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The first report of pathogenic bacteria isolated from blow flies *Lucilia sericata* (Diptera: Calliphoridae) in Sari city, North of Iran

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Abstract
Blow Flies are found worldwide, occurring nearly every place inhabited by human. They may transmit disease organisms to people. This study was done to isolate some of bacteria from *Lucilia sericata* in Sari City and determine their antibiotic susceptibility pattern. In this experimental study, totally 600 blow flies were collected from 3sites of Sari city (April–Dec 2016). Bacteria on the external surface and digestive system of each fly were identified by using standard tests based on Bergay' s technique. Antibiotic susceptibility test was performed by disc diffusion method based on CLSI. There were 2 peaks of fly activity in a year; (Jun-July and Aug-Sep). The most common isolated bacteria were *E. coli* (19%) and *S. aureus* (15.8 %). Antibiotic resistance was found in isolated. These findings suggest a potential role of blowflies in transmission of pathogenic bacteria with antibiotic resistance. Therefore, their population should be controlled in North of Iran.

Keywords: *Lucilia sericata*, medically important, antibiotic resistant bacteria, Iran

1. Introduction
Dipteran flies are one of the most abundant and important groups of insects, serving as vectors for some of the diseases affecting humans [1]. Blow flies include a number of species including common green bottle fly (*Lucilia sericata*), blue bottle fly and others. *Lucilia sericata* is a member of the family *Calliphoridae* and plays an important role in medicine [2, 3]. Blow flies are the most common type of flies found in and around buildings. It prefers warm and moist climates especially common in coastal regions, but it also is present in arid areas [4, 5]. The relative frequency and activity of flies has a straight relation with optimum temperature and humidity and rainfall. Very hot and very cold weather is not good for their activity [6]. There is a significant difference in the number of trapped flies among the seasons and months of the year. These flies are common in populated areas and are often found near meat-processing plants, garbage dumps, butcher shops and slaughterhouses [5, 6]. It is 10–14 mm long, slightly larger than a housefly, and has brilliant, metallic, blue-green or golden coloration with black markings [7]. They have robust bodies and wide heads. They are able to transport bacteria from septic environments to other substrates via contamination of their surfaces (feet, wings, bodies) and also by regurgitation of crop contents [8]. Since blow flies routinely move between infected materials such as dead animals or dung and human habitats, they may transmit diseases organisms to human [6, 9]. Flies have been implicated in the transmission of serious diseases such as anthrax, ophthalmia, typhoid fever, tuberculosis, cholera, and infantile diarrhea and have been demonstrated to harbor or transmit other pathogenic bacteria including *Salmonella spp., Proteus spp., Shigella spp., Chlamydia spp., Campylobacter jejuni, Klebsiella spp., Escherichia coli O157:H7, Yersinia pseudotuberculosis, and Helicobacter pylori*, the causative agent of gastric ulcer [10- 13]. Recently, these insects were reported to be involved in disease outbreaks including E. coli O157:H7 in Japan and Vibrio cholera in India [14, 15, 16]. Green bottle flies are classified as filth feeders that develop in and feed on dead animals, feces, garbage and decomposing plant materials [6, 10]. Because of their unsanitary habitats, they may carry pathogenic bacteria that can be transmitted to people and animals via mechanical transmission [17, 18]. The studies carried out in developed countries observed that there was a mutual relationship or significant relevance between fly control and a decrease of infection diseases. In the other words the researches show that by controlling the number of flies in

different areas many of infection diseases such as diarrheic diseases, trachoma, shigellosis, gastroenteritis diseases and nosocomial infections with multiple antibiotic-resistant bacteria considerably decreased [19-22].

Recently, antibiotic-resistant, bacteria have been found in flies in hospital environments or near animal production sites [23,24]. Antibiotic resistance is a serious public health problem, for it under mines treatment efficacy and poses greater health risks [9]. An increasing frequency of antibiotic resistance has been reported by researchers from all over of the world. There have been no studies about the contamination of antibiotic-resistant pathogenic bacteria by *Lucilia sericata* in Sari. In this regard, the objective of this study was to identify and determine the frequency of resistance and susceptible bacteria isolated from blowflies in different regain of Sari City, North of Iran.

