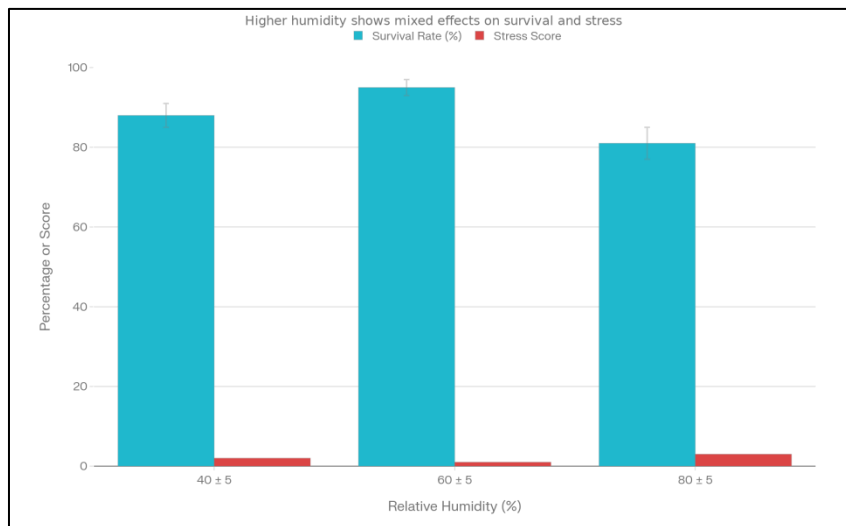


Figure 5 Physiological effects under different relative humidity levels

With regards to the role of the houseflies in mechanical bacterial transmission Table 5 shows different proportions of bacterial isolates of the outer surface, mouthparts and digestive system; although, a huge increase of positive samples is observed in high environmental conditions (30°C – 80% RH). Statistical analysis demonstrated that the environmental treatments have significant differences ($P \leq 0.05$). The graphical representation of the percentage of bacteria isolation by temperature and humidity is represented in Figure 5.

Table 5 Percentage of positive bacterial samples (%) under different environmental treatments

Environmental Treatment	External Surface	Mouthparts	Digestive System
20 °C – 40% RH	32	26	18
25 °C – 60% RH	58	46	35
30 °C – 80% RH	74	63	51

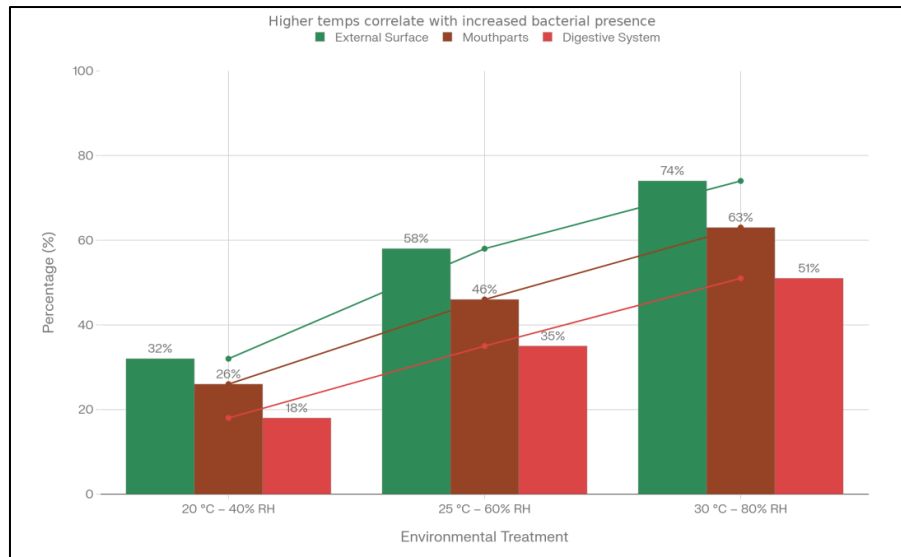


Figure 6 Bacterial Contamination Levels in Insects under Varying Temperature and Humidity

Comprehensively, these findings reveal that the environmental factors particularly high temperatures and humidity elevate the behavioral activity in the house flies with imposed physiological stress, and also amplify the capacity of the flies in carrying and transmitting pathogenic bacteria to other organisms.

4. Discussion

The present experiment demonstrated that temperature and relative humidity have great influence on the behavioral and physiological characteristics of housefly (*Musca domestica*)- the study correlates with the previous researches which placed environmental factors as the most significant determinants of activity and interrelations of the insects. [24].

There was also an increase in the behavioral data that showed locomotor activity, frequency of flight, the number of landings, and duration of contact with the higher temperature and moderate to high humidity levels [25]. This implies that the positive influences of the environment enhance the contacts between the flies and their environment which incorporates food and the potential surfaces that can be exposed to microbial contamination [26]. These results are in harmony with other reports which have revealed the stimulation effect of temperature and humidity on insect activity [27].

The physiological measurements indicated that when the environment was moderate, survival was recorded at the highest rate and when the environment was hot, or humid, the survival rate recorded was low and the stress levels were high [28]. This is an indication that extreme levels of environmental factors lead to physiological stress and this reduces the fitness of houseflies, as earlier researches have determined the impact of temperature and humidity levels to the survival and activity of insects [29].

Regarding the mechanical bacteria transmission, the findings indicated that higher bacterial isolation collection was on the exterior part, mouthparts and digestive system of flies in higher temperature and humidity [30]. This implies that there are environmental parameters which appear to enhance fly activity and that the same parameters enhance the chances of surface contamination and bacterial transfer, which is in line with earlier studies that found the connection between insect activity and microbial transfer [31].

In general, these findings suggest that temperature and humidity interactively affect the behavior and physiology and has direct relevance to the performance of houseflies as mechanical vectors of pathogenic bacteria [32]. This interaction identifies the intricacy in the bio-relationship between environmental factors, insect behaviour as well as pathogens transmission, and it shows the significance of environmental control in highly populated regions in which humans reside, in a bid to reduce the transmission of the pathogen [33].

Judging by these findings, environmental control (in the form of house temperature and humidity), is a significant measure that can help reduce housefly activity and bacterial transmission to improve the well-being of the populace, and fewer cases of mechanically transmitted diseases [34].

5. Conclusion

Behavioral and physiological responses of *Musca domestica* also greatly rely on the environmental factors that directly affect the potential of the insect to be a mechanical host to pathogenic bacteria. The climate and moisture in the areas of high population might be regulated to reduce the number of flies and limit the spread of microbes, thereby promoting the measures of the population health.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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