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THE GHOST ANT *TAPINOMA MELANOCEPHALUM* (FORMICIDAE) AS MECHANICAL VECTOR OF CLINICALLY IMPORTANT BACTERIA

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Abstract

The wide range of foraging ants have facilitated carries in their bodies a high microbial load that have been transported to different places. It is accepted that ants can act as mechanical vectors for the human pathogens, due to their natural behavior and wide distribution representing a high health risk. For this reason, the ghost ant *Tapinoma melanocephalum* (Formicidae) was studied in order to identify its contribution as a mechanical vector in two areas located near to public hospitals in the Barranquilla city, the food services area and the Adult Intensive Care Unit (ICU). Two types of samples were taken in each site: (1) Ants attracted to tuna baits and (2) sampling with swabs directly from soil. After sampling, 93 individuals distributed in 38 and 55 for ICU and the food services area, respectively were registered. Bacteria belonging to the genera *Staphylococcus, Bacillus, Enterobacter, Enterococcus, Micrococcus, Salmonella, Shigella*, and *Streptococcus* were present in both samples (ants and soil). However, only the genera *Enterobacter* and *Micrococcus* were identified in ant samples, suggesting that *T. melanocephalum* could be carrying pathogenic bacteria into this public hospital acting as a mechanical vector in these areas to both pathogens. Finally, it is important to verify whether this insect pest can act as mechanical vector of diseases associated to patient care in order to overcome the problem and reduce a potential health risk of nosocomial infections.

Key words: Urban Ants, Nosocomial infections, mechanical vectors.

Introduction

Within the family Formicidae stand urban ants or "tramps ants", due to their biological and ecological characteristics that have allowed them to adapt to urban environments; invading human houses and their workplaces; sometimes acting as mechanical vectors of some known pathogens [1,2]. Several previous studies have demonstrated the ability of urban ants to carry pathogens into hospital environments, which have been associated with nosocomial infections [2–13].

Brazil is the Latin American country where ants have been broadly studied as vectors of nosocomial diseases. The first research work was carried out in 1993, reporting 14 ant species being present in hospitals in the State of São Paulo, acting as mechanical vectors by transporting bacteria in their bodies [14]. In Colombia the study of urban ants as mechanical vectors of germs has had little importance, highlighting only one study in the department of Valle del Cauca where the presence of seven species of ants was registered, which were associated with 14 types of pathogenic bacteria found in clinics and hospitals belonging to local health system [4]. The preset study evaluated the presence of ghost ants and pathogenic bacteria in the Adults Intensive Care Unit and food service areas belonging to a public hospital (level IV) in the Barranquilla city. The study shows the relevance and impact that this issue has on public health; especially taking in account its relationship with hospital-acquired infections, which causes of human death and increased costs associated with prolonged stay of infected patients.

Methods

Samples collection

The designed protocol to collect samples was approved for the scientific committee of the hospital selecting two areas: Adult Intensive Care Unit (ICU) and the area of food services. The first are has critically ill patients who need specific hospitalization and intensive care. In the second selected area is located the unit that handle and provides all nutritious meals and snacks to hospitalized patients. Strategic and randomized sites were selected in these areas in order to put baits containing 3 g of tuna, which were previously autoclaved [15]. These baits were observed three times during 90 minutes; afterwards, all ants that reached the baits were collected using a sterile brush. These individual ants were stored in sterile Eppendorf tubes, which were sealed and transported to the Laboratory of Microbial Biotechnology at the Universidad del Atlántico. Additionally, with the aid of sterile swabs, floor samples were taken from selected sites in order to compare the bacterial flora carried by ants with these environments. Sample collecting procedures were repeated three times along seven months (August – February).

Sample processing and organism identification

The individual identification was performed following the taxonomic key described by Fernandez (2003) [1] and by comparison with ants preserved at the Entomology Museum of the University del Valle. Some ants were used for identification purposes and the remaining ones were deposited in test tubes containing nutrient broth. A same procedure was performed with samples collected using swabs. Posteriorly, 100 µL of nutrient solution were taken from each tube exhibiting positive bacterial growth, and then inoculated by using striated by exhaustion onto nutrient agar plates in order to isolate pure colonies of microorganisms present in the two samples (bait and drag swab). Afterwards, the morphology, identification and partial characterization of bacteria was performed using light microscopy, Gram staining, biochemical tests and manual identification by following the protocol described by Bergey`s (2005) [16].