2. Material and Methods

This experimental study was conducted at Entomology and Microbiology Laboratory in Mazandaran University of Medical Science. Flies were collected during the 3 seasons from (April–December 2016).

2.1 Sample collection

A total of 600 adult house blow flies were independently captured using aerial insect nets four times a month randomly from public places including slaughter house, urban (public park, garbage dump, public toilet) and rural (garbage dump, stalls and local maintain animals waste) were selected owing to the observed abundance of adult blow flies and existence of ecological conditions enhancing their survival and persistence in Sari city. some of blow flies trapped were shown on Fig2. Flies were transferred by zip-locked plastic sterile bag to the Laboratory, and to identified species level by morphological characters. The flies of the same genus and species *(Lucilia sericata*) were batched and then individually rinsed in 10 ml sterile physiological saline solution for two minutes. Also, the flies' gut was picked using a sterile forceps, squashed in a sterile plate, and mixed with 10 ml physiological sterile serum. The solution of external body and gut was centrifuged with 2000 rpm for 5 minutes. The suspension cultured on EMB and Blood agar for 24 hours at 37ºC for determining the specious of the bacteria. Some other exclusive media such as TSB agar, SS agar, TSI agar, TCBS agar, XLD agar, Manitol salt Agar, PSE Agar and other specific and selective media were used. Gram staining, microscopic examination and biochemical tests such as IMVIC, oxidative–fermentative test, Urea test and other standard diagnostic tests were used to selection and identification of bacteria. The resulting isolates were characterized morphologically and further identifications were carried out following the methods of Bergey's Manual of determinative bacteriology [25].

2.2 Antibiotic susceptibility test

Antibiotic sensitivity pattern of bacteria isolates was determined on Muller Hinton agar plates by Kirby-Bauer disc diffusion method according to NCCLS [26]. The antimicrobial agents tested were: tetracycline, ampicillin, ciprofloxacin, gentamicin, cephalexin, cotrimoxazole, nalidixic acid, nitrofurantoin, amikacin, vancomycin, kanamycin, oxacilline which were purchased from PatanTeb (Iran) Company. Resistance data were interpreted according to National Committee for Clinical Laboratory Standards (NCCLS).

3. Results

The propagation of flies was increase in spring and summer. In the present study flies had one peak in their activities in accordance with temperature and humidity level (Table 1). From middle of summer to early autumn, the highest number of flies was seen in Tir (June to July) and Mordad (almost July to August), and the lowest number of flies were seen in Azar (almost October to November) and Dey (November to December) (Table 2). In the present study at North of Iran the highest number of trapped flies was in spring and summer, and the lowest number of flies was in autumn and winter (Fig. 1). The result shows seasonal changes in the abundance and extension of *Calliphoridae* mostly depend on environmental factors such as height, rainfall, temperature, humidity and reproduction substrates. The contamination of flies in summer (17.5%) was higher than other time of the year in Sari County (Table2 and Fig. 2). The Abundance of flies in slaughter houses was more than the urban and rural area.

Among the 600 blow fly collected, 216 (36%) were found to carry one or more species of bacteria on the external surface or in the alimentary tract. Four species of gram-positive bacteria were isolated from the *Lucilia sericata* species of house flies (Table 3). The most common gram positive bacteria isolated was *Staphylococcus aureus* with 34 cases (15.8%) and other gram positive isolated bacteria were belonging to *bacillus spp.* (10.2%), *Streptococcus spp*. (9.2%) and *Enterococcus faecalis* (7.4%). Contamination percentage of gram positive bacteria on the surface of flies was near than their digestive tract. Six species of gram-negative *Enterobacteriaceae* species were isolated from the flies (Table 3). Contamination percentage of gram negative bacteria in the alimentary tract was significantly higher than that of the external surface. The most common gram negative isolated bacteria were *Escherichia coli* with 41 cases (19%) and other gram negative isolated bacteria were belonging to *Proteus mirabilis* (7.8%), *Klebsiella pneum*onia (13%), *Citrobacter spp.* (5.1%), and *Entrobacter aerogene*s (11.1%). Some of the bacteria isolated were shown in Fig. 3.

