Extraction And Evaluation Of Antidotes From *Musca Domestica* (Houseflies) Wings Against Its Body Pathogens

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Abstract

Musca domestica is an important insect vector of various myriads of microbes ranging from bacteria, fungi and parasites. They tend to infest food and drinks of human and animals causing severe diseases. Unfortunately, some of the microbes/diseases transmitted by this vector have developed resistance against various drugs (Drug Resistance Microbes). This current study evaluates the microbial loads on various body parts (Right wing, Left wing and Body Surface) of M. domestica captured from different locations in Ilaro, Ogun State and the antimicrobial properties of the body parts. One hundred and twenty samples of M. domestica were trapped from different locations including abattoirs, food vendors, house hold waste bin area and palm wine shops. Trapped flies were examined for Total Bacteria Count (TBC), Total Coliform Count (TCC) and Total Fungi Count (TFC) and the presence of antimicrobial agent on their body parts. Highest TBC and TCC $(2.42x10^2 \text{ and } 1.47x10^2)$ were found in flies trapped in Palm Wine Body Parts (PBP), while highest TFC $(2.40x10^2)$ was found in flies captured in Abattoir Body Parts (ABP). No bacteria, coliform and fungi growth were observed in the right wing of flies trapped in the four locations except the Food-Vendor Right Wings-0.18x10². (FRW). Cultural and biochemical characteristics of the bacteria isolate shows the presence of Proteus spp, Salmonella spp, Escherichia coli, Staphylococus aureus, Bacillus spp and Klebsiella spp. No bacteria growth was found in Palm-wine Right wings (PRW). No fungi growth was found in the PRW, House-hold Right wings and FRW. Other body parts carry various fungi including Mucor spp, Aspergillus spp, Fusarium spp, Rhizopus spp and Penicillium spp. Statistically, antimicrobial test of the water and ethanolic extract of the right-wing against test organism shows no significant difference (p>0.05) when compared. Dimethyl Sulphoxide (DMSO) shows no significant effect (p>0.05) against all the test organisms. Significantly (p<0.05), Ciprofloxacin shows better effect than the water and ethanolic extracts of the right wings as it exhibited the highest zone of inhibition (30.33 ± 0.58) and sensitivity against all the organisms. The right-wing extracts can be examined for other antimicrobials properties and can be enhanced to develop natural and novel antimicrobial agent for controlling microbes in food and water.

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I. Introduction

The emergence of drug resistant pathogens is a major problem in the medical world. They tend to present a great deal of threat to health due to the emergence of new strains which ruthless effect surpass the common antibiotics (Lee *et al.*, 2012). Many of these pathogens mutate from their usual form making it a thorn in the neck for medical researchers and practitioners to combat with common antibiotics. The growing concern about drugresistant microbes instigated scientists and researchers to intensify their search for natural novel alternative compounds that could be remedial to drug-resistant pathogens. The outcome of various concurrent field practical works by researchers birthed the use of natural compounds from insects. The search for these antimicrobial compounds from unusual natural sources will procreate potentially useful leads in the identification of new drugs and compounds (Lee *et al.*, 2012).

Insects are termed the largest group of living organisms (Feng *et al*, 2009) and are considered an underutilized and underexplored source of potential compounds and toxins which can be highly beneficial in modern medicine for the treatment of various ailments. In ancient China, insects are used in mystic art for the treatment of various uncommon ailments which underrates the efficacy of common antibiotics (Chou 1980; Zou 1982). Various insects such as bees, spiders, flies, cockroaches among others have been researched to have healing potentials. Insects parts (exuvium, wings), products (eggs, egg shell) and secretions have also been reported to have medicinal values (Feng *et al.*, 2009). Their healing potentials are associated to the presence of various antimicrobial compounds such as lysozyme, diptericin, colicin, defensin etc.

Houseflies are vectors for the spread of varying forms of diseases from dirty places to food or drinks. They are rated among the top three major insect vectors transmitting myriads bacteria, viruses and parasites that cause vector borne diseases in human, animals and plants (Byron, 2022). But despite their notorious nature and undesirable menaces caused by this insect vector, few researches have demonstrated their benefits in the medical world. *Musca domestica* is a cosmopolitan insect which is considered to possess various attributes which can ameliorate diseases and also serve as bioactive agent in the development of medicines. Few literatures have stated the neutralizing effect of one of the wings carrying the antidote for the deadly microbes embedded in the other wing (Rehab, 2014; Claresta *et al*, 2019; Asril *et al*, 2021)

Hence, the quest to extract and evaluate these antidotes from the wings and measure the time range to neutralize the pathogen has necessitated this study.

