

Antibiotic resistance patterns of bacterial strains isolated from *Periplaneta americana* and *Musca domestica* in Tangier, Morocco

Lamiaa Bouamama¹, Antonio Sorlozano², Amin Laglaoui¹, Mariam Lebbadi¹, Ahmed Aarab¹, Jose Gutierrez²

¹Department of Life Sciences, Faculty of Sciences and Techniques, Abdelmalek Essaâdi University, Tangier, Morocco

²Department of Microbiology, School of Medicine, University of Granada, Spain

³Service of Microbiology, University Hospital Virgen de las Nieves, Granada, Spain

Abstract

Background: Flies and cockroaches are two insects in close contact with human beings. They are carriers of human pathogenic bacteria on the external areas of their bodies or in their digestive tracts. This study examines *Periplaneta americana* and *Musca domestica* collected from the residential areas of six districts in Tangier, Morocco.

Methodology: In total, 251 bacteria were isolated from external areas of the participants' bodies and the antimicrobial susceptibility was calculated.

Results: The predominant bacterial species included *Escherichia coli* (17.9%), *Klebsiella* spp. (14.7%), *Providencia* spp. (9.6%), *Staphylococcus* spp. (15.1%) and *Enterococcus* spp. (11.6%). The study showed no difference between the species of bacterial strains from American cockroaches and houseflies. Carbapenems and aminoglycosides were active against 100% of the Gram-negative bacilli isolated in this study. *Staphylococcus* spp. strains were susceptible to linezolid, vancomycin, daptomycin, levofloxacin and cotrimoxazole, and no antibiotic resistance was found in *Enterococcus* spp.

Conclusions: In our setting, although both cockroaches and flies collected from residential areas may be vectors of human pathogenic bacteria, the infections caused by them are easily treatable as a result of the high susceptibility of their bacteria to antibiotics routinely used in the community or in hospitals.

Key words: *Periplaneta americana*, *Musca domestica*, bacteria, antimicrobial resistance

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Introduction

American cockroaches and houseflies are often found in intimate association with human beings and are present in large numbers in and around houses or hospitals and in urban areas and villages with poor sanitation and insalubrious conditions [1,2]. Furthermore, their feeding mechanisms and filthy breeding habits make them the ideal agents for harbouring and transmitting pathogenic bacteria [3-5].

The American cockroach comes in contact with human sewage through sewer systems where they can live, and from there also are able to get into bathrooms and basements [6]. Various bacteria may simply be carried on the insect's cuticle or be ingested and, some time later, regurgitated or excreted. Moreover, several species of bacteria of public health significance have been isolated from, or have passed through, cockroaches (*Periplaneta*

americana) and their digestive tract, such as *Staphylococcus aureus*, *Streptococcus* spp., *Enterobacteriaceae*, *Pseudomonas aeruginosa*, etc. [7-9]. Cockroaches collected in hospitals and households have been found to harbour multi-drug resistant bacteria and hospital cockroaches with drug-resistant *Klebsiella* spp. have been suggested to play a role in the epidemiology of nosocomial infections [7-9]. In addition, a neonatal unit infested with cockroaches [10] suffered an outbreak of nosocomial disease due to extended-spectrum β -lactamase-producing *Klebsiella pneumoniae*.

Houseflies have been suspected to be reservoirs and vectors for pathogens [11-13]. In addition, they have been found to carry multi-drug resistant bacteria in hospital environments and they may play a role in the transmission of human pathogens within hospitals [12-15].

In Morocco, pathogenic bacteria have been isolated from American cockroaches and houseflies collected in urban areas of Tangier [16], a city which records a high demographic growth and also a constant rhythm of urbanization, factors that lead to the emergence of insalubrious and under-developed districts. *Periplaneta americana* and *Musca domestica* are the most common insect species in Morocco because of the favourable environmental and climatic conditions [17]. There are no studies about the susceptibility of human pathogenic bacteria hosted by the common insects, *Periplaneta americana* and *Musca domestica*, found in Moroccan hospitals and households.

