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Faecal carriage of extended-spectrum β-lactamase-producing *Escherichia coli*: prevalence, risk factors and molecular epidemiology

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***Objectives*: The aim of this study was to investigate the epidemiology of faecal carriage of extended-spectrum β-lactamase (ESBL)-producing *Escherichia coli* in the community.**

***Patients and methods*: Faecal carriage with ESBL-producing *E. coli* was studied in 53 outpatients with urinary tract infection (UTI) due to these organisms, 73 household members, 32 non-household relatives and 54 unrelated patients. Clonal relatedness of the isolates was investigated using repetitive extragenic palindromic-PCR and PFGE, and ESBLs were characterized by PCR and sequencing. Multivariate analysis was performed to investigate risk factors for faecal carriage.**

***Results*: The prevalence of faecal carriage was 67.9% in patients with UTI, 27.4% in household members, 15.6% in non-household relatives and 7.4% in unrelated patients. Being a relative of a patient with UTI was independently associated with an increased risk of being a carrier. Among the relatives, multivariate analysis showed that those eating their main meal outside their own home >15 days during the previous month were less likely to be faecal carriers (OR = 0.2; 95% CI: 0.06–0.6; *P* = 0.007). The faecal isolates of patients with UTI were CTX-M-producers in 66.6% and SHV-producers in 33.3% of the cases, while the percentages for other population groups were 40% to 55.5% and 50% to 75%, respectively. Of the 19 families with >1 carrier member, 8 families had 2 members who shared clonally related isolates, 8 families had 2 members carrying different clones producing the same enzymes and there were 3 families where all members had different enzyme-producing clones.**

***Conclusions*: Our results suggest that both acquisition from a common source and person-to-person transmission might contribute to ESBL dissemination.**

Keywords: urinary tract infections, antimicrobial resistance, community-acquired infections, cephalosporin resistance

Introduction

Worldwide dissemination of plasmid-borne extended-spectrum β-lactamases (ESBLs) is a public health concern since community-acquired infections caused by ESBL-producing *Escherichia coli* (among other Enterobacteriaceae) are becoming increasingly frequent.[1] This phenomenon is mainly related to the surprisingly rapid spread of one specific family of ESBLs, the CTX-M enzymes, associated with highly efficient mobile genetic elements found in different *E. coli* clones.[2] The intercontinental spread of particular *E. coli* clones harbouring CTX-M-15 has also been described.[3] Very little is known about the epidemiology of the ESBLs from the SHV family within the community setting, even though, in some areas, SHV-producing *E. coli* have been found as a cause of community-acquired infections.[4,5]

Although a few studies have investigated the prevalence of faecal carriage with ESBL-producing *E. coli* in the community setting,[6–14] little is known of the frequency of carriage in patients with community-acquired urinary tract infections (UTIs), or in their family members, and of the risk factors involved in faecal carriage. Even though the food supply is a suspected source of generic antimicrobial-resistant *E. coli*,[15] recent data

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indicate that other variables, including foreign travel and household and community exposure, may be more important risk factors.[16] To the best of our knowledge, no specific studies have been performed for ESBL-producing *E. coli*.

The objectives of our study were to investigate the prevalence and risk factors of faecal carriage with ESBL-producing *E. coli* in different groups of persons in the community and the impact of cohabitation on the spread of these pathogens.

Patients and methods

The study was performed between June 2005 and September 2006 in the Health Area of the Virgen Macarena Hospital, serving a population of some 450 000 people and located in the north of Seville, Spain. A significant increase in the number of ESBL-producing *E. coli* has been noted since 2000 in our area,[17] which constituted 4.8% of all *E. coli* isolated from outpatients in 2003.[4] The vast majority of these isolates were clonally unrelated.[18] With regard to the type of ESBLs produced by these isolates, ~2/3 produced CTX-M enzymes (CTX-M-14 being the most frequent) and 1/3 SHV (mainly SHV-12).[18]

