



A Study on Skin Microbiome, Knowledge and Control of Malaria among Volunteers in Akure– A Malaria Endemic Area, South-West, Nigeria

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

The study was carried out to determine the skin microbial diversity, knowledge and control of malaria among volunteers in Akure. Skin surface of one hundred and fourteen (114) consented participants were swabbed at different locations (elbow, neck, and knee) on the body and examined for the microbial diversities using standard microbiological methods. Socio-demographic characteristics, knowledge and awareness of malaria and mosquito control among the participants were evaluated with the use of multiple choice questionnaires. The highest number of the participant were within the age range 31-40 years 32(28.1%) followed by age range 11-20 and 21-30 years (17.5%). The number of male and female participants were 67(58.8%) and 47(41.2%) respectively. It was noted that all the participants have heard about malaria before while 60(52.6%) of them have been diagnosed of malaria in the last 6 months, 28.9% of the participants have their personal mosquito repellent as at the time of this investigation while 71.1% did not have mosquito repellent. Bacterial counts (Log_{10} CFU/cm²) ranged from 5.70±0.13 (neck) to 6.51±0.05 (knee) while the highest fungal counts (Log_{10} SFU/cm²) was observed in knee (2.80±0.07) followed by elbow (2.61±0.04) and neck (1.91±0.05). Ten (10) bacterial and nine (9) different fungal species were identified as skin microbiome, the most occurred bacterial genus in the skin of the neck, elbow and knee were *Staphylococcus* (49.36%), *Bacillus* (42.23%) and *Staphylococcus* (43.75%) respectively

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while the most occurred fungal genus were *Aspergillus* (36.54%), *Aspergillus* (50%) and *Alternaria* (18.92%) in the neck, elbow and knee skin respectively. The results of the study will be important for control of malaria in endemic area and the skin microflora isolated could be explored as mosquito repellent to reduce malaria infection in endemic areas.

Keywords: Skin; mosquito repellent; microbiome; knowledge and awareness of malaria.

1. INTRODUCTION

Malaria is transmitted throughout Nigeria, with 76 percent of the population living in high transmission areas and 24 percent in low transmission areas [1]. The main mode of malaria transmission worldwide is through the bite of an infected female *Anopheles* mosquito as vector. Sequel to this many control measure such as the use of insecticide treated nets (ITNs), long lasting insecticide nets (LLINs), repellants, laticidal and indoor residual spray (IRS) taken to control and eradicate malaria have been channeled to malaria vector [2,3,4].

Humans differ in their attractiveness to mosquitoes and these differences remain relatively stable over time [5,6].

Because the human skin microbiota plays an important role in the production of body odours and is attractive to *A. gambiae* when grown on agar plates, a correlation between skin microbiota composition and a person's attractiveness to mosquitoes can be expected. Within humans, this may lead to selection for skin bacteria that are less attractive or even repellent to mosquitoes [6].

However, human attractiveness to mosquitoes may change when individuals are infected with *Plasmodium* parasites that cause malaria, for instance, children harbouring *Plasmodium* gametocytes attracted about twice as many mosquitoes as children without infection or with parasites in the asexual stage [7]. This increased attractiveness of infected children could be explained by an increase in body temperature, increased perspiration or by a change in breath composition and not as a result of microbial diversity on the skin.

The human skin microorganisms are most abundant in the vicinity of skin glands, where they metabolize the skin gland secretions [7].

Human sweat is odourless, only gets its characteristic smells after incubation with

bacteria [7]. The microbiota of the skin plays an important role in human odour production and the number of certain microorganisms is strongly correlated with the intensity of the odour emitted [8]. *Corynebacteria* also play a pivotal role in the generation of volatile fatty acids, which are associated with malodour [8]. When skin lipids are catabolized into long-chain fatty acids, it seems that only *corynebacteria* are capable of transforming these long-chain fatty acids into short- and medium-chain fatty acids (C2–C11), causing malodour [8]. The presence and diversity of human skin micro-flora may be useful as mosquito's attractant or repellent.

2. MATERIALS AND METHODS

2.1 Collection of Samples

This study was carried out at the Department of Microbiology, Federal University of Technology, Akure. During this study, one hundred and fourteen (114) participants who consented to the study purpose after it has been well explained to them as stated in the consent form were recruited. The identities of all the participants were not revealed to maintain the anonymity. The participants were given orientation about the study, all of which are not under any medication during the period of the study and have not had their bath 24hrs before swab sample collection.

The surface of the skin (inner-elbow, neck, knee fold) was gently swabbed with 70% ethanol for surface sterilization and allowed to dry, clean sterile moistened swabbed stick was used to swab about 2 cm by 2 cm square area of the pre-surface sterilized skin and gently caped with addition of 1.0 mL of sterile normal saline, the samples were collected in triplicates and transported to the laboratory in an ice pack within 1 hour of collection for further microbiological assay.

During sample collection, participants demographic information were collected with the aid of multiple choice structured questionnaire as well as their history of infection, knowledge and awareness of mosquitoes repellent.

