Dissemination of Enterobacteriaceae to Human Beings Through Houseflies in Different Areas of Lahore City

*MOHAMMAD HUMAYUN, *MUNAZA HASAN, **NASEER AHMAD CHAUDHRY

ABSTRACT
Acute gastroenteritis is probably the most important public health problem. The association of flies as mechanical vectors in transmission of the causative agent is evident due to their behavioural characteristics which ensure their contact with food and wastes of man and animals. The present study was of six months duration from 1st June, 2008 to 30th November, 2008, 7200 flies were used for bacteriological examination from six sanitary and six insanitary areas of Lahore city. This study revealed that bacterial contamination of houseflies varied with the site of its breeding. Overall frequency of orofaecal transmitting bacteria was higher in the insanitary areas whereas in the sanitary areas bacterial isolates were more of non-orofaecal transmitting group. It has become evident from this study that sanitary conditions should be improved to minimize the breeding of house-flies in Lahore.

Key words: Houseflies, Dissemination, Enterobacteriaceae

INTRODUCTION
The family Enterobacteriaceae is composed of a large number of closely related species of gram negative non-sporo forming rods that are found in soil, water, decaying matter and the large intestines of humans, animals and insects. Enterobacteriaceae the most frequently responsible for human enteric infections are Eschericia coli, Proteus vulgaris, Proteus mirabilis, Klebsiella pneumoniae, Pseudomonas aeruginosa, Citrobacter freundii, Salmonella typhimurium, Salmonella typhi, Shigella species and Serratia species1-4.
The association of houseflies as mechanical vectors in the transmission of the causative agents of diarrhea or gastroenteritis has been demonstrated in many countries of the world. The pathogens may invade either directly or after their multiplication in various contaminated foodstuffs. Control or eradication of fly population has coincided with a reduction in the incidence of gastroenteritis. They are common household pests; being associated closely with human beings and animals3,5.

Houseflies have been shown to be a menace to human health since ancient times and are recognized as transport hosts for a variety of bacterial, parasitic and viral agents5-13. They are forever in search of food and because of their high intake of nourishment they defecate very often, so that a well-fed insect may deposit faeces constantly with as little as 5 minute interval between each drop of faeces. This constant discharge of excreta is one of the many factors that make the housefly a very dangerous carrier of disease.5 The larvae of many species of biting and non-biting flies are laid down on filth or garbage whereas the adult flies often feed upon this material while doing so they may carry both in or on their bodies the putrefactive and faecal bacteria acquired from the substances on which they feed and also carry and distribute many disease producing species of bacteria to the food substances thus spreading these organisms10-13.

Bidawid et al8 studied the role of non-biting flies in the transmission of enteric pathogens in Beirut, Lebanon. Extracts were prepared from batches of 15 houseflies. They found 29 isolates of Shigella and 19 isolates of Salmonella from bacteriological studies of 72 batches of houseflies. Among the isolates from Musca domestica there were Shigella dysenteriae shiga, Shigella sonnei, Salmonella typhi, Salmonella enteridis and Salmonella typhimurium.

As the population of Lahore is increasing day by day and insanitary conditions also prevail in large part of the city, therefore the present study was planned to find out the role of houseflies as carriers of enterobacteriaceae in Lahore city.

MATERIALS AND METHODS
The present study included 7200 houseflies from six insanitary and six sanitary localities of Lahore. Five different places (garbage dumps, butcher’s shops/slaughter houses, sweet shops, open kitchens and gardens) of twelve different localities viz: six sanitary and six insanitary were selected from said places each month over a period of six months in the batches of 15 each. The insanitary localities were
Bakar Mandi, Juggian Shahabuddin, Mohammadi Kot, Kachi Abadi of Shahnoor, Sodhiwal and Icchra. The sanitary localities included Wahdat Colony, Allama Iqbal Town, New Muslim Town, Shadman Colony, Gulberg and GOR-1. They were then transported to the laboratory in sterilized test tubes which were fumigated with chloroform. Upon reaching the laboratory the extracts were prepared by grinding the flies in 15 ml of sterile normal saline solution.

Direct staining of extracts was done and subsequently the cultures were inoculated onto blood agar, Mac-Conkey’s agar, S S agar, XLD agar and TCBS agar plates at 37°C for 24 hours, aerobically. Next day the plates were examined for any growth and if needed incubation were extended to 48 hours. At the same time 1 ml of each well-mixed extract was added to a test tube containing 5 ml of Selenite broth and 5 ml of alkaline peptone water and they were incubated overnight at 37°C. Next day subcultures were made onto TCBS agar plates. These plates were incubated aerobically and studied next day for bacterial growth.