Control of these arthropod vectors would allow a reduction of the transmission of these pathogenic bacteria. In addition, in a hospital or clinic setting, a medical professional will not be determining whether or not a particular infection was caused by cockroach or house fly transmission; however, the susceptibility or resistance of the bacteria found on these insects in the geographical areas would be relevant to a patient who presents an infection. Also, once bacterial susceptibility to antibiotics is known, the degree of virulence of the bacteria is likely to be determined and therapeutic possibilities in the case of infection may be found.

In the present study we collected *Periplaneta americana* and *Musca domestica* from residential areas of six districts in Tangier and isolated human pathogenic bacteria from the external surfaces of these insects. Afterward, we determined the susceptibility of the bacterial strains to different antibiotics.

Methods and materials

Insect collection sites

Cockroaches and flies were collected from residential areas of six selected districts of Tangier, between March and October 2006, according to their socio-economic conditions (kind of population, urbanization and social level). The districts were Bendiban (BD), Banimakada (BM), Castilla (CA), Val fleuri (VAL), Place Mozart (PM) and Charf (CF). Banimakada and Bendiban are the popular districts of the city and the most underprivileged and under-equipped due to three main issues: high density of population, inadequate waste disposal and insufficient treatment network. Place Mozart and Charf are benefited by a favourable socio-economic situation, while Val fleuri and Castilla are situated between these two categories of districts.

Collection and identification of cockroaches and flies

Sixty American cockroaches (10 per district) and 600 houseflies (100 per district) were collected from the six selected sites during the period of the study (according to a 1:10 ratio). Flies were caught with sterilized nets near the houses, from garbage heaps and from open defecating grounds in each district and from 9:00 a.m. to 1:00 p.m., when the flies are active.

Cockroaches were caught at night from houses of the selected districts, directly by hand using a gallon container. Trapped cockroaches and flies were placed in sterile test tubes and subsequently taken to the laboratory and stored in a refrigerator at 4°C until the identification and processing for bacteria examination. Identification was made by examining the insect under a low-power microscope and following standard taxonomic keys.

Processing of external body of insect for bacteria isolation

The isolation of bacteria in cockroaches was conducted by adding 5 ml of sterile normal saline solution to a tube containing one cockroach. This was vortexed for 2 minutes to wash off any bacteria from the insect's external body. Vortexing was performed at the lowest possible speed to prevent insects from vomiting and contaminating the contents. Flies were pooled in batches of 10 houseflies each and then individually shaken thoroughly in sterile saline solution (5 ml) for 2 minutes [18]. The suspension washings were then serially diluted and inoculated on MacConkey agar, Chapman agar, and Bile Esculin agar. Plates were incubated for 24 hours at 37°C, and colonies with morphologies characteristic of Gram-negative bacilli, staphylococci, and enterococci were identified by Gram staining and biochemical tests. The species of Gram-negative bacilli were identified using the API 20E system (BIOMÈRIEUX, Marcy-l'Étoile, France), the staphylococci using the API Staph system (BIOMÈRIEUX), and the enterococci using the API 20 Strep system (BIOMÈRIEUX) [19].

Susceptibility determination

Microdilution was performed in a Mueller-Hinton broth, adjusting for Ca⁺⁺ and Mg⁺⁺, following Clinical and Laboratory Standards Institute (CLSI) guidelines [20]. Each antibiotic was dissolved as recommended by the manufacturer. The microdilution procedure for Gram-negative bacilli was performed using the following concentrations (in µg/ml): ampicillin, cefoxitin, ceftazidime, cefepime and amikacin (0.125 to 256); amoxicillin-clavulanate

Table 1. Distribution of Gram-negative bacilli, staphylococci and enterococci isolated from two insects in the six districts of Tangier.