2.1.1 Isolation and identification of skin microflora

The swab stick was gently Shaken for 2 to 3 minutes to enables the attached microorganisms released into the normal saline added, the saline was then introduced into 9.0 mL of sterile normal saline and rocked on the palm to make dilution factor of 10^{-1} , the step was repeated until dilution of 10^{-5} was attained. Different dilutions were pure plated on selective and differential media (Manitol salt, *Salmonella-Shigella*, potato dextrose, nutrient and Eosine methylene blue agar). The plates were incubated at 37 °C for 24 hours and 25 °C for 72 hours for enumeration of bacteria and fungi respectively. Biochemical characteristics of bacterial isolates were determined [9] while cultural, morphology and microscopy characteristics of isolated fungi were observed [10].

Data obtained were expressed as mean ± Standard Error of Mean, descriptive statistics and were statistically analysed using One-way ANOVA. The new Duncan Multiple Range test was used to compare means of different groups, bivariate logistic regression was used to compere different parameters. A *P*-value of < 0.05 was considered statistically significant.

3. RESULTS

3.1 Socio-Demographic Characteristics of the Participants

The socio-demographic characteristics of all the participants whose skin flora were examined during this are shown in Table 1. The highest number of the participant were within the age range 31-40 years 32(28.1%) followed by age range 11-20 and 21-30 years (17.5%). The number of male and female participants were 67(58.8%) and 47(41.2%) respectively.

Based on marital status and level of education, there was higher number of participants that were single 73(64%) and those that have tertiary 49(43%) level of education respectively. Also, larger number of participants were trader 41(36%) and student 37(32.5%) based on occupation, Christian 66(57.9%) and Yoruba 108(94.7%) tribe.

3.2 Participants' Knowledge and History of Malaria Infection

The result in Table 2 revealed the participants' knowledge and history of malaria infection during

this study. It was noted that all the participants have heard of malaria before, 60(52.6%) have been diagnosed of malaria in the last 6 months while 76(66.7%) have their relatives diagnosed of malaria in the last 6 months and the number of volunteers that agreed that mosquito alone can transmit malaria 106(93%) were significantly ($p < 0.001$) high.

3.3 Participants' Knowledge about Exposure to and Control of Mosquito Bites

The result showed in Table 3 revealed the participants' knowledge about exposure to and control of mosquito bites. In the knowledge of exposure to mosquito bites, it was noted that higher 110(96.5%) numbers of participants do experience mosquito bites whenever they are in a place where mosquitoes are, while 3.5% of them do not. The result of this study also showed that 28.9% of the participants do not experience mosquito bites whenever they are among those that have the mosquito bites.

Furthermore, in the knowledge about control of mosquito bites, 60.5% of the participants have heard of mosquito repellent while 39.5% have not, 28.9% of the participants have their personal mosquito repellent as at the time of this investigation while 71.1% did not have mosquito repellent. Among the participants (33 participants) that owns mosquito repellent, 25(75.76%) used spray, 8(24.24) are using cream while none of them used mosquito coil and plant based mosquito repellent. Among the participants that did not have mosquito repellent, 3.5% claimed that they did not have it due to non-availability, 28.1% claimed it was expensive while 39.5% claimed they were not aware of mosquito repellent. Statistically, all the factors examined were significant ($p \leq 0.05$).

3.4 Comparison of Microbial Load Associated with Different Parts of the Participants' Skin

Comparison of microbial load associated with different parts of the participants' skin (Table 4). It was noted that there was no significant ($p \leq 0.05$) difference in the mean bacterial counts of neck and inner part of elbow, bacterial counts (Log_{10} CFU/cm²) ranged from 5.70±0.13 (neck) to 6.51±0.05 (knee fold).

The result also showed that there was no significant ($p \leq 0.05$) difference between the fungal counts of inner part of elbow and knee fold, the

highest fungal counts (Log_{10} SFU/cm²) was observed in knee fold (2.80 ± 0.07) followed by inner-elbow (2.61 ± 0.04) and neck (1.91 ± 0.05). Generally, the lowest and highest microbial load were observed in neck and knee fold respectively.

Table 1. Socio-demographic characteristics of the participants

Socio-Demographic characteristics	Frequency	Percentage (%)
Age (Year) (Total = 114)		
1-10	18	15.8
11-20	20	17.5
21-30	20	17.5
31-40	32	28.1
41-50	16	14.0
51-60	8	7.0
Gender (Total = 114)		
Male	67	58.8
Female	47	41.2
Marital status (Total = 114)		
Single	73	64.0
Married	33	28.9
Widowed	8	7.0
Divorced	0	0
Level of Education (Total = 114)		
Informal	12	10.5
Primary	29	25.4
Secondary	24	21.1
Tertiary	49	43.0
Occupation (Total = 114)		
Pupils	21	18.4
Student	37	32.5
Trader	41	36.0
Farmer	4	3.5
Civil servant	7	6.1
Unemployed	4	3.5
Religion (Total = 114)		
Christian	66	57.9
Muslim	48	42.1
Others	0	0
Tribe (Total = 114)		
Igbo	6	5.3
Yoruba	108	94.7
Hausa	0	0
Others	0	0