**RESULTS**

The study was specifically oriented to detect the dissemination of enterobacteriaceae to human beings through houseflies. The whole study was of six months duration and the period extended from 1st June 2008 to 30th November 2008; 7200 flies were used for bacteriological examination from six insanitary and six sanitary localities of Lahore.

Comparison of Gram negative bacterial isolates of the insanitary and sanitary areas revealed that Eschericia coli isolated from insanitary areas were significantly higher (p<0.01) when compared with sanitary areas. The Enterobacter species and Citrobacter freundii isolated from the sanitary areas were significantly higher when compared with the insanitary areas (Table 1). The study thus revealed that overall frequency of isolates of enterobacteriaceae was higher in the insanitary areas as compared to sanitary areas (Fig. 1).

<table>
<thead>
<tr>
<th>Isolates</th>
<th>Insanitary areas (n=180)</th>
<th>Sanitary areas (n=180)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Escherichiacoli</td>
<td>95*</td>
<td>76</td>
</tr>
<tr>
<td>Proteusvulgaris</td>
<td>38</td>
<td>35</td>
</tr>
<tr>
<td>Proteusmirabilis</td>
<td>39</td>
<td>36</td>
</tr>
<tr>
<td>Klebsiellapneumoniae</td>
<td>29</td>
<td>32</td>
</tr>
<tr>
<td>Pseudomonasaeruginosa</td>
<td>24</td>
<td>27</td>
</tr>
<tr>
<td>Enterobacterspecies</td>
<td>17</td>
<td>27**</td>
</tr>
<tr>
<td>Citrobacterfreundii</td>
<td>7</td>
<td>18***</td>
</tr>
<tr>
<td>Salmonella typhimurium</td>
<td>10</td>
<td>8</td>
</tr>
<tr>
<td>Salmonellatyphi</td>
<td>5</td>
<td>4</td>
</tr>
<tr>
<td>Shigella species</td>
<td>9</td>
<td>8</td>
</tr>
<tr>
<td>Serratiaspecies</td>
<td>8</td>
<td>13</td>
</tr>
</tbody>
</table>

*Significantly more (p<0.01) when compared with sanitary areas
**Significantly more (p<0.02) when compared with insanitary areas
***Significantly more (p<0.05) when compared with insanitary areas

![Fig.1: Comparison of Gram negative bacterial isolates](image-url)
DISCUSSION

Houseflies are cosmopolitan, ubiquitous, synanthropic insects that serve as mechanical or biological vectors for various micro-organisms. They are a source of nuisance to human beings. The local distribution of flies is mainly dependent upon the existence of adequate breeding places. Since they feed upon and breed in septic substrates, houseflies are important in the transmission and dissemination of bacterial disease agents, especially in countries with poor sanitation. The presence of a large number of flies is a direct indication of the poor sanitary conditions of that particular locality. In the insanitary localities the usual feature has been found to be the presence of heaps of refuse and filth scattered here and there. Faecal matter is also scattered due to indiscriminate defecation and open latrines in most of the houses. The presence of kitchen wastage in the open and the preparation of sweets are freely exposed to the flies. Moreover, the meat shops and slaughterhouses provide a significantly rich source of feeding places for the flies. Aforesaid factors provide sufficient opportunities for the mechanical transmission of bacteria through its appendages, vomitus and the faecal matter.

The population of Lahore has been on the increase day by day. The insanitary conditions have been observed to be prevailing in larger parts of the city. In such conditions the abundance of houseflies and their menace to public health cannot be overlooked. Keeping in view this fact the present study was undertaken to assess the magnitude of the problem.

The results of bacterial study showed that Enterobacteriaceae viz: Eschericia coli, Proteus Vulgaris, Proteus mirabilis, Klebsiella pneumoniae, Pseudomonas aeruginosa, Enterobacter species, Citrobacter freundii, Salmonella typhimurium, Salmonella typhi, Shigella and Serratia species were isolated in the different sanitary and insanitary areas of Lahore. Bidawid et al conducted a study on the role of flies in the transmission of enteric pathogens in Beirut, Lebanon. They detected 29 isolates of Shigella and 19 isolates of Salmonella species from enteric bacteria detected were Proteus, Pseudomonas and Salmonella species. The frequency of Salmonella and Proteus species seemed to correlate more with accessibility of flies to dog excrement than to rendered meat. They concluded that the high incidence of enteric contamination of filth flies showed them as mechanical carrier of enteric disease.

The present study revealed that the bacterial contamination of houseflies varied with the site of its breeding. Overall frequency of orofaecal transmitting bacteria was higher in the insanitary areas whereas in the sanitary areas bacterial isolates were more of non-orofaecal transmitting group.

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