Study Site

II. Materials And Methods

The research work was conducted in the Science Laboratory Technology Department at The Federal Polytechnic, Ilaro, Ogun State, Nigeria.

Materials

1. Reagents:

Distilled water, Ethanol, Kovac's reagent, Normal saline, Gram stains, Hydrogen peroxide and Ludol's Iodine. 2. Equipment:

Autoclave, weighing balance, measuring cylinder, incubators, spatula, Spint, Petri-dishes, Inoculating loop, Beaker, Conical flask, Cork borer, Spirit lamp, Rotary evaporator, Shaker and Incubator.

3. Microbiological media:

Nutrient agar, MacConkey agar, Mueller Hinton agar and Potato dextrose agar

4. Miscellaneous:

Cotton wool, Aluminum foil, Ruler, weighing balance, Measuring cylinder, Filter paper, hand gloves, filter paper

Methodology

Collection of Flies

The project was conducted from June to August, 2024 in Ilaro town, Ogun State. One hundred and twenty samples of adult houseflies (*Musca domestica*) was obtained from different locations such as abattoirs, food vendors, house hold waste bin area and palm wine shops. The flies were collected using sweep net over a surface where houseflies converge. The flies were later constrained into labelled constructed cages (Ahmad *et al.* 2016).

Isolation of External Parasites

Flies collected were transferred into a well labelled specimen bottle with information such as date and location of the isolate using an aspirator. Deep freezing method was used to kill flies. Right and left wing of the flies' samples were detached and placed in a disinfected petri-dish for identification and to ascertain which of the two wings carry microbes and parasites.

Bacteriological analysis of the parasites

Wings and body of the houseflies were transferred into plain bottles and 5mls of sterile normal saline was added to the bottles and centrifuged for 15min at 3000rpm. 0.5ml of each sample was placed in labelled plain bottles containing 5ml of normal saline. The bottles were shaken vigorously to expel the microbes attached to the surface of the wings into the saline solution. Following the discard of the microbes, bottles containing normal saline solution and labelled appropriately for further use.

x10 serial dilution was done and 1ml of the inoculum from the original bacteria stock was collected and aseptically transferred into the first dilution bottle. Sample was diluted 3times in order to achieve an acceptable colony count. Bottles were closed immediately to avoid contamination of samples. Following this, 1ml of the inoculum was aseptically collected with a sterile syringe and cultured on 20ml Nutrient agar., potato dextrose agar and McConkey agar and was shake gently to evenly spread the inoculums in the medium. Plates were allowed to solidify, inverted and incubated at 35°C for 24hrs. Following the incubation period, plates were observed for the number of colonies. The number of colonies (total colony forming units in grams) were recorded and expressed in TCFUg⁻¹. The colonies further sub-cultured on fresh nutrient agar plate. The sub-cultured plates were inverted and incubated at 37°C for 24hrs to obtain pure isolates. Pure isolates obtained were subjected to several biochemical, motility test and gram stain following the standard procedure described by (Cheesbrough, 2023).

Isolation of Microorganisms

Each colony was isolated in a pure form by sub-culturing. Distinctive morphological properties of each pure culture such as colony form, elevation of colony and colony margin were observed. At the expiration of incubation, colonies were counted at 35^oC within 24hrs using colony counter. Count were expressed as colony forming unit ml⁻¹. Further microbial identification was done using the methods of (Akogun and Badaki, 1998).

Antibiotic Susceptibility Test

This was carried out using perforated filter paper contained in Ciprofloxacin. A colony of the test organism was inoculated into peptone water using a sterile wire loop. The turbidity was then compared against a reference 0.5 McFarland standard tube. The suspension was streaked on the surface of nutrient agar plate and the antibiotic disc was placed on it using a syringe. The plate was incubated at 37^oC for 24hrs. Zone of inhibition generated by the antibiotic disc was grouped as susceptible and resistant.

Statistical Analysis

The proportion of houseflies collected from each location and the frequency of the bacteria and parasite isolate from the housefly were analyzed using Statistical Package for Social Science (SPSS).

III. Results

Presented in table 1 below is the colony of microbial counts of left wings, right wings and body part of houseflies trapped in various locations. The result showed significant bacterial and fungal contamination of the various parts examined. The palm wine left wing (PLW) sample exhibited a total bacterial count (TBC) of 2.08 $\times 10^2$ cfu/mL, total coliform count (TCC) of 1.16 $\times 10^2$ cfu/mL and a total fungal count of (TFC) of 0.22 $\times 10^2$ cfu/mL.

