Lahore, Pakistan’dan toplanan Hamamboceklerinin Vucut yüzeylerinden İzole Edilen Patojen Bakteriler Arasında Dezenfektan Direncinin Yaygınlığı

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Prevalence of Disinfectant Resistance among Pathogenic Bacteria isolated from body surfaces of Cockroaches collected from Lahore, Pakistan

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Introduction

• Nosocomial infections and food-borne illness are very much common in developing countries and lethal for newborns as well as children and adults.

• 1.7 million hospital-associated infections, cause or contribute to 99,000 deaths each year in US.

• In Europe, 2/3rd of the 25,000 deaths each year cause by nosocomial infections.
Introduction

• Cockroaches are suspected as a cause of nosocomial infections and food borne illness

• Indoor infestation of cockroaches is a major cause of mechanical transmission of pathogens

• serve as a sort of public transit for the busy microbiological world (bacteria, fungi, viruses, parasites)
Cockroaches infestation and consequences
Habitat and ecology

- Cockroaches have survived on the earth for more than 300 million years virtually without change.
- Tropical insects and preferred a home that is both warm and moist.

- Found in abundance near leaking faucets or pipes, damp corners, drains, kitchens, bathrooms and maintenance rooms with sink taps.

- Their filthy breeding habits, feeding mechanisms and indiscriminate travel between filth and food make them efficient vectors of pathogenic bacteria.
Bacterial carriage of cockroaches

- Bacterial carriage role can be explained on the basis of their favorable hideouts in toilets during the daytime and frequently feeding on human feces, from which they acquired and have copious opportunity to disseminate pathogenic Bacteria, helminthes, fungi, and protozoa on food.

- In the evening time, they migrate into kitchens, bedrooms and living rooms where they deposit pathogens present on their body surface on kitchen utensils, clothing in bedrooms and on chairs in the living rooms.
Aims of study

• Despite that a number of studies have been done in other countries worldwide, to our knowledge there are few studies conducted on Microbial screening of cockroaches in the Pakistan.

• Hence, this study aims to isolate disinfectant resistance bacterial contaminants present on the external surfaces of cockroaches in houses, food stores and offices of urban areas of Lahore.
Hypothesis of research

• Cockroaches are possible mechanical vector of resistant bacteria in human dwellings
METHODOLOGY

❖ **Experimental site:** Entomology Research Laboratory, LCWU, Lahore.

❖ **Study area:** urban area of district Lahore

❖ **Collection and identification of cockroaches:**

- 200 samples collected from different houses, food stores and offices of urban area of Lahore
- Sticky traps, food baited traps and manually by hand
- Identification of adults by published pictorial key
**Microbial screening**

- Isolation of bacterial contaminants on selective and differential media

- **Inoculation of samples** on TSA, MSA, MacConkey agar, EMB, SS agar, BAP for 24-36 hrs at 37°C.

- **Identification of Bacteria** by colony morphology, gram staining, biochemical tests (oxidase, catalase, coagulase and IMViC test)
Efficacy of Common Disinfectants for Bacterial Isolates

- Efficacy was tested by **well diffusion method** against
  1. Germ kill ventocil IB
  2. G-cide as crystal HLD
  3. Germ kill ventocil FHC
- **Serial dilution of disinfectants** followed by pouring of disinfectants in holes made in MH agar streaked plates
- Sterile broth was used as negative control.
- **Zone inhibition diameter** was measured according to CLSI guidelines (2014) and % **RIZD** was also calculated and efficacy was denoted as (+) or (-).
Results

- Collection and Identification of Cockroaches
- 2 species of cockroaches were identified
  
a) Periplaneta americana
  
b) Blattella germanica
American Cockroach (*Periplaneta americana*)

- Adults are **reddish brown** with a yellowish figure 8 pattern on the light colored pronotum.
- Length of two inches.
- Young nymphs are greyish brown.
- Found in sewers and basements especially around pipes and drains.
German Cockroach (*Blattella germanica*)

- Adults have light brown bodies and two dark stripes on the pronotum
- Length of average 0.5 inches
- Both have wings extending to the end of their abdomen but they do not fly.
- Found in kitchens, storage areas especially where food being prepared or stored.
Bacterial Isolates

- Common Pathogenic bacteria isolates from cockroaches are *Klebsiella pneumonia*, *Escherichia coli*, *Staphylococcus aureus*, *Enterobacter aerogenes*, *Salmonella typhi*, *Shigella dysentriae* and *Pseudomonas aeruginosa*.