Table 2. Participants' Knowledge and History of Malaria Infection

Knowledge and history of malaria (Total number of respondents = 114)	Frequency (%)	p-Value (Significant at <0.05)
Have you heard of malaria before?		
Yes	114(100)	0.632
No	0(0)	
Were you diagnosed of malaria in last 6 months?		
Yes	60(52.6)	0.574
No	54(47.4)	
Were any of your relatives diagnosed of malaria in last 6 months?		
Yes	76(66.7)	<0.001
No	38(33.3)	
Do you agree that malaria can be transmitted by mosquito alone?		
Yes	106(93.0)	<0.001
No	8(7.0)	

Table 3. Participants' knowledge about exposure to and control of mosquito bites

Knowledge about exposure to mosquito bites (Total number of respondents = 114)	Frequency (%)	p-Value (Significant at <0.05)
Do you notice mosquito bite wherever you are in a place there are mosquitoes?		<0.001
Yes	110(96.5)	
No	4(3.5)	
Whenever people around you have mosquito bites, do you experience mosquito bite too?		<0.001
Yes	81(71.1)	
No	33(28.9)	
Knowledge about control of mosquito bites (Total number of respondents = 114)		0.025
Have you heard of mosquito repellent?		
Yes	69(60.5)	
No	45(39.5)	
Do you have mosquito repellent?		<0.001
Yes	33(28.9)	
No	81(71.1)	
If yes, Which mosquito repellent do you have? (Total number of respondents = 33)		0.003
Plant based	0	
Spray	25(75.76)	
Cream	8(24.24)	
Coil	0	
If no, why didn't you have mosquito repellent? (Total number of respondents = 81)		<0.001
Not available	4(3.5)	
Is expensive	32(28.1)	
Not aware of it	45(39.5)	

Table 4. Comparison of microbial load associated with different parts of the participants' skin

Parts of the skin	Bacteria counts (Log₁₀ CFU/cm²)	Fungal counts (Log₁₀ SFU/cm²)
Neck	5.70±0.13 ^a	1.91±0.05 ^a
Inner-Elbow	5.79±0.04 ^a	2.61±0.04 ^b
Knee fold	6.51±0.05 ^b	2.80±0.07 ^b

Values are means of microbial counts±SD, values in the same column carrying same superscript are not different significantly according to new Duncan's Multiple Range test at p≤0.05; Keys: Log₁₀ = Logarithm of numbers to base ten (10), CFU = colony forming unit, SFU = spore forming unit

3.5 Relationship between Bacterial and Fungal Counts of different Parts of Volunteers Skin

Relationship between bacterial and fungal counts of neck among the volunteers was presented in Fig. 1. It was observed that there was positive correlations (r = 0.095) that is, the higher the bacterial counts the higher the fungal counts, however, statistically there was no significant (p≤0.05) difference (p-Value = 0.313) between bacterial and fungal counts of the neck.

Relationship between bacterial and fungal counts of inner-elbow among the volunteers was presented in Fig. 2. It was noted that there was positive correlations (r = 0.156) between the fungal and bacterial counts of the inner-elbow but

there was no significant (p≤0.05) different between the counts (p = 0.098).

Relationship between bacterial and fungal counts of knee fold among the volunteers was presented in Fig. 3. It was observed that there was positive correlation (r = 0.156) between the bacterial and fungal counts but there was no significant (p≤0.05) difference in the counts (p= 0.096).

3.6 Biochemical Characteristics and Presumptive Identification of Bacteria Isolated from the Skin of Participants

The result showed in Table 5 revealed the identity of bacteria that were isolated from skin of participants. It was noted that ten (10) different bacterial species were identified as;

Staphylococcus aureus 61(23.19%), *Staphylococcus epidermidis* 52(19.77%), *Bacillus cereus* 49(18.63%), *Bacillus subtilis* 39(14.83%), *Streptococcus pyogenes* 19(7.22%), *Aeromonas hydrophila* 15(5.70%), *Corynebacterium xerosis* 10(3.80%), *Enterobacter aerogenes* 6(2.28%), *Pseudomonas aeruginosa* 7(2.66%) and *Klebsiella pneumoniae* 5(1.90%),