Statistical analysis

For statistical analysis, all tests were evaluated in triplicate. All descriptive analysis and plots were performed using SPSS (version 17.0) for Windows (SPSS, Inc., Chicago, IL). The hierarchical clustering analysis was carried out by using UPGMA (Unweighted Pair Group Method using Arithmetic Mean) with PAST3.11 software package (2016).

Results

Insect monitoring

A total number of 93 individuals of *T. melanocephalum* were collected during sampling, being 38 individuals captured in the ICU and 55 in the area of food service. Figure 1 show the abundance and ant distribution by area.

Human pathogens isolation and identification

A total of 71 bacteria strains distributed in eight genera were observed. A group of 39 strains was successfully associated to insect activity as mechanical vectors. It is important to point out that *T. melanocephalum* was found to have the highest bacterial diversity. The figure 2 show the distribution of bacteria genera in the food service area. Only three genera were isolated from these samples: *Bacillus*, *Enterococcus* and *Staphylococcus*, being just the genus *Bacillus* the only one that was present in both swab and ant *T. melanocephalum* samples. Eight genera of bacteria from the ICU were isolated and properly identified (Figure 3), five of which were being transported by *T. melanocephalum* inside the building. However, only Enterococcus sp., Micrococcus sp. and Enterobacter sp. were exclusively associated to the ant. This information was corroborated using statistics focused on presence of human pathogens at the hospital. The results show that five genera *Bacillus*, *Salmonella*, *Shigella*, *Staphylococcus,* and *Streptococcus* were isolated from hospital floor samples. Particularly, only the genus *Bacillus* was present at the kitchen area. In addition, the genera *Salmonella*, *Shigella* and *Streptococcus* were not found in ants. A higher percentage of similarity (60%) of microbial load similarity in the ant *T. melanocephalum*, regarding to both studied areas, was found in this study (Figure 4), due to the presence in both places of the genera *Staphylococcus* and *Enterococcus*. However, the ICU had a higher bacterial load than the feeding area, showing the *Enterobacter* and *Micrococcus* genera as unique pathogens in this area. By contrast, the microorganisms from the two floor areas were dissimilar, with less than 18% similarity.

Discussion

The *T. melanocephalum* activity at the Hospital located in the Barranquilla city (Colombia) was monitored during sampling in order to determine its participation as a mechanical vector of human pathogens. Previously, Teixeira et al. (2009)[15] reported that the ghost ant was associated to the microbiota present in a Brazilian University Hospital. According to Olaya et al. (2005)[4], this tramp ant specie has the highest incidence in hospitals (43.7%). The *Staphylococcus* sp. and *Enterococcus* sp were the most frequent genera in the feeding area; which coincides with the studies realized by De Zarzuela et al. (2004)[6] conducted in bathrooms, residential kitchens and food outlets. Commonly these genera are highly pathogenic and have been associated with nosocomial infections at clinic level [2–7,9,11–15,17–19]. The *Enterobacter* sp., *Enterocuccus* sp., and *Micrococcus* sp. resulted to be the most common genera in the Intensive Care Unit, a similar result when compared with findings showed in other researches [4,6,14,20]. Our results show that 31 isolates are considered a cause of nosocomial infections. This result is in agreement with the study realized by Olaya et al, (2005)[4] and Zarzuela et al (2004)[6], who not only identified similar microorganisms but also associate them to *T. melanocephalum* as the mechanical vector that increased load of human pathogens into Hospitals. The current information provide by this study should be taken in account, if we consider the important activities realized inside these places, and the human health risk associated with the presence of clinically important bacteria and other kind of pathogens; which are responsible of death in immunosuppressed patients who frequent this area. This information could lead to design strategies for the management of nosocomial infections by either reducing or controlling mechanical vectors in these areas. In this study, we observed the presence of *Enterobacter* sp. associated with ant *T. melanocephalum* locomotion[10]. *Enterobacter* sp. is one of the most common clinically important pathogen identified in hospitals. This pathogenic bacterium causes approximately 70% of urinary and intestinal infections around the world [21,22]. For this reason, it is necessary to control this insect pest and to improve the sterilization protocols inside hospital-related buildings in order to avoid potential and dangerous nosocomial infections.

Hospital-acquired infection (HAI)

According to statistics, several genera were identified as the main causes of nosocomial infections at the ICU (Figure 5), during a year sampling period in a hospital belonging to health system of the Barranquilla city. During the same time, the ICU evidenced a higher number of nosocomial infections, being 81 cases with origin in surgical sites, followed 65 cases of urinary infection mainly associated with treatment procedures (Figure 6). The results suggest that microorganisms isolated in this study are responsible for the occurrence of nosocomial infections at the adult ICU unit. In addition, we think that these infections could be linked to the locomotion of *T. melanocephalum* ant and its behavior inside the Hospital in where this ant acts as mechanical vector.