Bacteria	Total	<i>Musca domestica</i>						Total	<i>Periplaneta americana</i>						Total
		Districts							Districts						
		BD	BM	CA	CF	PM	VAL		BD	BM	CA	CF	PM	VAL	
<i>Acinetobacter lwoffii</i>	4	0	1	0	0	0	0	1	2	1	0	0	0	0	3
<i>Alcaligenes spp.</i>	2	0	0	1	0	1	0	2	0	0	0	0	0	0	0
<i>Citrobacter spp.</i>	3	0	0	0	0	0	0	0	1	0	0	0	0	2	3
<i>Enterobacter spp.</i>	21	0	0	2	1	3	1	7	5	3	1	1	3	1	14
<i>Escherichia coli</i>	45	5	3	2	6	9	1	26	2	1	1	8	6	1	19
<i>Klebsiella spp.</i>	37	1	3	1	2	4	2	13	6	9	2	3	2	2	24
<i>Leclercia adecarboxylata</i>	1	0	0	0	0	0	1	1	0	0	0	0	0	0	0
<i>Moellerella wisconsensis</i>	1	0	0	0	1	0	0	1	0	0	0	0	0	0	0
<i>Morganella morganii</i>	2	0	1	0	1	0	0	2	0	0	0	0	0	0	0
<i>Pasteurella spp.</i>	2	0	0	0	0	0	0	0	1	0	0	1	0	0	2
<i>Proteus spp.</i>	17	1	8	0	1	3	1	14	0	2	0	0	1	0	3
<i>Providencia spp.</i>	24	1	5	1	1	2	3	13	2	3	2	1	1	2	11
<i>Salmonella spp.</i>	8	0	1	0	2	0	0	3	2	0	0	2	1	0	5
<i>Serratia spp.</i>	11	0	0	0	2	0	2	4	4	0	1	1	0	1	7
<i>Shigella dysenteriae</i>	4	0	1	0	1	0	0	2	1	0	0	0	0	1	2
<i>Yersinia enterocolitica</i>	2	0	0	0	0	0	0	0	0	1	0	0	1	0	2
<i>Staphylococcus aureus</i>	17	1	0	1	3	3	0	8	5	1	0	0	1	2	9
<i>Staphylococcus coagulase-negative</i>	21	3	0	1	7	1	3	15	3	1	0	0	1	1	6
<i>Enterococcus spp.</i>	29	1	1	2	4	5	0	13	5	6	2	0	2	1	16
Total	251	13	24	11	32	31	14	125	39	28	9	17	19	14	126

Abbreviations: BD: Bendiban; BM: Banimakada; CA: Castilla; CF: Charf; PM: Place Mozart; VAL: Val fleuri.

(0.06/0.03 to 128/64); piperacillin-tazobactam (0.125 to 256, with a fixed concentration of tazobactam of 4 µg/ml); imipenem, ertapenem and meropenem (0.004 to 8); gentamicin and ciprofloxacin (0.06 to 128); cotrimoxazole (0.03/0.594 to 64/1216). The concentration used in the *Staphylococcus* spp. tests are as follows: vancomycin, erythromycin, clindamycin and levofloxacin (0.03 to 64); oxacillin (0.016 to 32); penicillin (0.002 to 4); gentamicin (0.125 to 256); cotrimoxazole (0.03/0.594 to 64/1216); linezolid (0.004 to 8); daptomycin (0.001 to 2). Finally, the following concentrations were used in the *Enterococcus* spp. tests: ampicillin, vancomycin and levofloxacin (0.03 to 64); linezolid (0.004 to 8); daptomycin (0.004 to 8).

The minimum inhibitory concentration (MIC) was defined as the lowest antibiotic concentration to completely inhibit bacterial growth, and the strains

were considered susceptible, intermediate, or resistant according to the recommendations of the CLSI.

The following strains were used as quality control in all procedures, in accordance with CLSI guidelines [20]: *E. coli* ATCC 25922 and *S. aureus* ATCC 29213.

Statistical analysis

The Fisher exact test for $r \times s$ tables was used to compare the clinical categories (in terms of susceptible or resistant) for each antibiotic tested by microdilution between the bacterial strains obtained in flies vs. cockroaches and the strains obtained in the six districts of Tangier. The presence of a difference between the groups with regard to the variable was the alternative hypothesis (H1). A p value ≤ 0.05 was considered significant.

Table 2. Antimicrobial susceptibility of Gram-negative bacilli isolated from American cockroaches and houseflies.