Faecal carriage of ESBL-producing *E. coli* was studied in three groups of adults (age >14 years) using a cross-sectional survey: group A patients included all patients diagnosed with community-acquired UTI [defined as the presence of symptoms relating to the urinary tract, pyuria (≥10 leucocytes per high-power field) and a positive urine culture (≥10^5 cfu/mL) in patients seen in the emergency department or primary care centres of our health area, with no history of hospital admission in the preceding month[4,5,19]], caused by ESBL-producing *E. coli* during the study period; group B included all available relatives of group A patients and was divided into two subgroups: the B1 subgroup ('household members'), which included all available members of the household of group A patients, and the B2 subgroup ('non-household relatives'), which included all available next of kin adults related to group A patients but not living in the same household; and group C ('unrelated controls') included unrelated persons, one per patient in group A, randomly chosen from those attended to in the hospital's Emergency Department during the week after detecting the group A index patient. We chose this latter group to provide some frequency matching for the underlying conditions and previous hospital admissions, since our previous studies led us to expect that such variables would be frequent among group A patients. Potential participants were excluded if ESBL-producing *E. coli* had previously been isolated from them, according to the Microbiology records.

Faecal carriage was studied by performing a rectal swab on all participants who agreed to participate. In order to be considered suitable for study, the rectal swab had to contain faecal material. Rectal swabs were obtained from all participants in <7 days after the diagnosis of UTI in the group A patient. Patients with an ESBL-producing *E. coli* isolate on a rectal swab were considered carriers.

The following data were recorded for each participant: age; gender; type of relationship with patient (whether partner or not); any chronic underlying disease; severity of co-morbidity (according to the Charlson index[20]); previous healthcare contact (admission to any acute care hospital or long-term care facility, dialysis, specialized home care or intravenous therapy during the previous 3 months); invasive procedures (urinary catheter, vascular catheter, surgery and endoscopic procedure) during the previous week; receipt of antimicrobials, proton pump inhibitors or other antacids during the previous 2 months; contact with pets; travel abroad during previous 2 months; dietary habits; contact with raw meat products; cooking and consumption of meat products; and how frequently the main meal (that is, the most copious meal of the day) had been eaten outside their own home during the previous month. All participants chose lunch as their main meal. This latter was recorded as a categorical variable and included the following strata: <7, 7–15 and >15 days during the previous month. The crude OR was similar for <7 and 7–15 days, so that we recoded it as a dichotomous variable (≤15 and >15 days).

The study was approved by the local Ethics Committee. Written informed consent was obtained from all participants.

Microbiological studies

The isolates studied included urine isolates from group A patients and faecal isolates from all participants. Rectal swabs were spread onto 2 MacConkey agar plates, one supplemented with 2 mg/L cefotaxime and the other with 2 mg/L ceftazidime (for use within 5 days of preparation), using a blood agar plate (BA) as a control. Only samples yielding enteric flora on BA were considered suitable. When the culture showed positive on selective media, at least three colonies plus each distinct morphotype were selected for subsequent characterization. Bacterial identification was performed using biochemical tests (oxidase, triple sugar iron, urea and indole). Screening for ESBL production was carried out with 30 μg cefotaxime and ceftazidime discs (Oxoid Ltd, Basingstoke, UK), and with and without 10 μg clavulanic acid, according to the CLSI guidelines.[21]

Isoelectric focusing[22] and PCR assay were used for preliminary characterization of the β-lactamase produced. PCR conditions and previously described group-specific primers (SHV,[23] CTX-M-9,[24] CTX-M-1[25] and TEM[23]) were used to amplify the *bla* genes, depending on the results of isoelectric focusing. Amplicons were sequenced by an ABI PRISM 377 sequencer (Applied Biosystems, Perkin Elmer, Foster City, CA, USA). Clonal relationship was determined for all isolates of a given family using repetitive extragenic palindromic (REP)-PCR.[26] To confirm clonality, all isolates whose REP-PCR profile was indistinguishable were further assessed by PFGE analysis using *Xba*I.[27] Isolates were defined as representing the same clone according to the Tenover's criteria.[28]

Statistical analysis

The prevalence of carriage was calculated as the percentage of carriers among participants of each group. To investigate the risk factors associated with faecal carriage within groups, carriers were compared with non-carriers in terms of exposure to the different variables studied. For independent samples, categorical variables were compared using the χ^2 or Fisher exact test, and continuous variables using Student's *t*-test or the Mann–Whitney *U*-test, as appropriate. Odds ratio (OR) and 95% confidence intervals (CIs) were calculated. The adjusted OR was calculated using conditional logistic regression (the group A patient served as the matching variable), except for analysing the risk factors for carriage in the group A patients, where logistic regression was used. Data were analysed using the STATA 9.2 software package.