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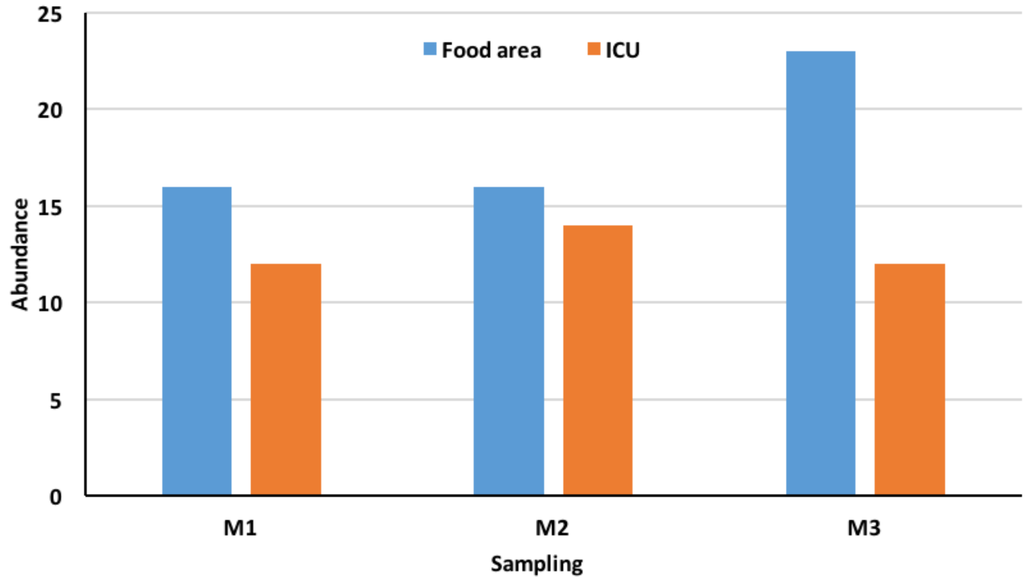

Figure 1. Abundance and ant distribution by area.

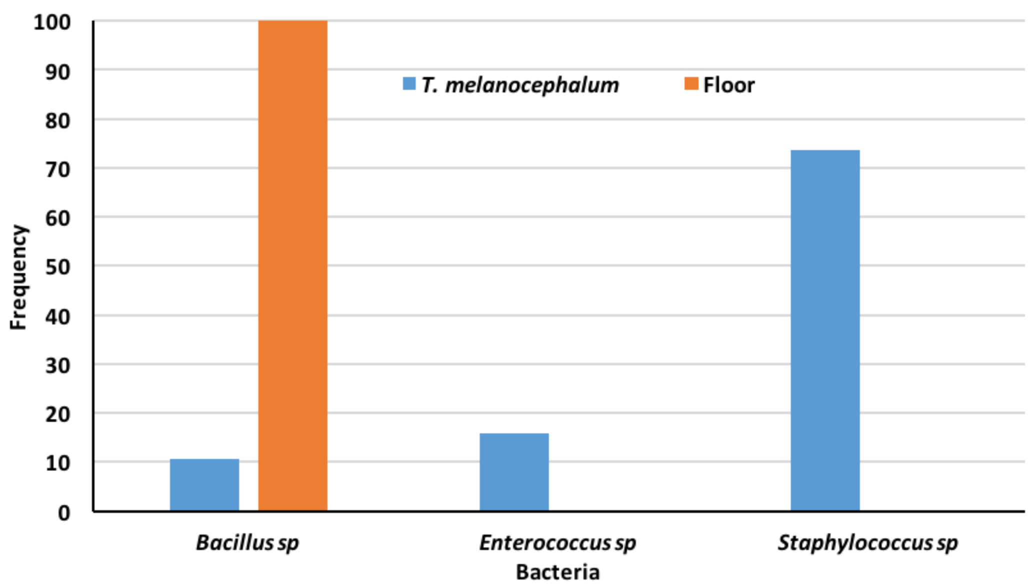

Figure 2. Distribution of bacteria genera in the food service area.