Conversely, the palm wine body parts (PBP) showed a TBC, TCC and TFC of 2.42×10^2 cfu/mL, 1.41×10^2 cfu/mL and 0.68×10^2 cfu/mL respectively. In marked contrast, no growth (NG) was observed for palm wine right wings (PRW). TBC of 1.64×10^2 cfu/mL, TCC of 1.08×10^2 cfu/mL and TFC of 0.53×10^2 cfu/mL was observed in the household body parts (HBP). Significantly, no growth was exhibited in the TBC and TFC of household right wings (HRW) except a TCC of 0.28×10^2 cfu/mL.

As for flies trapped in Abattoir, The Total Bacterial Count (TBC) ranged from 0.23×10^2 to 1.00×10^2 cfu/ml with the highest count found on Abattoir Body Parts (ABP). On the other hand, the Total Coliform Count (TCC) varied between 0.70×10^2 and 1.20×10^2 cfu/ml and Total Fungi Count (TFC) ranging from 0.11×10^2 to 2.40×10^2 with ABP indicating the highest value in both. No growth was observed for TCC on ARW.

Flies captured at Food Vendor location showed significant variations in their various body parts. The Bacteria Count (TBC) exhibited a notable range of 0.18 x 10² to 1.86 x 10²cfu/ml, manifesting variable bacterial loads across different body parts of the flies with Food Vendor Body Parts (FBP) harboring the highest bacteria count of 1.86 x 10²cfu/ml. However, Total Coliform Count (TCC) varies from 0.81 x10² to 1.24 x10² cfu/ml. Food Vendor Body Part (FBP) and Food Vendor Left Wing (FLW) showed comparable TCC values, indicating potential contamination sources. Specifically, No Growth (NG) was observed in Food Vendor Right Wing (FRW) implying minimal coliform presence on this body part.

Total Fungi Count (TFC) exhibited substantial fungi contamination. Count ranged from 1.26×10^2 to 1.92×10^2 cfu/ml with FBP revealing the highest count of 1.92×10^2 cfu/ml. No fungi growth was observed in the FRW. In summary, PBP exhibited the highest TBC and TCC among all flies captured in different locations, while ABP and ALW revealed the highest fungal count. In contrast, FRW showed the least TBC with no growth observed in PRW, HRW, ARW AND FRW.

	Table 1: Colony of	microbial counts	
Plates	TBC (cfu/ml)	TCC (cfu/ml)	TFC (cfu/ml)
PLW	2.08 x 10 ²	1.16 x 10 ²	0.22 x 10 ²
PBP	2.42 x 10 ²	1.47 x 10 ²	0.68 x 10 ²
PRW	NG	NG	NG
HLW	1.91 x 10 ²	0.98 x 10 ²	$0.42 \ge 10^2$
HBP	$1.64 \ge 10^2$	1.08 x 10 ²	0.53 x 10 ²
HRW	NG	$0.28 \ge 10^2$	NG
ALW	$0.80 \ge 10^2$	0.72×10^2	2.24 x 10 ²
ABP	$1.00 \ge 10^2$	$1.20 \ge 10^2$	2.40 x 10 ²
ARW	0.23 x 10 ²	NG	0.11 x 10 ²
FLW	$1.09 \ge 10^2$	0.81 x 10 ²	1.26 x 10 ²
FBP	1.86 x 10 ²	1.24 x 10 ²	$1.92 \ge 10^2$
FRW	0.18 x 10 ²	NG	NG

Table 1: Colony of microbial counts

KEYS: TBC- TOTAL BACTERIAL COUNT TCC- TOTAL COLIFORM COUNT

TFC- TOTAL FUNGAL COUNT

PLW- PALMWINE LEFT WINGS PBP- PALMWINE BODY PARTS PRW- PALMWINE RIGHT WINGS

HLW- HOUSEHOLD LEFT WINGS HBP- HOUSEHOLD BODY PARTS HRW- HOUSEHOLD RIGHT WINGS

FLW- FOOD VENDOR LEFT WINGS FBP- FOOD VENDOR BODY PARTS FRW- FOOD VENDOR RIGHT WINGS

ALW- ABATTOIR LEFT WINGS ABP- ABATTOIR BODY PARTS ARW- ABATTOIR RIGHT WINGS

NG- NO GROWTH

Table 2 revealed the various bacteria isolated from each part of houseflies trapped from different locations. The cultural and biochemical characteristics of bacterial isolates from houseflies' body parts revealed diverse microbial populations. Bacterial isolates from Palmwine Left Wings (PLW) and Abattoir Left Wings (ALW) exhibited similar cultural characteristics, with thin cream to gray colonies and rod-shaped morphology, suggesting the presence of *Proteus spp*.