Antibiotic	All Gram-negative bacilli			Gram-negative bacilli in MD			Gram-negative bacilli in PA		
	MIC ₅₀	MIC ₉₀	Susceptibility (%)	MIC ₅₀	MIC ₉₀	Susceptibility (%)	MIC ₅₀	MIC ₉₀	Susceptibility (%)
AMP	32	>256	44	32	>256	42.7	32	256	45.3
AMC	2/1	16/8	83.7	2/1	16/8	83.1	2/1	16/8	84.2
PTZ	0.5/4	1/4	98.9	0.5/4	2/4	97.8	0.5/4	1/4	100
FOX	2	64	82.6	2	64	83.1	4	32	82.1
CAZ	≤0.125	2	98.9	0.25	2	97.8	≤0.125	2	100
FEP	≤0.125	2	99.4	≤0.125	2	98.9	≤0.125	1	100
IMI	0.06	0.25	100	0.06	0.25	100	0.06	0.25	100
ETP	0.008	0.03	100	0.008	0.03	100	0.008	0.03	100
MEM	0.03	0.06	100	0.03	0.06	100	0.03	0.06	100
GM	0.25	0.5	100	0.25	1	100	0.25	1	100
AK	1	4	100	1	4	100	1	4	100
SXT	0.25/4.75	32/608	87.5	0.25/4.75	>64/1216	80.9	0.25/4.75	1/19	93.7
CIP	≤0.06	0.5	97.3	≤0.06	1	97.8	≤0.06	0.5	96.9

Abbreviations: AMP: ampicillin; AMC: amoxicillin-clavulanate; PTZ: piperacillin-tazobactam; FOX: cefoxitin; CAZ: ceftazidime; FEP: cefepime; IMI: imipenem; ETP: ertapenem; MEM: meropenem; GM: gentamicin; AK: amikacin; SXT: cotrimoxazole; CIP: ciprofloxacin; MD: *Musca domestica*; PA: *Periplaneta americana*.

Results

Bacterial isolation

Among the 251 bacteria isolated from American cockroaches and houseflies collected in the six districts of Tangier, 184 (73.3%) belonged to the group of Gram-negative bacilli, 38 (15.1%) to staphylococci, and 29 (11.6%) to enterococci. A total of 125 bacteria were found in *Musca domestica* and 126 bacteria in *Periplaneta americana* (Table 1).

Sixteen genera of the Gram-negative bacilli isolated from the two species of insects were human pathogenic, *i.e.*: *Escherichia coli* (24.5%); *Klebsiella* spp. (20.1%); *Providencia* spp. (13%); *Enterobacter* spp. (11.4%); *Proteus* spp. (9.2%); *Serratia* spp. (6%); *Salmonella* spp. (4.4%); *Shigella dysenteriae* (2.2%); *Acinetobacter lwoffii* (2.2%); *Citrobacter* spp. (1.6%); *Alcaligenes* spp. (1.1%); *Morganella morganii* (1.1%); *Yersinia enterocolitica* (1.1%); *Pasteurella* spp. (1.1%); *Leclercia adecarboxylata* (0.5%) and *Moellerella wisconsensis* (0.5%) (Table 1).

Seventeen *S. aureus* (44.7%) and 21 coagulase-negative staphylococci (55.3%) were isolated from cockroaches and flies. Twenty-nine enterococci were found to be carried by these two species of insects (55.2% of *E. faecium*, 17.2% of *E. durans/hirae*, 13.8% of *E. faecalis*, 10.4% of *E. casseliflavus* and 3.4% of *E. avium*) (Table 1).

Higher numbers of these bacteria were found in Bendiban, Banimakada, Charf and Place Mozart, while the minimum number of bacteria was found in Castilla (Table 1).

The most frequent bacteria isolated from houseflies and American cockroaches coming from all districts of the city were *E. coli*, *Klebsiella* spp. and *Providencia* spp. In addition, *Enterobacter* spp., *Klebsiella* spp. and *Serratia* spp. were more frequently isolated from American cockroaches in comparison with houseflies, while *Proteus* spp. and coagulase-negative staphylococci were more frequently isolated from houseflies, in comparison with American cockroaches. *Citrobacter* spp., *Pasteurella* spp. and *Y. enterocolitica* were present only in *Periplaneta americana*, while *M. morganii* was isolated only from *Musca domestica* (Table 1).