Most of isolated bacteria from *Lucilia sericata* in the current study were pathogenic. Also, *E. coli and Staphylococcus aureus* were the most abundant bacteria that isolated from this flies (Table 3). Antibiotic resistance was tested for two most isolated bacteria cultures obtained from flies to determine their disc sensitivity or resistance to 12 commonly used antibiotics (Table 4). The percentage of antimicrobial susceptibility (sensitive, resistant and intermediate) obtained for this bacteria is presented in Table 3. In this study, *E. coli* isolates were susceptible to ciprofloxacin, gentamycin, nalidixic acid, nitrofurantoin, amikacin and resistance to 4 of 9 antibiotics tested containing tetracycline, ampicillin, cephalexin and Cotrimoxazole. While among all the isolates *S. aureus* resistance to 2 of 7 antibiotics tested containing tetracycline and ampicillin and they were sensitive to ciprofloxacin, gentamicin, amikacin, vancomycin, kanamycin and oxacillin. The result showed that vancomycin had high activity against *S. aureus* strains and ampicillin had low activity against these bacteria. ciprofloxacin, gentamycin and amikacin were active against both *E. coli* and *S. aureus*.

Table 1: The mean humidity and temperature range in variable month.

Variables Month	April	May	Jun	July	August	September	October	November	December
Temperature Range (min to max)	14-20	16-24	20-28	23-31	24-34	21-29	14-24	9-18	4-14
Humidity%	70	71	68	66	66	69	70	71	73

Table 2: Contamination Percentage & Frequency of Blow Files (*Lucilia Sericata*) in Sari.

season	Variables Month	Number of trapped flies(Percentage)	Total Number of trapped flies(Percentage)in each seasons	Contaminated number (Percentage)	Total contaminated (Percentage) of each seasons
Spring	March- April	65(10.9%)	202(33.6%)	13(6.01%)	63(10.5%)
	April- May	67(11.2%)		21(9.72%)	
	May-Jun	70(11.7%)		29(13.42%)	
Summer	Jun-July	82(13.6%)	243(40.5%)	33(15.3%)	109(18.16%)
	July-Aug	90(15%)		45(20.83%)	
	Aug-Sep	71(11.9%)		31(14.35%)	
Autumn	Sep-Oct	60(10 %)	155(25.83%)	20(9.25%)	44(7/34%)
	Oct-Nov	52(8.7%)		14(6.5%)	
	Nov-Dec	43(7.2%)		10(4.62%)	
Total		600(100%)		216(100%)	216(36%)

Table 3: Contamination percentage of bacteria isolated from *Lucilia sericata* (n =216).

Bacteria		Alimentary Tract		external body surface		Total bacteria	
		No	%	No	%	No	%
Gram positive bacteria	*Staphylococcus aureus*	15	7	19	8.8	34	15.8
	Bacillus. subtilis	9	4.1	13	6	22	10.2
	Streptococcus spp.	8	3.7	12	5.5	20	9.2
	Enterococcus faecalis	11	5.1	5	2.3	16	7.4
	Total G+	43	20	49	22.6	92	42.6
Gram negative bacteria	*E. Coli*	23	10.6	18	8.3	41	19
	Proteus mirabilis	12	5.6	5	2.3	17	7.8
	Klebsiella pneumonia	15	7	13	6	28	13
	Citrobacter.spp	9	4.1	5	2.3	14	6.5
	Entrobacter aerogenes	14	6.5	10	4.6	24	11.1
	Total G-	73	33.9	51	23.5	124	57.4
G+ &G- bacteria	Total G+&G-	116	53.9	100	46.1	216	100

Table 4: Antimicrobial susceptibility patterns of *E. coli* and *S. aureus* isolated from blow flies.

Antimicrobial agent	Microorganism	
	S. aureus	***E. coli***
Tetracycline	R	R
Ampicillin	R	R
Ciprofloxacin	S	S
Gentamicin	S	S
Cephalexin	NT	R
Cotrimoxazole	NT	R
Nalidixic acid	NT	S
Nitrofurantoin	NT	S
Amikacin	S	S
Vancomycin	S	NT
Kanamycin	S	NT
oxacillin	S	NT
Antimicrobial agent	Microorganism	
	S. aureus	*E. coli*
Tetracycline	R	R
Ampicillin	R	R
Ciprofloxacin	S	S
Gentamicin	S	S
Cephalexin	NT	R
Cotrimoxazole	NT	R
Nalidixic acid	NT	S
Nitrofurantoin	NT	S
Amikacin	S	S
Vancomycin	S	NT
Kanamycin	S	NT
oxacillin	S	NT