However, Palmwine Body Parts (PBP) and Food Vendor Body Parts (FBP) isolates shared characteristics consistent with *Salmonella spp* and *Bacillus spp*, respectively. Household Left Wings (HLW) and Household Right Wings (HRW) isolates displayed similar properties, indicating the presence of *Klebsiella spp*. Abattoir Body Parts (ABP) and Food Vendor Right Wings (FRW) isolates shared characteristics with *Staphylococcus aureus*. *E. coli* was suspected in isolates from PBP, Abattoir Right Wings (ARW), and Food Vendor Left Wings (FLW). Meanwhile, *Vibrio spp* was identified in Food Vendor Body Parts (FBP) isolates.

These findings highlight the potential role of houseflies in transmitting various bacterial pathogens.

Locati on	Isolates	Cultural Characteristic	Shape	Gram Staini	Citrat e	Urea se	Catalas te	Indo le	Suspected Organism
		s		ng	Test	Test	Test	Test	
PLW	A	Thin cream to gray colour	Rods	-	-	+	+	+	Proteus spp
	В	Abundant, Opaque, golden yellow	Coccu s	+	-	-	+	-	Staphylococcus aureus
PBP	Α	White moist	Rods	-	-	•	+	+	Escherichia coli
	В	Dark sheen on SS agar	Rods	-	-	•	+	-	Salmonella spp
PRW	NG	NG	NG	NG	NG	NG	NG	NG	NG
HLW	Α	Slimy White, Raised elevation	Rod	-	+	+	+	-	Klebsiella spp
HBP	A	Dark sheen on SS agar	Rods	-	-	-	+	-	Salmonella spp
	В	Greyish –white colony	Rods in chain	+	+	-	+	-	Bacillus spp
HRW	Α	Slimy White, Raised Elevation	Rod	-	+	+	+	-	Klebsiella spp
ALW	A	Thin cream to gray colour	Rods	-	-	+	+	+	Proteus spp
	В	White moist	Rods	-	-	•	+	+	Escherichia coli

Table 2: Cultural and biochemical characteristics of bacterial isolates

-									
ABP	Α	Abundant,	Coccu	+	-	-	+	-	Staphylococcus aureus
		Opaque,	s						
		golden yellow							
	Α	Slimy White,	Rod	-	+	+	+	-	Klebsiella spp
		Raised							
		Elevation							
ARW	Α	Greyish -white	Rods	+	+	-	+	-	Bacillus spp
		colony	in						
			chain						
FLW	Α	White moist	Rods	-	-	-	+	+	Escherichia coli
FBP	Α	Greyish -white	Rods	+	+	-	+	-	Bacillus spp
		colony	in						
			chain						
	В	Yellowish	Rod	-	+	-	+	+	Vibrio spp
		colony							
	С	White moist	Rods	-	-	-	+	+	Escherichia coli
FRW	А	Abundant,	Coccu	+	-	-	+	-	Staphylococcus aureus
		Opaque,	S						
		golden yellow							

Keys: PLW- PALMWINE LEFT WINGS PBP- PALMWINE BODY PARTS PRW- PALMWINE RIGHT WINGS

HLW- HOUSEHOLD LEFT WINGS HBP- HOUSEHOLD BODY PARTS HRW- HOUSEHOLD RIGHT WINGS

ALW- ABATTOIR LEFT WINGS ABP- ABATTOIR BODY PARTS ARW- ABATTOIR RIGHT WINGS

FLW- FOOD VENDOR LEFT WINGS FBP- FOOD VENDOR BODY PARTS FRW- FOOD VENDOR RIGHT WINGS NG- NO GROWTH

Table 3 revealed the cultural and microscopic characteristics of fungal isolates. The isolates from Palmwine Left Wings (PLW) and Household Body Parts (HBP) were identified as Aspergillus spp. based on their umbonate elevation, dark green color, and rapid growth. Palmwine Body Parts (PBP) isolates were classified as Mucor spp. With the exhibition of raised elevation, grey color, and very rapid growth. Household Left Wings (HLW) isolates suspected Rhizopus spp., characterized by umbonate elevation, spongy dark brown color, and rapid growth.