Results

During the study period, there were 54 cases of community-acquired UTIs caused by ESBL-producing *E. coli* (group A patients); the rectal swab was unsuitable in one of them and was excluded. Group B included 105 relatives in all; the B1

Table 1. Characteristics of participants; data are expressed as the number of patients (percentages) except where specified

	Group A ($n = 53$)	Subgroup B1 ($n = 73$)	Subgroup B2 ($n = 32$)	Group C ($n = 54$)
Median age, years (interquartile range)	69 (52–75)	43 (23–63)	42 (33–49)	68 (55–79)
Female gender	37 (70)	41 (56)	22 (69)	31 (57)
Previous hospital admission	19 (36)	6 (8)	4 (13)	22 (41)
Chronic underlying disease	42 (79)	22 (30)	10 (31)	44 (81)
Urinary catheter	11 (21)	1 (1)	0	5 (9)
Previous antimicrobial treatment	38 (72)	8 (11)	2 (6)	6 (11)

subgroup comprised 73 household members of 40 group A patients; the remaining patients either lived alone (7) or had only one household member, who could not be included for various reasons. For 32 group A patients (80%), the whole household was studied. In total, 81% of adult household members took part in the study (73/90). Non-participants and participants had similar age and gender distributions. Subgroup B2 comprised 32 next of kin not living in the same household as 20 group A patients; all of them reported visiting the home of their index group A patient at least twice a week. Finally, group C included 54 unrelated persons. Participant characteristics are shown in Table 1. ESBL-producing *E. coli* had not been previously isolated from any of the participants.

Prevalence of faecal carriage and risk factors

The prevalence (95% CI) of faecal carriage was 67.9% (51.7–84.0) in group A patients (36/53), 23.8% (15.6–31.9) in group B (25/105) and 7.4% (0.5–14.3) in group C (4/54). The prevalence of carriage was significantly more frequent in group A patients than in all other groups ($P < 0.01$ for all comparisons). Also, prevalence among group B was higher than in group C ($P = 0.01$). Regarding subgroups, prevalence was 27.4% (17.1% to 37.6%) in subgroup B1 (20/73) and 15.6% (3.1% to 28.0%) in subgroup B2 (5/32); the difference was not statistically significant ($P = 0.1$).

To investigate the risk factors for faecal carriage, we carried out separate analyses on group A patients and on their relatives (group B); the number of carriers in group C patients was too low for a specific analysis to be made. The univariate analysis of risk factors for faecal carriage in group A patients is shown in Table 2. Noteworthy findings were that the prevalence of carriage was similar for males and females (69% and 68%, respectively) and that receipt of antimicrobial therapy was not protective for carriage, although the low number of patients precluded the analysis of specific antimicrobials. For the multivariate analysis, diabetes mellitus, chronic pulmonary disease and urinary catheter (all with univariate P values <0.1) were included in the model; in the end, only diabetes mellitus was found to be independently associated with an increased risk of faecal carriage (OR = 12.8; 95% CI: 1.5–107.1; $P = 0.005$). Adding previous and present urinary antimicrobial treatment did not significantly affect the results. The univariate analysis of risk factors for faecal carriage in group B is shown in Table 3. Being a relative of a group A carrier patient was not associated with an increased risk of carriage; also, previous healthcare contact was not associated with increased risk of carriage. There were no significant differences in other variables such as contact with raw meat products, cooking or travel abroad. The variables introduced in the multivariate conditional regression analysis were frequently eating the main meal outside the home, female gender (both with a univariate P value <0.1) and household condition; only the first was independently associated with carriage. Controlling for household condition, people who had eaten their main meal outside their home >15 days in the previous month were less likely to be faecal carriers (OR = 0.2; 95% CI: 0.06–0.6; $P = 0.007$); 40/43 of the participants who reported having eaten their main meal outside the home >15 days in the previous month had eaten it in a restaurant or somewhere similar.