NT= not test, S= Sensitive, R= Resistance

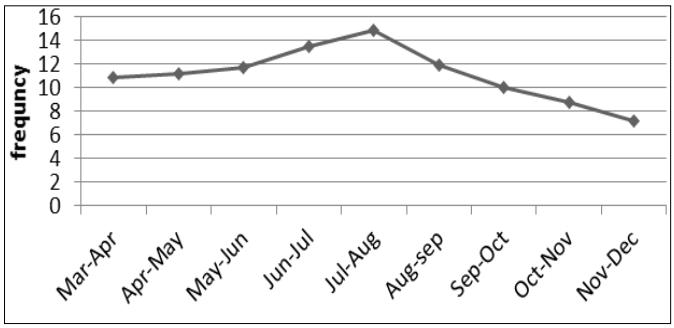

Fig 1: Frequency of Blow flies in each month

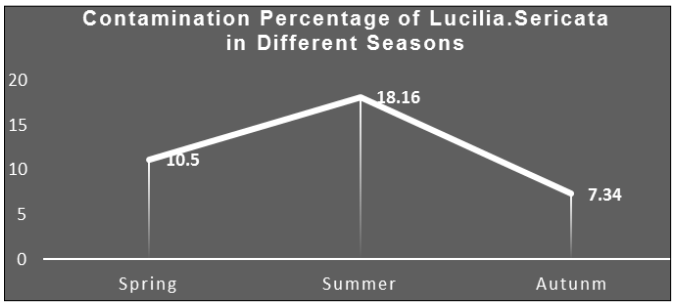

Fig 2: Contamination percentage of bacteria isolated from *Lucilia Sericata*

Fig 3: Some of bacteria isolated from *Lucilia sericata*

Fig 4: Some of trapped Blow flies *Lucilia sericata* in Sari City

4. Discussion

Mechanical transmission of various pathogenic agents such as bacteria by blow fly, *the Lucilia Sericata*, has been confirmed [1, 10, 19, 27]. Poultry houses, slaughter houses, hospitals, and other types of environments are sites where house flies can reproduce. In sometimes of the year, especially in seasons with optimum temperature and humidity and rainfall, the weather condition is convenient for flies' reproduction. They can transmit pathogen bacteria to human's food and living environment largely in warm seasons [5, 6]. The monthly abundance range of flies was significant in June, July and August in accordance with the rainfall, temperature and humidity. Similar observations were reported by Phasuk *et al*. [6]. who found *C. megacephala* in Brazil was abundant in fall (March–May) and summer (November–February). In India, the abundance of *C. megacephala* increased in correlation with the beginning of the rainy season and declined in the dry hot season [5].

Analysis of the data showed a significantly negative correlation for the populations of *C. megacephala* and *C. rufifacies* with the temperature and relative humidity. Seasonal changes in the abundance and distribution of *Calliphoridae* frequently depend on environmental variables such as altitude, rainfall, temperature, humidity, fauna, flora, breeding substrates and human disturbance [28, 6, 8]. The current study suggests that environmental factors such as the temperature and humidity are correlated with the abundance of blow flies in North of Iran. The abundance of flies in the Sari city was high in the majority of months. This can be due to appropriate weather and environment for their reproduction and breeding. According to the reports and observations, garbage dumping has been the most serious problem in Sari city. The accumulation of the garbage helps the proliferation of flies and the transmission of diseases. Waste management and improving environmental sanitation in these islands is an effective way to decreasing fly population and fly control. The abundance and relative frequency of medically important flies in the North of Iran, is very important. Most of the year time, especially in end of spring, summer and early autumn, the weather condition is so appropriate for flies' reproduction. The accumulation of wastes in this area also helps that. Therefore, effective strategy needs to be taken to control fly population in these seasons.