Susceptibility to antimicrobial agents

In general, Gram-negative bacilli isolated from both types of insects were deemed very susceptible to the antibiotics tested. Carbapenems and aminoglycoside antibiotics were found to be active against 100% of Gram-negative bacilli strains. In addition, the following showed excellent activity, although their effectiveness was not 100%: cefepime, ceftazidime, piperacillin-tazobactam, ciprofloxacin, cotrimoxazole, amoxicillin-clavulanate and cefoxitin. Only ampicillin showed low activity against these

Table 3. Percentage of susceptibility of Gram-negative bacilli isolated from two insects collected in the six districts of Tangier.

Antibiotic	In BD	In BM	In CA	In CF	In PM	In VAL
AMP	47.1	32.5	42.9	57.1	48.6	33.3
AMC	82.4	83.7	85.7	82.9	86.5	80.9
PTZ	97.1	97.7	100	100	100	100
FOX	79.4	88.4	78.6	85.7	83.8	71.4
CAZ	100	100	100	97.1	100	95.2
FEP	100	100	100	100	100	95.2
SXT	100	79.1	78.6	88.6	86.5	90.5
CIP	97.1	100	92.8	94.3	97.3	100

Abbreviations: AMP: ampicillin; AMC: amoxicillin-clavulanate; PTZ: piperacillin-tazobactam; FOX: ceftazidime; FEP: cefepime; SXT: cotrimoxazole; CIP: ciprofloxacin; MD: *Musca domestica*; PA: *Periplaneta americana*. BD: Bendiban; BM: Banimakada; CA: Castilla; CF: Charf; PM: Place Mozart; VAL: Val fleuri.

bacterial strains (56% of the strains were found resistant to this antibiotic). Moreover, species of Gram-negative bacilli isolated from houseflies were significantly more resistant to cotrimoxazole than those from cockroaches ($p = 0.001$) (Table 2).

On the other hand, Gram-negative bacilli from Banimakada and Val fleuri were significantly more resistant to ampicillin than those from other districts ($p = 0.021$). Up to 57.1% of bacterial strains from Charf were susceptible to ampicillin, while Gram-negative bacilli from Banimakada and Castilla were significantly more resistant to cotrimoxazole ($p = 0.014$). Cefoxitin showed a difference of 17 percentage points in the susceptibility of strains between the districts of Val fleuri and Banimakada (Table 3).

About 68.4% of *Staphylococcus* spp. showed resistance to penicillin; this percentage was greater in those isolated from cockroaches (73.3%) than in those from flies (65.2%) ($p = 0.011$). *Staphylococcus* spp. strains from cockroaches showed significantly more resistance to gentamicin and erythromycin than those from houseflies ($p = 0.009$ and $p = 0.001$, respectively) (Table 4).

One *S. aureus* isolate showed resistance to oxacillin (MRSA), but 33.3% of coagulase-negative staphylococci were methicillin-resistant.

The species of staphylococci coming from Bendiban, Charf, and Val fleuri were significantly more resistant to penicillin than those from other districts; also, species from Banimakada and Castilla were more resistant to oxacillin (methicillin-resistant staphylococci) than those from the rest of the districts. Staphylococci from Banimakada were also more resistant to gentamicin ($p < 0.05$ in all cases) (Table 5).

Enterococcus spp. from both insects and all the districts of the city were found very susceptible to all antibiotics tested (Table 6).

Discussion

American cockroaches and houseflies have been considered transmitters and spreaders of pathogenic bacteria in hospitals and households or residential areas [21]. In this study, 251 human pathogenic bacteria were isolated from the external bodies of these two species of insects, where we found *Salmonella* spp., *Shigella dysenteriae* and *Yersinia enterocolitica*, bacteria that cause typhoid, paratyphoid fever, dysentery and enterocolitis, among others diseases in humans.

Although the predominant bacteria on cockroaches were *Klebsiella* spp., *E. coli*, and *Enterococcus* spp. and those on flies were *E. coli* and coagulase-negative staphylococci, as other authors have found [9,17,21], no relevant differences were observed in the species of bacterial strains found in *Periplaneta americana* and *Musca domestica*. This can be explained by the fact that there are no differences between the environments in which these insects may be found (human and animal excrement, garbage heaps, open defecating grounds, etc.).