Isolate B matched Penicillium spp., exhibiting umbonate elevation, light green color, rapid growth, and septate hyphae. No fungal growth was observed for Household Right Wings (HRW) and Palmwine Right Wings (PRW). These findings indicate fungal contamination with potential health implications.

			CULTU RAL							MICROS COPY		
Samp	Isol	Elevation	Margin	Colon	Grow	Reverse	Hyphae	Conidia	Conidiosp	Vescicle.	Conidi	Suspected
les	ate			(PDA)	th Rate	Side		head	ores	shape	a shape	organism
PLW	A	Umbonate	Entire	Dark	Rapid	Yellow	Non-	Radiate	Branched	Globose	Round	Aspergillus Spp
				green			Septate					
PBP	A	Raised	Entire	Grey	Very	Whitish	Non-	Radiate	Branched	Globose	Spheri	Mucor spp
					Rapid		Septate				cal	
PRW	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG
HLW	A	Unbonate.	Entire	Spong	Very	Pale	Unbrane	Bi-seriate	Simple,	Globose	Spheri	Rhizopus spp.
				y dark	Rapid	white	hed		long and		cal	
				brown	-				branched			
HBP	A	Umbonate	Entire	Dark	Rapid	Yellow	Non-	Radiate	Branched	Globose	Round	Aspergillus Spp
				green			Septate					
	В	Umbonate	Entire	Light	Rapid	Cream	Septate	Bi-seriate	Branched	Globose	Round	Penicillium spp.
				green		colour						
HRW	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG

Table 3: Cultural and microscopic characteristics of fungal isolates

Keys: PLW- PALMWINE LEFT WINGS PBP- PALMWINE BODY PARTS PRW- PALMWINE RIGHT WINGS

HLW- HOUSEHOLD LEFT WINGS HBP- HOUSEHOLD BODY PARTS HRW- HOUSEHOLD RIGHT WINGS NG- NO GROWTH

The cultural and microscopic characteristics of fungal isolates from houseflies' body parts identified a diverse range of fungal species. Fungal isolates from Abattoir Left Wings (ALW) were identified as Mucor spp and Fusarium spp, due to their raised entire elevation, grey to brown-yellow coloration on PDA, and radiate conidia head. Aspergillus spp was suspected in isolates from Abattoir Body Parts (ABP), Abattoir Right Wings (ARW), and Food Vendor Left Wings (FLW), characterized by umbonate elevation, dark green coloration, and non-septate hyphae.

Rhizopus spp was identified in one ABP isolate, distinguished by its unbonate elevation, spongy dark brown coloration, and unbranched hyphae. Penicillium spp was detected in one Food Vendor Body Parts (FBP) isolate, showing umbonate elevation, light green coloration, and bi-seriate conidia. Mucor spp was also found in FBP isolates. No fungal growth was observed in Food Vendor Right Wings (FRW). These findings highlight the potential role of houseflies in transmitting fungal pathogens.

			CULTU RAL							MICROS COPY		
Samp les	Isol ate	Elevation	Margin	Colou (PDA)	Grow th Rate	Reverse Side	Hyphae	Conidia head	Conidiosp ores	<u>Vescicle</u> shape	Conidi a shape	Suspected organism
ALW	Α	Raised	Entire	Grey	Very Rapid	Whitish	Non- Septate	Radiate	Branched	Globose	Spheri cal	Mucor spp
	В	Raised	Entire	Brown - yellow	Rapid	Whitish	Septate	Radiate	Long Branched	Globose	Round	Fusarium spp.
ABP	Α	Umbonate	Entire	Dark green	Rapid	Yellow	Non- Septate	Radiate	Branched	Globose	Round	Aspergillus Spp
	в	Raised	Entire	Red	Rapid	Whitish	Septate	Radiate	Long Branched	Globose	Round	Fusarium spp.
	Α	<u>Unbonate</u>	Entire	Spong y dark brown	Very Rapid	Pale white	Unbranc hed	Bi-seriate	Simple, long and branched	Globose	Spheri cal	Rhizopus spp.
ARW	Α	Umbonate	Entire	Dark green	Rapid	Yellow	Non- Septate	Radiate	Branched	Globose	Round	Aspergillus Spp
FLW	Α	Umbonate	Entire	Dark green	Rapid	Yellow	Non- Septate	Radiate	Branched	Globose	Round	Aspergillus Spp
FBP	Α	Raised	Entire	Grey	Very Rapid	Whitish	Non- Septate	Radiate	Branched	Globose	Spheri cal	Mucor spp
	в	Umbonate	Entire	Dark green	Rapid	Yellow	Non- Septate	Radiate	Branched	Globose	Round	Aspergillus Spp
	С	Umbonate	Entire	Light green	Rapid	Cream colour	Septate	Bi-seriate	Branched	Globose	Round	Penicillium 5pp
FRW	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG	NG