To determine whether being a relative of a group A patient was associated with increased risk of carriage, and controlling for other variables, we performed a multivariate analysis using conditional regression that included all individuals from groups B and C. We employed different models, all of which included the variable 'being a relative', and adding various potential confounders (underlying diseases, age, sex, previous antimicrobial use and dietary habits); in each model, the variables were added individually and in pairs. None of these variables significantly changed the association between being a relative of a group A patient and faecal carriage; therefore, being a relative of a group A patient increased the risk of carriage, even when controlling for underlying disease, age, sex, previous antimicrobial use and dietary habits; OR (95% CI) for household members and non-household members were 16.1 (4.1–63.6) and 6.2 (1.2–29.8), respectively.

ESBL characterization and clonal relatedness of ESBL-producing E. coli

Nine people (four from group A and five from subgroup B1) carried more than one different strain of ESBL-producing *E. coli*. Table 4 shows the types of ESBL found in the different groups. A CTX-M-producing isolate was carried by 66.6%, 52% and 50% of carriers in groups A, B and C, respectively, with an SHV-producing isolate being carried by 33.3%, 52% and 75%, respectively; the prevalence of SHV in faecal isolates was somehow more frequent in isolates obtained from patients in groups B and C than in group A, although the difference was not statistically significant [14/29 (48.3%) versus 12/36 (33.3%), $P = 0.1$]. The CTX-M enzymes belonged to the CTX-M-9 group (54 CTX-M-14 and 11 CTX-M-9) and the CTX-M-1 group (6 CTX-M-32 and 5 CTX-M-15); 41/49 SHV enzymes were SHV-12. No TEM ESBL types were found.

Table 2. Univariate analysis of risk factors for faecal carriage with ESBL-producing *E. coli* in group A patients (patients with UTIs caused by these organisms); data are expressed as the number of cases (percentage) except where specified

	Carriers ($n = 36$)	Non-carriers ($n = 17$)	P[a]
Median age, years (interquartile range)	68 (48–77)	70 (62–75)	0.6
Female gender	25 (69)	12 (71)	0.9
Previous healthcare contact	16 (44)	5 (29)	0.2
Previous hospital admission	15 (42)	4 (24)	0.1
Long-term care facility	0	1 (6)	0.3[b]
Charlson index >1	16 (44)	4 (24)	0.1
Diabetes mellitus	16 (44)	1 (6)	0.005
Chronic pulmonary disease	2 (6)	4 (24)	0.07[b]
Neoplasia	3 (8)	1 (6)	1.0[b]
Chronic renal insufficiency	3 (8)	1 (6)	1.0[b]
Recurrent UTI	23 (64)	12 (71)	0.6
Urinary catheter	10 (28)	1 (6)	0.08[b]
Previous antimicrobial treatment	27 (75)	11 (65)	0.5[b]
Cephalosporins	8 (22)	2 (12)	0.4[b]
Fluoroquinolones	12 (33)	4 (24)	0.4
Trimethoprim/sulfamethoxazole	6 (17)	4 (24)	0.7[b]
Present active antimicrobial treatment	26 (72)	14 (82)	0.6[b]
Antacid therapy	24 (67)	11 (65)	0.8
Contact with pet	14 (39)	7 (41)	0.8
Consumption of water from public water supply	20 (56)	13 (76)	0.1
Main meal outside home >15 days previous month	9 (25)	4 (24)	1.0[b]
Consumption of chicken, mean days previous month (SD)	6.8 (5.6)	7.2 (5.3)	0.8[c]
Consumption of pork, mean days previous month (SD)	3.4 (3.7)	2.5 (2.4)	0.6[c]
Consumption of beef, mean days previous month (SD)	3.4 (3.5)	4.4 (3.4)	0.2[c]

[a]All P tests were performed using χ^2, except where indicated, [b]Fisher test and [c]Mann–Whitney U-test.