All Gram-negative bacilli isolated in this study were susceptible to carbapenems and aminoglycosides. Carbapenems are exclusively used in hospitals, while the aminoglycoside antibiotics are only preferably used there. Our strains, obtained in non-hospital environments, showed high susceptibility to these groups of antibiotics. These microorganisms were also susceptible to other antibiotics commonly used in the community, such as

Table 4. Antimicrobial susceptibility of staphylococci isolated from American cockroaches and houseflies.

Antibiotic	All staphylococci			Staphylococci in MD			Staphylococci in PA		
	MIC ₅₀	MIC ₉₀	Susceptibility (%)	MIC ₅₀	MIC ₉₀	Susceptibility (%)	MIC ₅₀	MIC ₉₀	Susceptibility (%)
LNZ	0.25	0.5	100	0.25	0.5	100	0.25	0.5	100
VAN	0.25	1	100	0.25	1	100	0.5	1	100
DAP	0.125	0.25	100	0.06	0.25	100	0.125	0.25	100
PEN	0.25	1	31.6	0.25	0.5	34.8	0.25	4	26.7
GM	1	4	94.7	1	4	100	2	8	86.7
ERY	0.25	8	89.5	0.25	0.5	91.3	0.25	8	86.7
CLI	0.25	0.5	92.1	0.25	0.5	91.3	0.25	0.5	93.3
LEV	0.06	0.125	100	0.06	0.25	100	0.06	0.125	100
SXT	0.125/2.375	0.25/4.75	100	0.125/2.375	0.25/4.75	100	0.125/2.375	0.25/4.75	100
OXA	0.25	2	78.9	0.25	2	73.9	0.25	2	86.7

Abbreviations: LNZ: linezolid; VAN: vancomycin; DAP: daptomycin; PEN: penicillin; GM: gentamicin; ERY: erythromycin; CLI: clindamycin; LEV: levofloxacin; SXT: cotrimoxazole; OXA: oxacillin.

Table 5. Percentage of susceptibility of staphylococci isolated from two insects collected in the six districts of Tangier.

Antibiotic	In BD	In BM	In CA	In CF	In PM	In VAL
PEN	25	50	50	30	50	16.7
GM	91.7	50	100	100	100	100
ERY	91.7	100	100	90	83.3	83.3
CLI	100	100	100	90	83.3	83.3
OXA	91.7	50	50	80	83.3	66.7

Abbreviations: PEN: penicillin; GM: gentamicin; ERY: erythromycin; CLI: clindamycin; OXA: oxacillin.

Table 6. Antimicrobial susceptibility of enterococci isolated from American cockroaches and houseflies.

Antibiotic	All enterococci			Enterococci in MD			Enterococci in PA		
	MIC ₅₀	MIC ₉₀	Susceptibility (%)	MIC ₅₀	MIC ₉₀	Susceptibility (%)	MIC ₅₀	MIC ₉₀	Susceptibility (%)
LNZ	0.5	2	100	1	2	100	0.5	1	100
VAN	1	2	100	1	2	100	1	2	100
DAP	0.2	0.5	100	0.125	0.5	100	0.25	0.5	100
AMP	0.5	1	100	1	1	100	0.5	1	100
LEV	1	2	100	1	2	100	1	1	100

Abbreviations: LNZ: linezolid; VAN: vancomycin; DAP: daptomycin; AMP: ampicillin; LEV: levofloxacin.

amoxicillin-clavulanate, ciprofloxacin or cotrimoxazole. There were no notable differences in the susceptibility of strains from flies compared to those of cockroaches. In any case, bacterial strains were not submitted to pressure from a specific antibiotic in their environment, as they did not come from clinical samples of patients infected and subsequently subjected to antibiotic treatment.

Only ampicillin showed a lower activity against these bacterial strains, essentially due to the presence of natural resistance in the bacterial species (*Klebsiella* spp., *Enterobacter* spp., *Providencia* spp., or *Serratia* spp.) [22]. These results contrast with those of Pai *et al.* [9] in which bacterial strains from households showed multi-drug resistance; but coincide with those of Rahuma *et al.* [21] and Elgderi *et al.* [23], both in Libya. In the works of the latter, the *Enterobacteriaceae* isolated from insects (flies or cockroaches) in hospitals were found significantly more resistant to antibiotics than those isolated from the same insects collected from streets.