 Table 4: Cultural and microscopic characteristics of fungal isolates

Keys: FLW- FOOD VENDOR LEFT WINGS FBP- FOOD VENDOR BODY PARTS FRW- FOOD VENDOR RIGHT WINGS

ALW- ABATTOIR LEFT WINGS ABP- ABATTOIR BODY PARTS ARW- ABATTOIR RIGHT WINGS

Table 5 revealed the antibacterial study, measuring the inhibitory effects of water and ethanol extracts at three different concentrations (100 mg/ml, 50 mg/ml, and 25 mg/ml) against six bacterial pathogens: *Staphylococcus aureus, Escherichia coli, Klebsiella sp, Proteus sp, Bacillus sp* and *Salmonella sp*. The extracts are compared to CPX (Ciprofloxacin) as a positive control, and DMSO (Dimethyl sulfoxide) as a negative control (which shows no antibacterial effect). Presented in the table is the mean inhibition zones (with standard deviation) for each extract and CPX, indicating how effective they are in preventing bacterial growth.

CPX (Ciprofloxacin) shows the highest inhibition across all pathogens and concentrations, indicating it is an effective broad-spectrum antibiotic. DMSO as a negative control shows no inhibition (0.00 mm) across all treatment. The ethanol extract generally shows better antibacterial activity than the water extract at most concentrations. The highest inhibition zone against Staphylococcus aureus was at 100 mg/ml, the ethanol extract had a zone of 10.67 ± 2.52 mm which is higher than the water extract (8.67 ± 2.08 mm). Both extracts generally show a dose-dependent relationship of which higher concentrations (100 mg/ml) yield greater zones of inhibition than lower concentrations (25 mg/ml).

A p-value below 0.05 suggests a statistically significant difference in inhibition zone between different concentrations. Staphylococcus aureus exhibited good susceptibility to both water and ethanol extracts, with statistically significant inhibition across the concentrations tested (p<0.05) *Escherichia coli* showed weaker inhibition by the extracts and the differences between the concentrations of the ethanol extract are not statistically significant (P>0.05) while against water concentration was statistically significant (P<0.05). *Klebsiella sp* and

Bacillus sp. also demonstrate statistically significant inhibition (P<0.05), particularly at higher concentrations. *Proteus sp.* showed relatively lower susceptibility with no significant relationship (P>0.05) between inhibition zone and concentrations. *Salmonella spp.* showed no zone of inhibition in the various water extract concentration while the ethanolic extract on the other hand exhibit statistical variation (P<0.05) in the various concentration with the least concentration of 25mg/ml showing no zone of inhibition.

The test organisms show different bacteria sensitivity to the extracts. *Staphylococcus aureus* was the most susceptible in the water extract with *Salmonella spp*. showing no susceptibility. In the ethanol extract, *Escherichia coli* was the most susceptible while *Klebsiella sp*. was the least susceptible.