ESBL-producing *E. coli* isolates found in the urine of group A patients were all unrelated. Of the 36 group A patients who were also faecal carriers, their faecal isolate was clonally related to their urine isolate in 27 (75%) cases (51% of the whole series of group A patients).

Analysis of ESBL-producing E. coli carriage within families studied

Fifty-one group A patients had at least one family member studied; the median number of relatives studied per group A patient was 2 (range, 1–5). Household members were studied in 40 group A patients; in 18 of these group A patients (45%), one or more household member was detected as a carrier. Relatives not living in the same household were studied in 20 group A patients; for 5 of these group A patients (25%), one or more relatives not living in the same household were carriers. The difference was not statistically significant ($P = 0.1$).

Grouped together, of the 51 group A patients with a relative (whether members of their household or not) studied, we found at least one carrier relative in 21 of them (41.2%). We analysed the features of group A patients associated with having at least one carrier relative; crude analysis showed an increased risk of finding a carrier relative in males and in those with urinary catheter; the probability of finding a relative carrier was also higher when the number of relatives studied was >2 compared with ≤2 (12/18, 66.6% versus 9/33, 27.2%, $P = 0.006$). After controlling for having >2 relatives studied, being male (OR = 11.0, 95% CI: 1.7–68.7) and having a urinary catheter (OR = 8.4, 95% CI: 1.1–64.7) were still associated with an increased probability of finding a relative carrier.

We analysed the types of ESBL and the clonal relatedness of the ESBL-producing *E. coli* isolates within the 21 families where at least two carriers were detected. Two families had to be excluded because some of the isolates were not available for typing. As a result, we analysed clonal relatedness of isolates within 19 families (Figure 1). Clonally related isolates of ESBL-producing *E. coli* were obtained from >1 member in 8 (42.1%) families (8 group A patients plus one of their relatives in 7 of them, and 2 in one of them); in every case, clonally related isolates harboured the same ESBLs: SHV-12 in 4 families, CTX-M-14 in 2, CTX-M-9 in 1 and CTX-M-15 in 1. Seven relatives lived in the same household as the group A patients and two did not. It was noted that 6 relatives of 5 group A patients carried an isolate that was clonally related to that causing the UTI in their group A family member. In a further 8 families, isolates from all members were clonally unrelated, although at least 2 members of each family harboured the same ESBL (SHV-12 in 5 families and CTX-M-14 in 3); 6 of these relatives lived in the same household as the group A patient; 3 did not. Finally, in only 3 families did all members carry unrelated *E. coli* producing different ESBLs. We noted that 8/10 group A patients who were faecal carriers and had urinary catheterization also had a relative

Table 3. Univariate analysis of risk factors for faecal carriage with ESBL-producing *E. coli* in members of B group; data are expressed as the number of cases (percentage) except where specified

	Carriers ($n = 25$)	Non-carriers ($n = 80$)	P[a]
Median age, years (interquartile range)	48 (24–71)	40 (28–53)	0.4
Female gender	10 (76)	44 (55)	0.06
Household member of group A patient	20 (80)	53 (66)	0.1
Relative of a carrier group A patient	19 (76)	59 (76)	0.9
Previous healthcare contact	4 (16)	8 (10)	0.4[b]
Previous hospital admission	3 (12)	7 (9)	0.6[b]
Long-term care facility	1 (4)	1 (3)	0.4[b]
Charlson index >1	5 (20)	12 (15)	0.5
Diabetes mellitus	2 (8)	4 (5)	0.6[b]
Chronic pulmonary disease	0	3 (4)	1.0[b]
Neoplasia	1 (4)	3 (4)	1.0[b]
Chronic renal insufficiency	0	0	—
Recurrent UTI	0	2 (3)	1.0[b]
Urinary catheter	0	1 (1)	1.0[b]
Previous antimicrobial treatment	2 (8)	8 (10)	1.0[b]
Antacid therapy	9 (36)	30 (38)	0.8
Contact with pet	11 (44)	22 (38)	0.1
Consumption of water from public water supply	22 (88)	65 (81)	0.4
Main meal outside home >15 days previous month	4 (16)	39 (49)	0.004
Consumption of chicken, mean days previous month (SD)	8.8 (3.7)	8.3 (5.8)	0.2[c]
Consumption of pork, mean days previous month (SD)	5.8 (3.9)	4.8 (3.4)	0.2[c]
Consumption of beef, mean days previous month (SD)	3.8 (3.9)	3.2 (4.0)	0.3[c]