- **Bacterial Infestation on cockroaches**
  - Both species have no significantly different bacterial load
  - All cockroaches were found to be infected with at least 1 bacterium
E. coli
K. Pneumoniae and P. aeruginosa
Efficacy of common disinfectants for bacterial isolates

• Bactericide effect depends on diffusion process of disinfectants in the media.
• The diffusion process depends on numerous factors including number, size and shape of particles.
• Besides polarity of samples and temperature, the pH of solvents also can affect the results of bactericides.
• Germ kill Vantocil FHC to be the strongest antimicrobial agent irrespective of the dilutions when compared with the other disinfectants.
• Disparity from recommended dose of disinfectant may be due to the water of different sources used to dilute the disinfectants.
Table: Means ± SEM of inhibition zone diameter using different concentration of disinfectant against bacteria isolated from cockroaches.

<table>
<thead>
<tr>
<th>Bacterial isolate</th>
<th>Zones of growth inhibition diameter (mm)</th>
<th></th>
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<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Germ kill vantocil IB (v/v)</td>
<td>G-cide as crystal HLD (v/v)</td>
<td>Germ Kill Vantocil FHC (v/v)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>12.5%</td>
<td>25%</td>
<td>50%</td>
<td>12.5%</td>
</tr>
<tr>
<td>E. coli</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>16.14±0.84</td>
<td>14.28±0.94</td>
<td>21±1.0</td>
<td>19±3.6</td>
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<tr>
<td>S. aureus</td>
<td>14.77±2.38</td>
<td>12.11±1.90</td>
<td>22.11±1.68</td>
<td>19.42±2.63</td>
</tr>
<tr>
<td>S. typhi</td>
<td>16±1.41</td>
<td>14±1.67</td>
<td>12±1.67</td>
<td>13.6±0.98</td>
</tr>
<tr>
<td>S. dysentriae</td>
<td>14±1.76</td>
<td>12±2</td>
<td>16±3.06</td>
<td>12.66±1.15</td>
</tr>
<tr>
<td>P. aeruginosa</td>
<td>15±1</td>
<td>13±1</td>
<td>11±1</td>
<td>21.66±2.73</td>
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<tr>
<td>K. pneumoniae</td>
<td>18.25±1.03</td>
<td>17.25±0.75</td>
<td>15.5±0.96</td>
<td>22.66±6.68</td>
</tr>
<tr>
<td>P. mirabilis</td>
<td>22±3.06</td>
<td>17.33±2.40</td>
<td>13.33±3.33</td>
<td>24±1.5</td>
</tr>
<tr>
<td>% RIZD</td>
<td>45.45</td>
<td>18.18</td>
<td>-9.18</td>
<td>70</td>
</tr>
</tbody>
</table>
Table: Effectiveness of selected disinfectants agents on bacteria isolated from cockroaches.

<table>
<thead>
<tr>
<th>Bacterial isolate</th>
<th>Antibacterial activity of surface disinfectants (++, +, -)</th>
<th>Germ kill vantocil IB</th>
<th>G-cide as crystal HLD</th>
<th>Germ Kill Vantocil FHC</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
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<td>12.5% (v/v)</td>
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<td>50% (v/v)</td>
</tr>
<tr>
<td>E. coli</td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>S. aureus</td>
<td>++</td>
<td>++</td>
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<td>S. typhi</td>
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<tr>
<td>S. dysentriae</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>P. aeruginosa</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>++</td>
</tr>
<tr>
<td>K. pneumonia</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>++</td>
</tr>
<tr>
<td>P. mirabilis</td>
<td>++</td>
<td>+</td>
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</tr>
</tbody>
</table>

*++ (Heavy Growth of microorganisms), + (Moderate growth of microorganisms), - (No growth observed), a (MIC for S. typhi), b (MIC for S. typhi, S. dysentriae), c (MIC for E. coli, K. pneumoniae).
Conclusion

• Gram negative bacterial isolates were relatively higher than Gram positive isolates

• Important key points of food borne illness
  - Source of food ingested
  - Residential type
  - Level of hygiene

• Presence of cockroaches in nosocomial environment and isolation of nosocomial agents from cockroaches may develop a connection in proving hypothesis of research
• Cockroaches highly adapted for diverse land environment (HSP, acclimatization)

• infestation rate of the cockroaches can related to the residential types, availability of food sources, sanitary conditions and climatic conditions
Some Related Research in Pakistan


• HAFSA MEMONA, FARKHANDA MANZOOR, & AFTAB AHMAD ANJUM. 2014. Cockroaches (Periplaneta americana L. and Blattella germanica) as Potential Vectors of Nosocomial Infections in Hospitals of Lahore, Pakistan. BIOLOGIA (PAKISTAN) 2014, 60 (2), 295-297

Prevention is better than cure!

Don’t give bacteria a free ride.
Say no to cockroach
say no to disease
THANKS