	- Po	Shive conti	of and Diff.	o as negativ	c control)		
Concentration	Water	CPX	DMSO	Ethanol	CPX	DMSO	Pathogen
(mg/ml)	Extract			Extract			
100	$8.67^{\rm c}\pm2.08$	$28.67^{a} \pm$	0.00 ± 0.00	$10.67^{b} \pm$	$27.67^{a} \pm$	0.00 ± 0.00	
		0.58		2.52	0.58		Staphylococcus
50	$4.67^{b} \pm$	-	-	$6.67^{b} \pm 1.53$		0.00 ± 0.00	aureus
	1.53						
25	$0.00^{\mathrm{a}} \pm 0.00$	-	-	$1.33^a \pm 2.31$		0.00 ± 0.00	
100	7.00 ^b ±	$30.67^{a} \pm$	0.00 ± 0.00	11.33 ^b ±	30.33 ^a ±	0.00 ± 0.00	
	1.00	0.58		2.08	0.58		Escherichia
50	$3.00^{a} \pm 2.65$	-	-	6.67 ^{ab} ±	-	-	coli
				2.08			
25	1.00 ^a ±	-	-	$3.00^{a} \pm 3.61$	-	-	
	1.732						
100	$5.67^{b} \pm$	26.33ª ±	0.00 ± 0.00	$9.00^{b} \pm 1.00$	28.33ª ±	0.00 ± 0.00	
	1.53	0.58			0.58		Klebsiella spp
50	$1.33^{a}\pm2.31$		0.00 ± 0.00	$2.00^{\text{b}} \pm 3.46$	-	-	
25	$0.00^{a}\pm0.00$		0.00 ± 0.00	$0.00^{a}\pm0.00$	-	-	
100	$7.67^{b} \pm$	$28.67^{a} \pm$	0.00 ± 0.00	10.67 ^b ±	$28.67^{a} \pm$	0.00 ± 0.00	
	4.16	0.58		4.16	0.58		Proteus spp
50	$5.00^{a}\pm4.36$	-	-	$7.33^a\pm3.51$	-	-	
25	$1.00^{\rm a}\pm1.73$	-	-	$3.67^a\pm3.51$	-	-	
100	$6.00^{b} \pm$	25.33ª ±	0.00 ± 0.00	$8.33^{b} \pm 1.15$	25.67ª ±	0.00 ± 0.00	
	2.00	0.58			0.58		Bacillus spp
50	$1.00^{\rm a}\pm1.73$	-	-	$2.67^a\pm2.52$	-	-	
25	$0.00^{\rm a}\pm0.00$	-	-	$0.67^{\rm a}\pm1.15$	-	-	
100	$0.00^{\rm b}{\pm}0.00$	$30.33^{a} \pm$	0.00 ± 0.00	10.33 ^b ±	$31.33^{a} \pm$	$0.00^{b} \pm$	
		0.58		1.05	0.58	0.00	Salmonella spp
50	$0.00^{a} \pm 0.00$	-	-	$6.\overline{67^{b}} \pm 0.00$	-	-]
25	$0.00^{a} \pm 0.00$	-	-	$0.00^{a} \pm 0.00$	-	-	

Table 5: Antimicrobial Test of the Right-wing Extracts against test Organisms (using Ciprofloxacin as
positive control and DMSO as negative control)

Result represent mean \pm standard deviation. Means followed by the same letter in each column are not significantly different according to Duncan's Multiple Range at P <0.05.

IV. Discussion

Flies are known to have close association with various microbes and parasites at every stage of their growth (Nayduch and Burrus, 2017). Various microorganisms find a peaceful abode in this arthropod and this makes them an insect of concern for entomologist and other researchers since they also have close relationship with human and animals. Despite having close association with deadly microorganism of economic importance, they still survive the co-habitation and this mechanism of survival is a mystery. Few reports have it that the mechanism of flies' survival despite having close relationship with harmful microbes is due to the presence of anti-microbial compounds in their wings and the digestive tract (Asril *et al*, 2021)

This current study evaluates the presence of various microbes in flies' wings and other body parts and the potential of the wing extracts as a natural approach in neutralizing the effect of the harmful microorganisms present their in. Table 1 shows the total bacterial count (TBC), total coliform count (TCC) and total fungi count (TFC) present in flies trapped in various human habitation zone.

Total Bacteria Count ranges from 0.18×10^2 to 2.42×10^2 cfu/mL. The highest TBC found in flies captured in palm wine body parts (PBP) align with previous studies, which reported high bacterial loads on houseflies found in food and beverage environments (Sasaki *et al.* 2000; Graczyk *et al.*, 2001). In addition, the Household Body Parts (HBP) also showed notable bacterial contamination with TBC of 1.64×10^2 cfu/mL. In contrast, no growth was observed for the Palm Wine Right Wing (PRW) and Household Right Wings (HRW). This suggest the potential of the right wings of houseflies in having antimicrobial compounds that neutralizes the

presence of harmful bacteria. This finding is consistent with the work of (Atta, 2014; Asril, 2021) who reported the right wings of flies not exhibiting any pathogenic bacteria after laboratory analysis.

Analysis shows the presence of Coliform which ranged from 0.28×10^2 to 1.47×10^2 cfu/mL with the highest Total coliform Count (TCC) found in PBP (1.47 x 10^2 cfu/mL. This finding agrees with the previous studies which reported high coliform counts on houseflies trapped in environment void of hygiene (Chavasse *et al.* 1999; Khan *et al.* 2012). Significantly, no growth was observed on the Abattoir Right Wings (ARW) and Food Vendor Right Wings (FRW) indicating minimal or no coliform presence on these body parts.