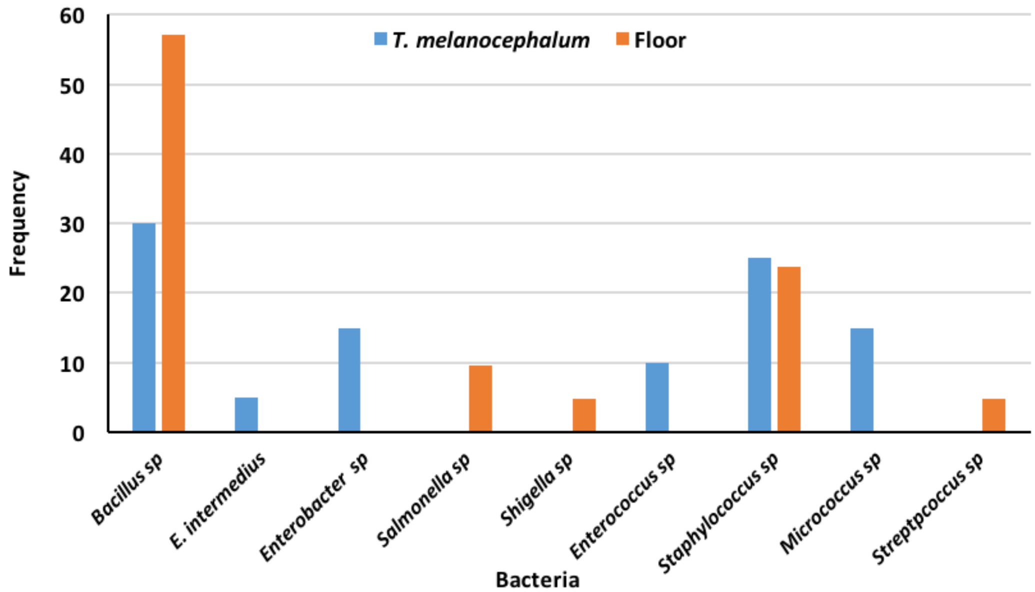

Figure 3. Frequency of Bacteria genera obtained from the ICU.

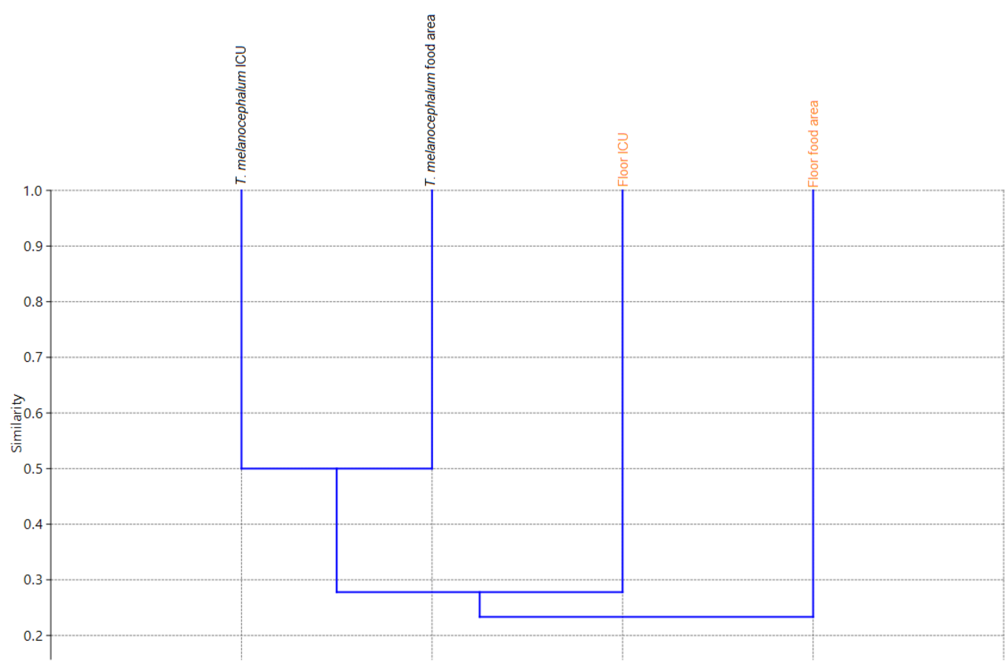

Figure 4. Cluster analysis using the Jaquard's similarity values of samples. Dendrogram obtained from UPGMA (Unweighted Pair Group Method using Arithmetic Mean).