[a]All P tests were performed using χ^2 except where indicated, [b]Fisher test and [c]Mann–Whitney U-test.

Table 4. Types of ESBLs produced by *E. coli*; some isolates produced more than one type of ESBL and some patients carried more than one different strain of ESBL-producing *E. coli*; data are presented as the number of isolates with feature specified (percentage of patients)

	Group A				
Type of ESBL	urine isolates ($n = 53$)	faecal isolates ($n = 36$)	Subgroup B1 faecal isolates ($n = 20$)	Subgroup B2 faecal isolates ($n = 5$)	Group C faecal isolates ($n = 4$)
CTX-M	34 (64.1)	24 (66.6)	11 (55.5)	2 (40)	2 (50)
SHV	18 (33.9)	12 (33.3)	10 (50)	3 (60)	3 (75)
Specific ESBL					
CTX-M-14	23	17	9	2	3
CTX-M-9	5	4	2	—	—
CTX-M-32	4	1	1	—	—
CTX-M-15	2	2	1	—	—
SHV-12	14	11	11	3	2
unspecified SHV	4	1	3	—	—
Not sequenced	2	2	—	—	—

n, number of patients.

sharing either a clonally related strain (2) or unrelated *E. coli* harbouring the same ESBL (6).

Among the participants, 28 sexual partners were studied. In 7 couples, both partners were carriers (clonally related isolates in 2 and clonally unrelated isolates in 5, of which 3 produced the same ESBL and the remaining 2 a different ESBL); in 15 couples, only one partner was a carrier; finally, in 6 couples, neither partner was a carrier.

Discussion

The rapid and extensive spread of ESBL-producing *E. coli* in the community setting is an intriguing phenomenon in contemporary epidemiology,[1] whose main components (reservoirs and means of transmission) have not been explained. While several indirect findings suggest that the acquisition of ESBL-harbouring isolates may be mediated by food,[29–31] little is known of the dynamics of

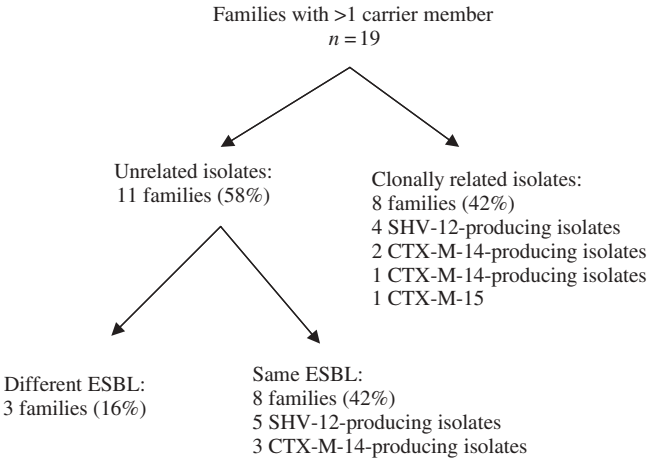

Figure 1. Molecular epidemiology of ESBL-producing *E. coli* within the 19 families with more than one carrier member.

how these enzymes spread among humans, a problem which is of interest for control purposes.