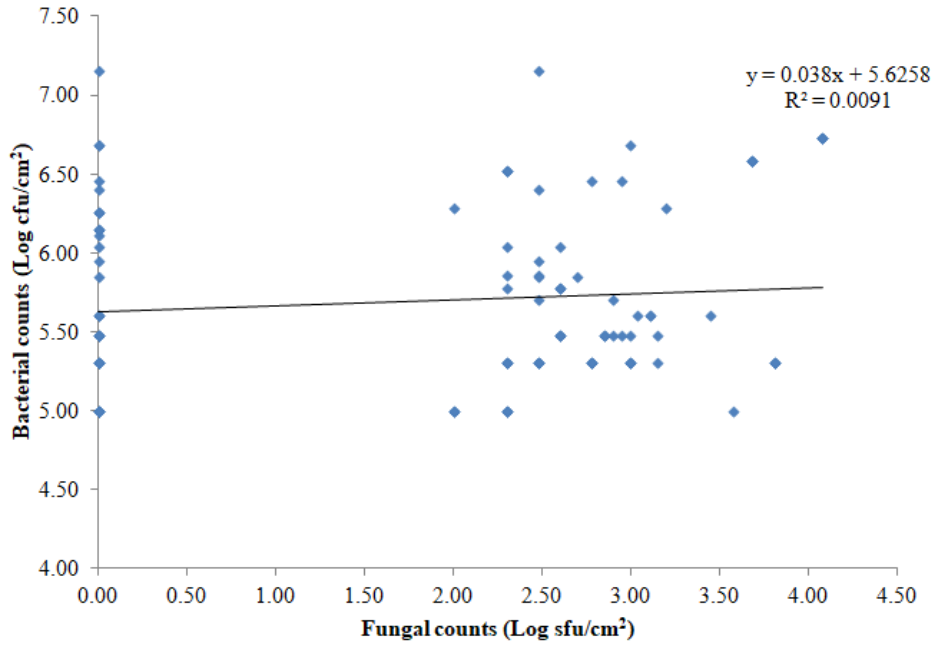


Fig. 1. Relationship between Bacterial and Fungal Counts of Neck among the Volunteers
 $p - Value = 0.313$

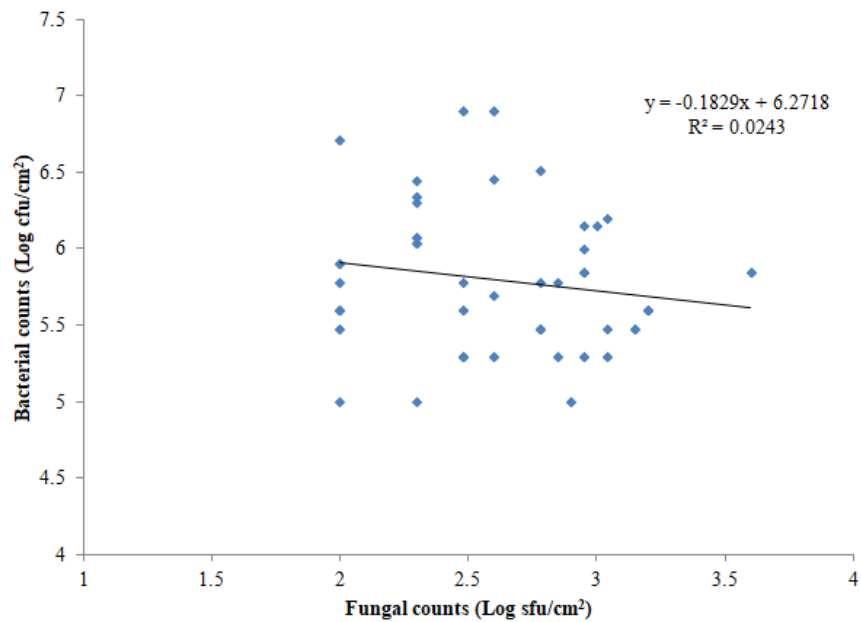


Fig. 2. Relationship between Bacterial and Fungal Counts of Elbow among the Volunteers
 $p - Value = 0.098$

Table 5. Biochemical characteristics and presumptive identification of bacteria isolated from the skin of participants

Gram stain	Cell shape	Urea se	VP	M R	Oxida se	Citrat e	Motility	Indo	Catalas e	slop e	TSI But t	H ₂ S	G as	Lactos e	Sucros e	Sugar fermentation				Coagulas e	SS	Presumptiv e identificatio n (Total = 263)
																Glucos e	Manto l	Maltos e	Galactos e			
-	Rd	-	+	-	-	-	+	+	+	Y	Y	-	+	+	+	+	+	+	+	N	-	<i>Aeromonas hydrophila</i> (n=15)
-	Rd	+	+	-	-	+	-	-	+	Y	Y	-	+	+	-	+	-	+	-	N	-	<i>Klebsiella pneumoniae</i> (n=5)
-	Rd	-	+	-	-	+	+	-	+	R	Y	-	+	+	+	+	+	+	+	N	-	<i>Enterobacter aerogenes</i> (n=6)
+	Rd	-	-	+	-	+	-	-	+	R	Y	+	-	-	+	+	-	+	+	N	-	<i>Corynbacterium xerosis</i> (n=10)
+	Rod	-	+	-	-	+	-	-	+	N	N	-	N	-	+	+	N	N	N	N	+	<i>Bacillus subtilis</i> (n=39)
+	Rod	-	+	-	-	+	+	-	+	N	N	-	N	-	+	+	N	N	N	N	+	<i>Bacillus cereus</i> (n=49)
-	Rd	-	-	-	+	+	+	-	+	R	R	-	-	-	-	-	+	-	-	N	-	<i>Pseudomonas aeruginosa</i> (n=7)
+	Cc	+	+	+	-	+	-	-	+	Y	Y	-	-	+	+	+	+	+	+	+	-	<i>Staphylococcus aureus</i> (n=61)
+	Cc	+	+	+	-	+	-	-	+	Y	Y	-	-	+	+	+	-	+	+	-	-	<i>Staphylococcus epidermidis</i> (n=52)
+	SC	+	+	+	-	+	-	-	+	Y	Y	-	-	+	+	+	+	+	+	+	-	<i>Streptococcus pyogenes</i> (n=19)