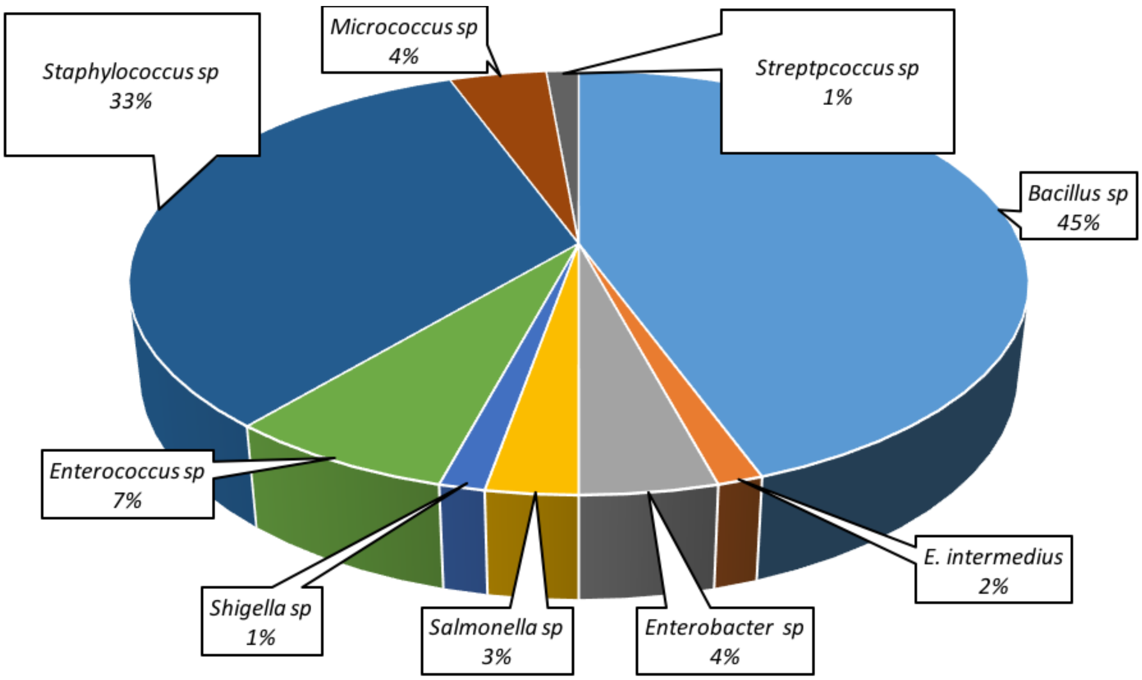

Figure 5. Bacteria associated to nosocomial infections during the sampling at the public hospital.

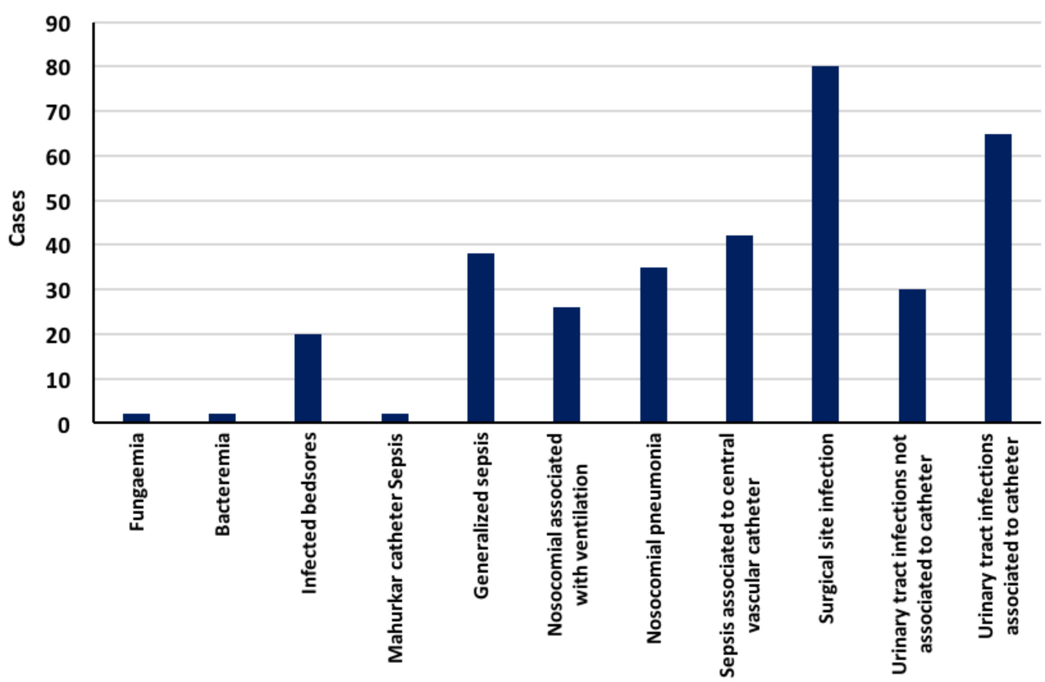

Figure 6. Nosocomial infections reported at the public hospital during the sampling (one year).