Recent studies suggest flies may play an important role in the spread of multidrug-resistant bacteria [29]. In the present study, a total of 216 samples (from 600 blow flies) as positive cultures were obtained. Most of isolated bacteria from *Lucilia sericata*in the current study were medically important. These findings agree with the results of Aigbodion F. *et al* [12]. In the current study, *E. coli* (19%) and *S. aureus* (15.8%) were isolated from blow flies in a non-hospital environment. Results showed that this flies may not only act as mechanical vectors but may also act as a biological vector of bacteria in digestive tracts. These studies demonstrate the ability of flies to carry bacteria with pathogenic potential to human beings and animals.

E. coli, a microorganism typically found in feces, was isolated from *muscids* and *caliphorids* in many studies [15, 16, 30, 31]. The percentage of bacteria isolated from guts was more than the body surface of the flies in the present study. These findings agree with the results of Pava-Ripoll M [1, 10] that show the prevalence of food-borne pathogens were three times greater in the guts than on the body surfaces of the flies. They were collected one hundred flies from the dumpsters of 10 randomly selected urban restaurants and then identified using taxonomic keys before being individually dissected. In their study *Cronobacter spp.,* (14%) *Salmonella* spp., and *Listeria monocytogenes* (3%) were detected using the PCR-based BAX systemQ7. Their selective flies were belonging to the housefly, *Musca domestica* (47%), the blowflies, *Lucilia cuprina* (33%) and *Lucilia sericata* (14%).

In the present study, different bacteria were isolated from *Lucilia sericata*. The bacteria were belonging to *S. aureus, B. subtilis spp., Streptococcus spp*., *Enterococcus faecalis, E. coli*, *Proteus mirabilis*, *Klebsiella pneumonia*, *Citrobacter. spp*., *Entrobacter aerogenes*. and *Yersinia enterocolitica*, these findings agree with the results of Aigbodion. I. F et.al in Benin City, Nigeria [12] which showed presence of *Micrococcus spp., Kelebsiella spp., Lactobacillus spp., Staphylococcus spp., Corynebacteria spp., Streptococcus spp., and E. coli.* on the external body parts of *phaenicia sericata* collected from three locations of city center. Their studies show that *S. aureus* (35%) and *Lactobacillus spp*. (20%) were the most species found in *P. sericata.* Their findings about *S. aureus* are in agreement with the present

finding.

One of a serious global public health problem today in developing countries is antimicrobial resistance. Many studies demonstrate the potential of flies to transmit multidrug-resistant bacteria from animals to the urban environment [30,31]. *E. coli* (77%), *Klebsiella pneumonia* (13%), *Pseudomonas aeruginosa* (6%), *Aerommas hydrophila* (4%) have also been isolated from flies including *Chrysomya megacephala*, *Aldrichina grahami*, *Lucilia sericata*, *Boettcherisca peregrima*, *Muscina stabulans* and *Bercaea cruenta* by other researchers in China. They show all isolates were resistant to amoxicillin, ticarcillin, cephalothin and cefuroxime. No resistance to meropenem and imipenem was observed [29].

Almeida et.al was determined the isolation frequency and microbial sensitivity profiles of *E. coli*, *Salmonella spp*., and *Staphylococcus spp*. isolated from synanthropic fly on dairy farms located in Northern Paraná, Brazil. they reported that the least effective drugs against Staphylococcus strains were penicillin G and tetracycline, whereas the most effective was sulfamethoxazole in association with trimethoprim, oxacillin, and gentamicin. Ceftriaxone, ciprofloxacin, enrofloxacin, and gentamicin were the most effective antibiotics against *E. coli* and *Salmonella Spp.* Tetracycline was the least effective antibiotic against the isolates. The *Staphylococcus spp*. strains isolated did not show the phenotype of oxacillin resistance. Ampicillin and tetracycline were the least effective antibiotics against *E. coli* [32]. Resistance to these antibiotics has been also observed in other studies [31, 33]. Moreover, present study showed that potentially pathogenic bacteria isolated from these insects were also found to have an increased resistance to antibiotics. These insects may also play some role for the transmission of bacteria with antibiotic resistance in different environment of regions in North of Iran.

5. Conclusion

The results of the current study confirm that flies are much more than a nuisance and that they pose potentially serious health risks as mechanical and biological vectors and the present study report that houseflies collected in urban and rural and slaughterhouse may be involved in the spread of drug resistant bacteria and may increase the potential for human and animal exposure to drug resistant bacteria. Therefore, they have to be controlled and density of their population should be reduced undertaken different vector control approaches.