Keys: -:negative to the test, +: positive to the test, N- Not determined, Cc- cocci shape, SC – streptococci shape, Y- yellow, R- red, Butt- bottom, n- number of isolate, VP- Voges-Proskauer test, MR- Methyl Red test, TSI- triple sugar iron, H₂S- hydrogen sulphide gas production, SS- spore staining, SH- starch hydrolysis, n - number

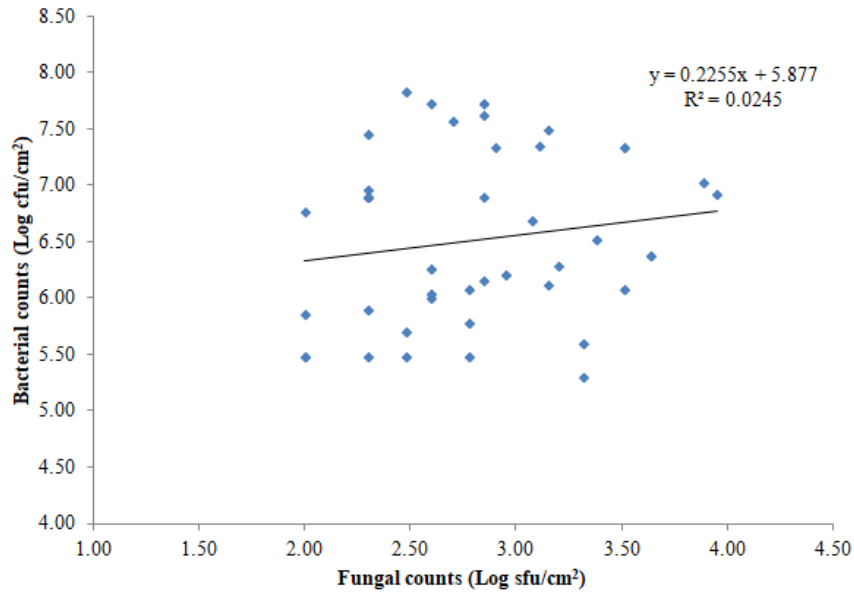


Fig. 3. Relationship between Bacterial and Fungal Counts of Knee Among the Volunteers
 $p - \text{Value} = 0.096; r = 0.156$

3.7 Relative Abundance of Bacterial Genus Isolated from the Skin of Participants

Relative abundance of bacterial genus isolated from the skin of participants is shown in Fig. 4. The relative abundance of the bacterial genus isolated from the neck were; *Staphylococcus* (49.36%), *Bacillus* (29.94%), *Streptococcus* (10.19%), *Pseudomonas* (4.45%), *Corynbacterium* (4.14%), *Aeromonas* (1.27%) and *Enterobacter* (0.64%). Also, the relative abundance of bacterial genus isolated from inner-elbow were; *Bacillus* (42.23%), *Staphylococcus* (36.51%), *Streptococcus* (6.53%), *Aeromonas* (6.27%), *Klebsiella* (3.54%), *Enterobacter* (2.18%), *Pseudomonas* (1.63%) and *Corynbacterium* (1.09%) while *Staphylococcus* (43.75%), *Bacillus* (27.72%), *Aeromonas* (8.97%), *Corynbacterium* (6.52%), *Streptococcus* (4.89%), *Enterobacter* (4.08%), *Pseudomonas* (2.45%) and *Klebsiella* (1.63%) were the genus of bacteria present in knee fold.

3.8 Morphological Identification of Fungi Isolated from the Skin of Participants

The result showed in Table 6 revealed the identity of fungi that were isolated from skin of participants. It was noted that nine (9) different fungal species were identified as; *Aspergillus niger* 31(21.83%), *Alternaria alternate*

30(21.13%), *Aspergillus flavous* 28(19.72%), *Exophiala dermatididis* 15(10.56%), *Malassezia furfur* 10(7.04%), *Candida tropicalis* 9(6.34%), *Candida albicans* 8(5.63%), *Fusarium oxysporum* 7(4.93%) and *Rhizopus stolonifer* 4(2.82%).

3.9 Relative Abundance of Fungal Genus Isolated from the Skin of Participants

Relative abundance of bacterial genus isolated from the skin of participants is shown in Fig. 5.