Flies trapped in Abattoir exhibited the highest fungal contamination with ABP having the most count (2.40 x 10^2 cfu/mL). This result is consistent with previous findings, which reported high fungal loads on houseflies in environments with high organic matter and humidity (Oliveira *et al.*, 2013; Singh *et al.*, 2015).

The cultural and biochemical of bacteria examination shows the presence of six bacterial spp which include *Proteus spp, Salmonella spp, Escherichia coli, Klesiella spp, Bacillus spp* and *Staphylococcus aureus*. All these bacteria species were detected in various body parts of the houseflies from various location except the PRW which exhibited no growth. These findings corroborate the report of (Rebab, 2014) who demonstrated the presence of bacteria and fungi in the left wing of houseflies after the dipping of both left and right wings in different sterile water for several minutes.

Klebsiella spp present in the HRW have been reported to produce antimicrobial peptides called bacteriocin which have antibacterial activity against closely related species. The presence of *Bacillus spp* in the ARW can serve as protection against *E. coli* contaminated drinks due to their antibiotic effect. The presence of *Bacillus spp* in the right wings is a plus as it has been reported to carry defensive mechanism called bacteriocins such as mersacidin, subpeptin, JM4-B, subtilosin A and sublancin. (Simons, 2020). The enzymes and secondary metabolites they produce can inhibit the growth of harmful bacteria, fungi spp and other parasites (Zhao *et al*, 2017; Claresta, *et al.*, 2020).

On the other hand, cultural and microbial examination of the various body parts of houseflies for the presence of fungi spp in Table 3 and 4 indicated the presence of various fungi such as *Aspergillus spp*, *Mucor spp*, *Rhizopus spp*, *Penicillium spp* and *Fusarium spp*. All parts examined showed the presence of at least one fungi spp except the PRW, HRW and FRW. *Aspergillus spp* was isolated from the ARW. *Aspergillus spp* are often isolated from house dust, compost heaps and dead vegetation (Kumar *et al*, 2010). The result shows how various body parts of houseflies can serve as reservoir for various harmful fungi except the right wings. Some findings have also reported the right wings of housefly carrying antimicrobial properties while the left wings and other body parts carries various debilitating microorganism.

Table 1, 2, 3 and 4 shows the potential antimicrobial activities of the right wing of housefly. However, the results as shown in Table 5 demonstrate the significance of the right-wing extracts of housefly having antimicrobial activities against various prominent microorganisms. From observation, both the water and ethanolic extract of the right-wing exhibit varying degrees of inhibition against the test organisms. Kinglsey, (2002) reported the potency of the ethanolic extract of the right wings of *M. domestica* to subdue the activity of bacteria and fungi. The minimum inhibitory concentration (MIC) values for both the water and ethanol extracts ranges from 25 to 100 mg/mL. This indicate that the extracts have moderate to high antimicrobial activity. The result from this study also shows that the effect of the extract is dose dependent.

It has been reported in some studies that flies are highly contagious vector having various debilitating microbes on their body and for them to survive, they must carry some antimicrobial properties on their body (Kingsley, 2002). Secondary metabolites are reported to be the source of antimicrobial agents on their body which is mostly peculiar to their body surface, right wing and part of their gut (Ali *et al.*, 2018).

Various microorganisms such as *E. coli, Salmonella spp, S. aureus, Klebsiella spp* and *Proteus spp* have been reported to exhibit moderate to high zone of inhibition (ZOI) when subjected to insect secondary metabolites. Among all the test agent, Ciprofloxacin shows the most promising antimicrobial effect than the ethanolic and water extract of the right-wings. Statistically, *E. coli* and *Salmonella spp* shows the highest level of sensitivity to Cipro. In contrast, no zone of inhibition was observed when the microorganisms were subjected to Dimethyl Sulphoxide (DMSO). This indicates that DMSO may not be the right agent to subdue the effect of the microbes highlighted.

V. Conclusion

It is apparent from this study that both the ethanolic and water extracts of the right wing of *M. domestica* contain antimicrobial agents. Although CPX shows better effect on the test organisms but antimicrobial resistance against the test organism in the long run may set in and this could make the test organism insensitive to this antibiotic. Therefore, both water and ethanolic extracts can serve as alternative and can serve as a natural and novel means of controlling microbial agents in food and water through scientific enhancement.

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