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7. References

1. Pava-Ripoll M, Goeriz Pearson ER, Miller KA, Ziobro CG. Prevalence and relative risk of *Cronobacter spp*., *Salmonella spp.*, and *Listeria monocytogenes* associated with the body surfaces and guts of individual filth flies. Applied and Environmental Microbiology. 2012; 78(22):7891-7902.
2. Espra AS, Tabugo Sharon RM, Torres Mark AJ, Gorospe JG, Manting EM, Gorospe CG. Describing dimorphism in wing shapes in the Blowfly *Lucilia sericata* meigen *(Diptera: Calliphoridae)* using geometric morphometries. Advances in Environmental Biology. 2012; 9(19):64-70.
3. Whitworth T. Keys to the genera and species of blow flies (*Diptera: Calliphoridae*) of America north of Mexico. Proceedings of the Entomological Society of Washington. 2006; 108:689-725.
4. Islam T, Al-Maruf A, Mannan A, Rakibur Rahman H, Tarafder M, Samad A, *et al*. Isolation and identification of associated bacteria and maggots from myiasis affected wounds of cattle and goats in Bangladesh. Journal of Advanced Veterinary and Animal Research. 2015; 2(2):95-100.
5. Wall R, Howard JJ, Bindu J. The seasonal abundance of blowflies infesting drying fish in south-west India. Journal of Applied Ecology. 2001; 38:339-348.
6. Phasuk Jumnongjit, Tharawoot Thitima, Chanpaisaeng Jariya. Seasonal abundance of Blow flies (*Diptera: Calliphoridae*) in three urban parks of Bangkok Thailand. Kasetsart Journal Natural Science. 2013; 47:828-834.
7. Sanei-Dehkordi A, Khamesipour A, Akbarzadeh K, Akhavan AA, Rassi Yavar, Oshaghi MA, *et al*. Experimental colonization and life table of the Calliphora vicina (Robineau-Desvoidy) (Diptera: Calliphoridae). Journal of Entomology and Zoology Studies. 2014; 2(6):45-48.
8. Sukontson KL, Bunchoo M, Khantawa B, Piangjai R, Somsak Y, Sukontason K. Comparison between Musca Domestica and Chrysomya Megacephala as carriers of bacteria in Northern Thailand. The Southeast Asian Journal of Tropical Medicine and Public Health. 2007; 38(1):38-44.
9. Barreiro C, Albano H, Silva J, Teixeira P. Role of flies as vectors of foodborne pathogens in rural areas. ISRN Microbiology, Hindawi Publishing Corporation. 2013; 718780:1-7.
10. Parva-Ripoll M, Goeriz Pearson ER, Miller KA, Ziobro CG. Detection of Foodborne Bacterial Pathogens from Individual Filth Flies. Journal of Visualized Experiments. 2015; (96):52372-52379.
11. Ma Qun, Fonseca Alicia, Liu Wenqi, Fields AT, Pimsler ML, Spindola AF, *et al*. Proteus mirabilis inter kingdom swarming signals attract blow flies, The ISME Journal. 2012; 6:1356-1366.
12. Aigbodion FI, Egbon IN, Obuseli AJ. Pathogens of medical importance isolated from *Phaenicia Lucilia sericata* (*Diptera: Calliphoridae*) in Benin City Nigeria. Pakistan Journal of Biological Sciences. 2013; 16(23):1791-5.
13. Singh B, Crippen TL, Zheng L, Fields AT, Yu Z, Ma Q *et al*. A metagenomic assessment of the bacteria associated with *Lucilia sericata* and *Lucilia cuprina* (Diptera: Calliphoridae). Applied Microbiology and Biotechnology. 2015; 99(2):869-83.
14. Kobayashi M, Sasaki T, Saito N, Tamura K, Suzuki K, Watanabe H, *et al*. Houseflies: not simple mechanical vectors of enterohemorrhagic Escherichia coli O157:H7. American Journal of Tropical Medicine and Hygiene. 1999; 61(4):625-9.
15. Sasaki T, Kobayashi M, Agui N. Epidemiological potential of excretion and regurgitation by *Musca domestica (Diptera:* muscidae) in the dissemination of *Escherichia coli O157: H7* to food. Journal of Medical Entomology. 2000; 37:945-949.
16. Fotedar R. Vector potential of house flies (*Musca domestica*) in the transmission of Vibrio cholera in India. Acta Tropica. 2001; 78:31-34.
17. Holt PS, Geden CJ, Moore RW, Gast RK. Isolation of *Salmonella enterica* Serovar Enteritidis from Houseflies