The relative abundance of fungal genus isolated from the neck were *Aspergillus* (36.54%), *Candida* (25%), *Malassezia* (25%), *Fusarium* (7.69%) and *Alternaria* (5.77%). The relative abundance of fungal genus isolated from inner-elbow were *Aspergillus* (50%), *Alternaria* (23.96%), *Exophiala* (12.5%), *Candida* (11.46%) and *Fusarium* (2.08%) while those that were isolated from knee fold were *Alternaria* (18.92%), *Rhizopus* (18.92%), *Exophiala* (17.57%), *Aspergillus* (16.22%), *Fusarium* (14.86%), *Malassezia* (9.46%) and *Candida* (4.05%).

4. DISCUSSION

The socio-demographic of the participants showed that the highest number were within the age range 31-40 years, the number of male were higher than female, based on marital status and

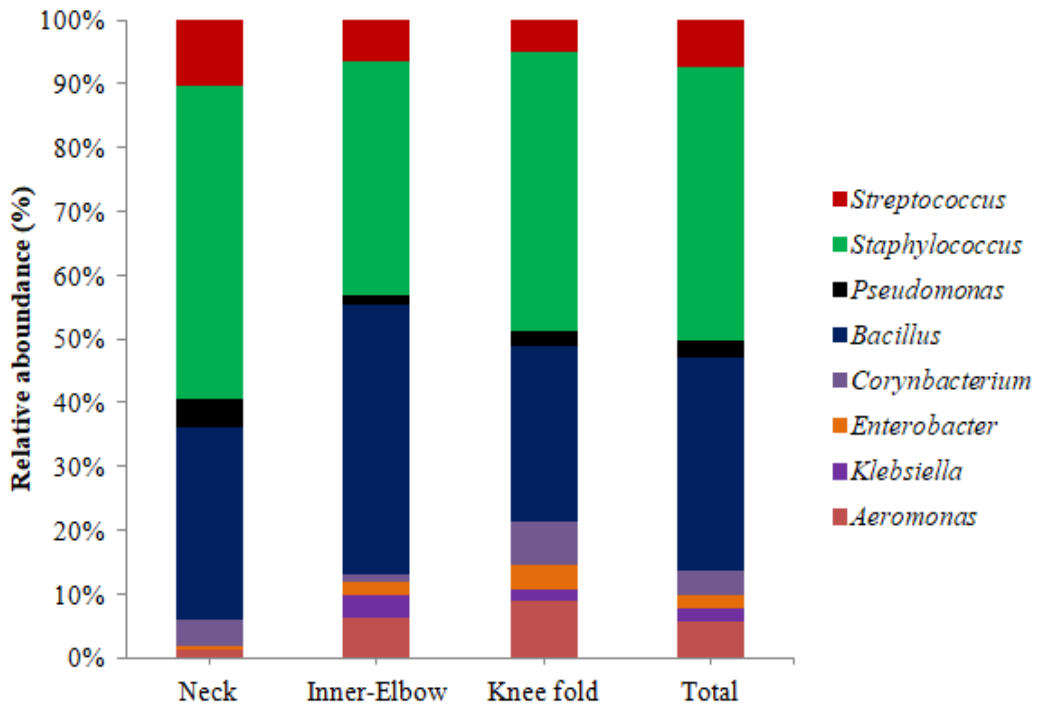


Fig. 4. Relative Abundance of Bacterial Genus Isolated from the Skin of Participants