As with Gram-negative bacilli, all staphylococci were susceptible to antibiotics exclusively used in a hospital environment (linezolid, vancomycin and daptomycin). However, unlike the bacilli, they were also susceptible to levofloxacin and cotrimoxazole. The less active antibiotic against this genus was penicillin, a logical fact due to the existence of the common β -lactamase. In addition, these strains maintained high susceptibility to aminoglycosides, macrolides and lincosamides.

Only one methicillin-resistant *S. aureus* was obtained, whereas 33.3% of coagulase-negative staphylococci were methicillin-resistant. Recent studies have showed an increase in the prevalence of methicillin-resistant *S. aureus* (MRSA) in Moroccan hospitals (from 14.4% in 1996 to 20% in 2004) [24,25]. On the other hand, coagulase-negative staphylococci usually show higher rates of resistance to methicillin than *S. aureus* [26] and this is reflected in our work.

No antibiotic resistance was found in *Enterococcus* spp., but recent studies in Morocco have isolated enterococci species from food samples and said species have shown a higher resistance to erythromycin, ciprofloxacin, and levofloxacin while showing susceptibility to penicillin and gentamicin [27].

Regarding the differences found on the distribution of strains in the different districts, it is remarkable that the highest concentration of resistant bacteria was found in Banimakada and Castilla

districts, the first of which is characterized as the most populated of the six districts studied, while Castilla has a health centre near which we collected the insects.

In conclusion, in our setting, although both the cockroaches and the flies collected from residential areas may be vectors of human pathogens, they can cause infections (wound infections, diarrhoea, pneumonia, etc.) that are easily treatable because of the high susceptibility of their bacteria to antibiotics routinely used in the community or in hospitals.

References

1. Baker LF (1981) Pests in hospitals. J Hosp Infect 2: 5-9.
2. Oothuman P, Jeffery J, Aziz HA, Baker EA, Jegathesan M (1989) Bacterial pathogens isolated from cockroaches transported from pediatric wards in peninsular Malaysia. Trans Roy Soc Trop Med Hyg 83: 133-135.
3. Cloarec A, Rivault C, Fontaine F, Leguyader A (1992) Cockroaches as carriers of bacteria in multi-family dwellings. Epidemiol Infect 109: 483-490.
4. Rivault C, Cloarec A, Leguyader A (1993) Bacterial load of cockroaches in relation to urban environment. Epidemiol Infect 110: 317-25.
5. Graczyk TK, Knight R, Gilman RH, Cranfield MR (2001) The role of non-biting flies in the epidemiology of human infectious diseases. Microb Infect 3: 231-35.
6. Brenner RJ, Koehler PG, Patterson RS (1987) Health implications of cockroach infestations. Infec Med 4: 349-58.
7. Fotedar R, Shriniwas UB, Banerjee U, Samantray JC, Nayar E, Verma A (1991) Nosocomial infections: cockroaches as possible vectors of drug-resistant *Klebsiella*. J Hosp Infect 18: 155-59.
8. Fotedar R, Shriniwas UB, Verma A (1991) Cockroaches (*Blattella germanica*) as carriers of microorganisms of medical importance in hospitals. Epidemiol Infect 107: 181-87.
9. Pai HH, Chen WC, Peng CF (2005) Isolation of bacteria with antibiotic resistance from household cockroaches (*Periplaneta americana* and *Blattella germanica*). Acta Trop 93: 259-65.
10. Cotton MF, Wasserman E, Pieper CH, Theron DC, van Tubbergh D, Campbell G, Fang FC, Barnes J (2000) Invasive disease due to extended spectrum beta-lactamase-producing *Klebsiella pneumoniae* in a neonatal unit: the possible role of cockroaches. J Hosp Infect 44: 13-17.
11. Echeverria P, Harrison BA, Tirapatk C, McFarland A (1983) Flies as a source of enteric pathogens in a rural village in Thailand. Appl Environ Microbiol 46: 32-36.
12. Fotedar R, Banerjee U, Samantray JC, Shriniwas SS (1992) Vector potential of the hospital house flies with special reference to *Klebsiella* species. Epidemiol Infect 109: 143-47.
13. Fotedar R, Banerjee U, Shriniwas SS, Verma A (1992) The house fly (*Musca domestica*) as a carrier of pathogenic micro-organisms in a hospital environment. J Hosp Infect 20: 209-15.
14. Khalil K, Lindblom GB, Mazhar K, Kaijsher B (1994) Flies and water as reservoirs for bacterial enteropathogens in urban and rural areas in and around Lahore, Pakistan. Epidemiol Infect 113: 435-44.