(*Musca domestica*) Found in Rooms Containing *Salmonella* Serovar Enteritidis-Challenged Hen. Applied and environmental Microbiology. 2007; 73(19):6030-6035.
18. Chaiwong T, Srivoramas T, Sueabsamran P, Sukontason K, Sanford MR, Sukontason KL. The blow fly *Chrysomya megacephala* and the house fly *Musca domestica* as mechanical vectors of pathogenic bacteria in Northeast Thailand. Tropical Biomedicine. 2014; 31(2):336-346.
19. Graczyk TK, Knight R, Gilman RH, Cranfield MR. The role of nonbiting flies in the epidemiology of human infectious diseases. Microbes and Infection. 2001; 3:231-235.
20. Esrey SA. Effects of improved water supply and sanitation on ascariasis, diarrhoea, dracunculiasis, hookworm infection, schistosomiasis and trachoma. Bulletin of World Health Organization. 1991; 69:609-621.
21. Cohen D, Green M, Block C, Slepon R, Ambar R, Wasserman SS, *et al*. Reduction of transmission of shigellosis by control of houseflies (*Musca domestica*). Lancet. 1991; 337:993-997.
22. Emerson PM, Lindsay SW, Walraven GEL, Faal H, Bogh C, Lowe K. Effect of fly control on trachoma and diarrhoea. Lancet. 1999; 353:1401-1403.
23. Ahmed A, Ghosh A, Schal C, Zure KL. Insects in confined swine operations carry a large antibiotic resistant and potentially virulent enterococci community. BMC, Microbiol. 2011; 11:23-25.
24. Vazirianzadeh B, Shams SS, Rahdar M, Hajhossien R, Mehdinejad M. Identification of bacteria which possible transmitted by *Musca domestica* (*Diptera: Muscidae*) in the region of Ahvaz SW Iran. Jundishapur Journal of Microbiology. 2008; 1(1):28-31.
25. Bergey DH, Buchanan RE, Gibbons NE. Bergey's manual of determinative bacteriology. Baltimore. Williams & Wilkins, 1974.
26. National Committee for Clinical Laboratory Standards (NCCLS). Performance Standards for Antimicrobial Disk Susceptibility Tests. Approved Standard NCCLS. Villanova. PA. fifth ed. 1993.
27. Graczyk TK, Knigh Ronald, Tamang Leena. Mechanical transmission of human protozoan parasites by insects. Clinical Microbiology Reviews. 2005; 18:128-132.
28. Amat E, Tonus MMA, Albertino RJ. A survey of necrophagous blowflies (Diptera: Oestroidea) in the Amazonas-Negro inter fluvial region (Brazilian Amazon). Revista Brasileira de Entomologia. 2016; 60:57-62.
29. Liu Y, Yang Y, Zhao F, Fan X, Zhong W, Qiao D, *et al*. Multi-Drug resistant gram-negative enteric bacteria isolated from flies at Chengdu Airport China. The Southeast Asian Journal of Tropical Medicine and Public Health. 2013; 44(6):988-996.
30. Davari B, Kalantar E, Zahirnia A, Moosa-Kazemi SH. Frequency of resistance and susceptible bacteria isolated from houseflies. Journal of Arthropod-Borne Diseases. 2010; 4(2):50-55.
31. Zurek LB, Ghosha A. Insects represent a link between food animal farms and the urban environment for antibiotic resistance traits, Applied and Environmental Microbiology. 2014; 80(12):62-67.
32. Almeida JL, Giuffrida R, Andrade APR, Chaves MP. Muscoid Diptera as potential vectors of bacterial agents on dairy farms in the northern region of Paraná. Brazil. Semina Ciências Agrárias Londrina. 2014; 35(6):3127-3138.
33. Alam MJ, Zurek L. Association of Escherichia coli O157:H7 with Houseflies on a Cattle Farm. Applied and Environmental Microbiology. 2004; 70(12):7578-7580.