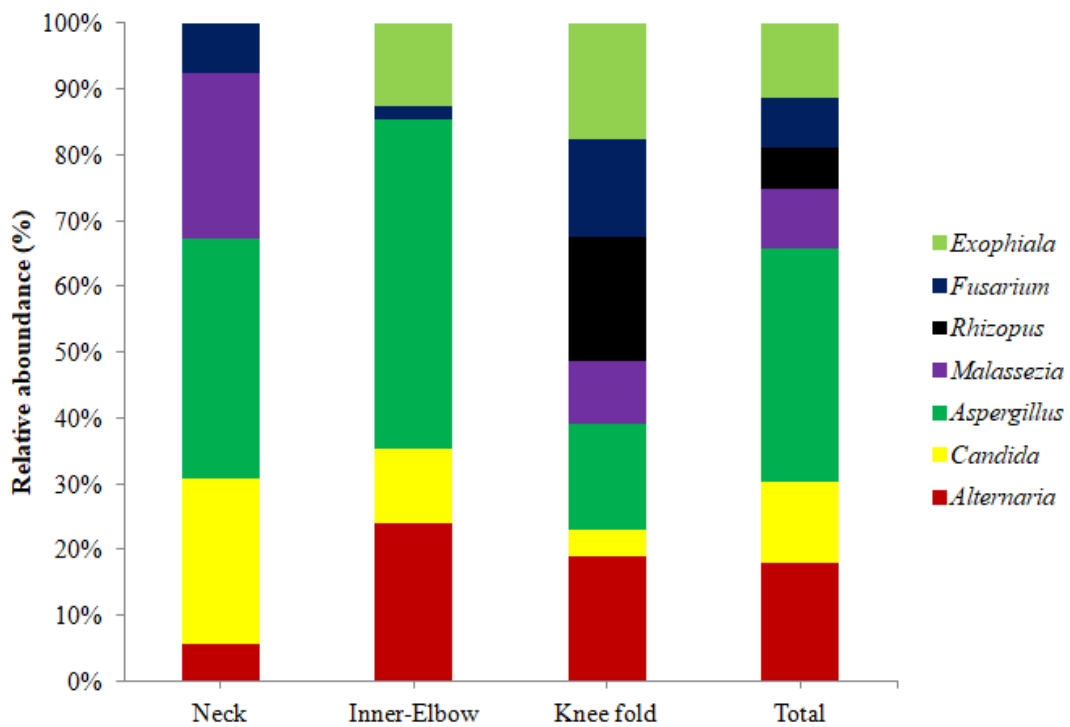


Fig. 5. Relative abundance of fungi genus Isolated from the skin of participants

Table 6. Morphological identification of fungi Isolated from the skin of participants

Microscopic and cultural characteristics	Probable organism
Brown to black colonies on potato dextrose agar (PDA), have short conical conidia and consisting of large catenate conidia chains	<i>Alternaria alternate</i> (n= 30)
Colonies are cream, raised, entire, smooth and butyrous on PDA, elongated budding cells are present with pseudohyphae.	<i>Candida albicans</i> (n= 8)
Whitish, with round oval shape, presence of blastoconidia which reproduce by budding	<i>Candida tropicalis</i> (n= 9)
Bright yellow to green colonies on potato dextrose agar. Has the same colour of green with a tinge of yellow on the reverse side of the plate. Conidiophores coarsely rough, heads vary in size, loosely radiate, phialides borne directly on the vesicle and conidia are globose	<i>Aspergillus flavous</i> (n= 28)
It appeared as whitish colonies which turned black due to the formation of black conidia on potato dextrose agar and spread rapidly. Under microscopic investigation, the hyphae were septate and profusely branched. Conidia were bonded in chains at the hips of sterigmata. The conidia are seriated and compact head forming chains	<i>Aspergillus niger</i> (n= 31)
Cream to yellowish brown yeast like colonies on PDA, slow growth but rapidly at 37 °C	<i>Malassezia furfur</i> (n= 10)
Rapidly growing, whitish in colour on PDA, not or scarcely septate; rhizoids and stolons present; brown sporangiophores, solitary or in tufts on the stolons, diverging from the point at which the rhizoids form; round sporangia and ovoid sporangiophores.	<i>Rhizopus stolonifer</i> (n= 4)
White thick mycelium and white colour at bottom of plate on potato dextrose agar. Microconidia are oval and produced on simple short phialides, septate and chlamydo spores are present	<i>Fusarium oxysporum</i> (n= 7)
Brownish-black colonies with grey borders, slightly elevated with black pigment on the reverse side of the plate on potato dextrose agar. The colonies were irregular and varied in size and spread largely on the plates, a pigmented hypha developed appearing with filaments, Conidia are globular and epithelial in shape	<i>Exophiala dermatididis</i> (n= 15)

Key: n = number of isolates

level of education, majority were single and have tertiary level of education respectively. Also, larger numbers of participants were trader and student based on occupation, Christian and Yoruba dominated the religion and tribe. It has been established that socio-demographic characteristics of participants used in a study could influence the outcome of the study [11,12], therefore, the variations in the socio-demographic of the participants could have influence in the outcome of the study. However, the use of statistical tool to analyze the results obtained in this study may have eliminated the biased influence of the socio-demographic of the participants.

The participants' knowledge and history of malaria infection during this study was evaluated, it was noted that all the participants have heard of malaria before, higher numbers of participants have been diagnosed of malaria as well as their relatives in the last 6 months. The fact that all participants have heard of malaria before could be attributed to the continual awareness of malaria through different media and the occurrence and high transmission of malaria in

the study area, Ondo State, Nigeria [13,14,15]. Higher number of participants together with their relatives with case of malaria in last 6 months as revealed in this study is not surprising, because studies have shown that people living in malaria endemic area usually suffer at least one or two episodes of malaria in a year [16,17,4,18,15]. Also, the number of participants that agreed that mosquito alone can transmit malaria were significantly ($p < 0.001$) high. Knowledge of mosquito as a vector of malaria could prevent or predisposes people to malaria infection [19, 20, 21,17]. Since the number of the participants that agreed to the fact that mosquito alone can transmit malaria is not 100%, it is suggested that there should be continual awareness through different media about mosquito as vector of malaria. This finding will assist in decision making towards improved malaria prevention and control.