15. Sramova H, Daniel M, Absolonova V, Dedicova D, Jedlickova Z, Lhotova H, Petras P, Subertova V (1992) Epidemiological role of arthropods detectable in health facilities. *J Hosp Infect* 20: 281-92.
16. Bouamama L, Lebbadi M, Aarab A (2007) Bacteriological analysis of *Periplaneta americana* L. (Diptera; Blattellidae) and *Musca domestica* L. (Diptera; Muscidae) in ten districts of Tangier, Morocco. *Afr J Biotechnol* 6: 2038-42.
17. Boulesteix G, Le Dantec P, Chevalier B, Dieng M, Niang B, Diatta B (2005) Role of *Musca domestica* in the transmission of multiresistant bacteria in the centers of intensive care setting in sub-Saharan Africa. *Ann Fr Anesth Reanim* 24 : 361-65.
18. Fotedar R (2001) Vector potential of houseflies (*Musca domestica*) in the transmission of *Vibrio cholerae* in India. *Acta Trop* 78: 31-34.
19. Murray PR (1999) Manual of Clinical Microbiology. Seventh edition. ASM Press, Washington, DC.
20. Clinical and Laboratory Standards Institute (2007) Performance standards for antimicrobial susceptibility testing; seventeenth informational supplement. CLSI publication M100-S17. Wayne, Pa.
21. Rahuma N, Ghenghesh KS, Ben Aissa R, Elamaari A (2005) Carriage by the housefly (*Musca domestica*) of multiple-antibiotic-resistant bacteria that are potentially pathogenic to humans, in hospital and other urban environments in Misurata, Libya. *Ann Trop Med Parasitol* 99: 795-802.
22. Navarro Risueño F, Miró Cardona E, Mirelis Otero B (2002) Interpretive reading of the antibiogram of enterobacteria. *Enferm Infecc Microbiol Clin* 20: 225-34.
23. Elgderi RM, Ghenghesh KS, Berbash N (2005) Carriage by the german cockroach (*Blattella germanica*) of multiple-antibiotic-resistant bacteria that are potentially pathogenic to humans, in hospitals and households in Tripoli, Libya. *Ann Trop Med Parasitol* 100: 55-62.
24. Kesah C, Ben Redjeb S, Odugbemi TO, Boye CSB, Dosso M, Ndinya Achola JO, Koulla-Shiro S, Benbachir M, Rahal K, Borg M (2003) Prevalence of methicillin-resistant *Staphylococcus aureus* in eight African hospitals and Malta. *Clin Microbiol Infect* 9: 153-56.
25. Borg MA, Kraker M, Scicluna E, Bruinsma NS, Tiemersma E, Monen J, Grundmann H (2007) Prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) in invasive isolates from southern and eastern Mediterranean countries. *J Antimicrob Chemother* 60: 1310-15.
26. Stefani S, Varaldo PE (2003) Epidemiology of methicillin-resistant staphylococci in Europe. *Clin Microbiol Infect* 9: 1179-86.
27. Valenzuela AS, Ben Omar N, Abriouel H, Lopez RL, Ortega E, Cañamero MM, Galvez A (2008) Risk factors in enterococci isolated from foods in Morocco: Determination of antimicrobial resistance and incidence of virulence traits. *Food Chem Toxicol* 46: 2648-52.

Corresponding author

Dr. Jose Gutierrez
Departamento de Microbiología
Facultad de Medicina
Avda de Madrid 11
E-18012 Granada
Spain
Email: josegf@ugr.es

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