Furthermore, higher percentage of the participants in this study do experience mosquito bites whenever they are in a place where mosquitoes are while 28.9% stated that they do not experience mosquito bites whenever they are

in the midst of those that are experiencing mosquito bites, this confirms that humans differ in their attractiveness to mosquitoes [5]. Knowledge about control of mosquito bites was evaluated among the participants and it was noted that, 60.5% of the participants have heard of mosquito repellent and few participants have their personal mosquito repellent as at the time of this investigation. The understanding of the preventive and control measure of malaria varies from community to community and among individual, these and some other factors may influence peoples attitude towards the use of mosquito repellent, this also corroborate the study of Omoya and Ajayi [22]. Among the participants that own mosquito repellent, some used spray, and few used cream while none of them used mosquito coil and plant based mosquito repellent. The use of spray and cream by the respondents to repel mosquitoes could be due to availability and continual advertisement of the products on the media, the use of mosquito coil which produce smoke may have been avoided because of severe health consequences which has been reported by Romi *et al.* [23], Stevenson [24], and Kiplang'at [2]. Among the participants that did not have mosquito repellent, this study revealed that non-availability, expensive price tag and non-awareness of mosquito repellent were statistically significant ($p \leq 0.05$) factors for not being in possession of mosquito repellent. Therefore, there is need to intensify on the awareness of mosquito repellent and production of safe and affordable ones in the market.

Microbiota on the human skin plays a major role in interactions between disease vectors, their population and diversity displays a qualitative and quantitative correlation [25]. This study compare of microbial load associated with different parts of the participants' skin was examined in this study, it was noted that there was no significant ($p \leq 0.05$) difference in the mean bacterial counts of neck and hand, while the leg has the higher bacterial counts. Also, there was no significant ($p \leq 0.05$) difference between the fungal counts of hand and leg, while the neck has the least. The microbiota of the skin plays an important role in human odour production and the number of certain microorganisms is strongly correlated with the intensity of the odour emitted [25]. Therefore, the higher bacterial load observed in the leg may influence the mosquito bite among the participants.

Furthermore, relationship between bacterial and fungal counts of neck, hand and leg among the volunteers was studied and showed that skin fungal load have site-specific correlational patterns with co-colonizing bacteria, this corroborates the findings of Leung *et al.* [26] who stated that skin fungal community had site-specific correlation with bacterial community. The specific correlations between the fungal and bacterial microflora on the skin may influence differential attractiveness of humans to mosquitoes.

It was noted that ten (10) different bacterial species were identified from the skin and in decreasing order of occurrence they are; *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Bacillus cereus*, *Bacillus subtilis*, *Streptococcus pyogenes*, *Aeromonas hydrophila*, *Corynebacterium xerosis*, *Enterobacter aerogenes*, *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*. Also, nine (9) different fungal species were identified from the skin and in decreasing order of occurrence they are; *Aspergillus niger*, *Alternaria alternate*, *Aspergillus flavus*, *Exophiala dermatididis*, *Malassezia furfur*, *Candida tropicalis*, *Candida albicans*, *Fusarium oxysporum* and *Rhizopus stolonifer*. Occurrence of these bacteria and fungi as a skin microbial flora have been reported worldwide [27,28,29,30,3].

Composition and abundance of the human skin microbiota influences the relative degree of attractiveness of a human to the malaria mosquito *An. gambiae* [3]. Therefore, relative abundance of bacterial and fungal genus isolated from different parts of participants' skin was studied. In bacteria; *Staphylococcus* spp., *Bacillus* spp., and *Streptococcus* spp. were more abundant in the neck; *Bacillus* spp. and *Staphylococcus* spp., were more abundant in the hand while *Staphylococcus* spp., *Bacillus* spp. and *Aeromonas* spp. were more abundant in leg region. Abundance of *Staphylococcus* spp. in the skin have been previously reported to have positive correlation with attractiveness of the individuals to *An. gambiae* [31,27]. The abundance of *Staphylococcus* spp. in the neck, hand and leg may influence the attractiveness of mosquito however, mosquito may be more attracted to the leg region among the participants because of higher bacterial compositions. Also, the most abundant of fungal genus in the neck were *Aspergillus* spp., *Candida* spp., and *Malassezia* spp. In hand region, *Aspergillus* spp., *Alternaria* spp., *Exophiala* spp. and *Candida* spp.

were more abundant while *Alternaria* spp., *Rhizopus* spp., *Exophiala* spp., *Aspergillus* spp. and *Fusarium* spp. were more abundant in leg. Skin mycobiome population (fungal community membership) found on its occupants may influence the bacterial community composition variations and variation in bacterial species on the human skin may explain the variation in mosquito attraction between humans [32,26]. In this study, there was higher abundance of fungi in hand and leg and this may have influence the bacterial diversity.

5. CONCLUSION

The uses of environment friendly and biodegradable natural repellent of microbial origin have received renewed attention as agents for mosquitos' control. The cohort of the volunteers used in this study is a representative model of the people living in Akure and environ while subjects in this study may not represent other Akure and Nigerian individuals, this study is nonetheless fundamental in beginning to understand the skin microbiota of Nigerians for studies on mosquito repellent or attractant.

CONSENT

As per international standard or university standard, Participants' written consent has been collected and preserved by the authors.

ETHICAL APPROVAL

As per international standard or university standard written ethical approval has been collected and preserved by the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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