The prevalence of faecal carriage of ESBL-producing Enterobacteriaceae, both in outpatients (studied mostly from faecal samples of patients with acute diarrhoea) and healthy persons, has been investigated in several studies.[6–14] In all of these, *E. coli* accounted for the vast majority of isolates. In studies from three different areas in Spain (Madrid, Barcelona and Zaragoza), the prevalence of faecal carriage has significantly increased in recent years, reaching rates of between 5.5% and 8.1% during 2002 and 2004.[6,8,11] The prevalence of faecal carriage was 1.4% in York (UK) in 2003,[7] 2.4% in Lebanon[9] and 7% in India.[10] A higher rate was found in Saudi Arabia (15.4%).[12] Ben-Ami *et al.*[14] found that 10.8% of the patients studied on admission to hospital in Israel were faecal carriers. A significant increase in the prevalence of faecal carriage (from 0.1% in 2001 to 1.7% in 2005) was also observed in healthy children in Bolivia and Peru.[13] The prevalence of faecal carriage found among patients attending the Emergency Department in our study (7.4%) was similar to that found in other Spanish studies. Interestingly, the prevalence of faecal carriage was found to be significantly higher in relatives of patients diagnosed with community-acquired UTI caused by ESBL-producing *E. coli* (23.8%), and being a relative of such patients was independently associated with an increased risk of being a carrier (the risk was higher in household members than in non-household relatives), indicating acquisition from a common source or person-to-person transmission within families. In fact, we found at least one carrier relative in 45% of the infected patients.

The horizontal spread of ESBLs and ESBL-producing *E. coli* has been observed in community outbreaks caused by enteropathogens.[31,32] To the best of our knowledge, there are no previous studies investigating the risk factors associated with faecal carriage in healthy individuals in non-outbreak situations. Ben-Ami *et al.*[14] studied patients on admission to hospital and found that poor functional status, current antibiotic use, chronic renal insufficiency, liver disease and use of histamine$_2$-receptor antagonists were the independent risk factors for faecal carriage. However, that study included a wide range of ESBL-producing Enterobacteriaceae (only 17/31 isolates were *E. coli*), and a third of their patients were admitted from a long-term care facility. Such differences in methodology and population preclude any comparison with our data. We investigated the risk factors for faecal carriage among relatives of patients with community-acquired UTI caused by ESBL-producing *E. coli* and found that frequently eating the main meal outside the home was a protective variable, suggesting that home food may be a source of these isolates. Most relatives carrying ESBL-producing *E. coli* had no previous healthcare contact, confirming that acquisition of ESBL-producing *E. coli* may be unrelated to healthcare centres in many cases. On the other hand, we found that male gender and urinary catheter use in patients with UTI due to ESBL-producing *E. coli* were associated with an increased risk of having a carrier relative; urinary catheterization may increase the risk of person-to-person transmission of urinary isolates. Taken together, the data suggest that both common acquisition and person-to-person transmission may explain the increased risk of carriage in relatives of infected patients. It is worth noting that previous antimicrobial use, which has invariably been found to be a risk factor in community-onset infections caused by ESBL-producing *E. coli*,[4,5,33,34] was not a risk factor in faecal carriage. This further reinforces the idea that antimicrobials act by selecting ESBL-producing isolates in previously colonized persons.

The results of the molecular investigation support the hypothesis that both person-to-person transmission and acquisition from a common source contribute to the spread of ESBL-producing *E. coli* in the community. In families with more than one carrier member, the proportion of members sharing clonally related strains or clonally unrelated strains harbouring the same ESBL was the same: 42.1% in each case. Sexual transmission of extraintestinal pathogenic *E. coli* has been well demonstrated,[35,36] although in the eight families where more than one member shared clonally related ESBL-producing *E. coli*, the strain was shared by sexual partners in only two families. These data indicate that the epidemiology of colonization with ESBL-producing *E. coli* is complex and that explaining the dynamics of acquisition and transmission requires complex longitudinal studies.

An outstanding finding of our study is the fact that SHV-producing isolates are frequent colonizers, and we fear that, although CTX-M enzymes presently predominate in ESBL-producing *E. coli* in the community, SHV enzymes could also spread in the community setting. We have previously found that plasmids harbouring SHV-12, extracted from isolates causing community-onset infection in our area, were mostly unrelated, while those harbouring CTX-M-14 were related.[18] Further studies are required to obtain more detailed knowledge of the epidemiology of the SHV enzymes in the community.

Our data demonstrate that ESBL-producing *E. coli* causing community-acquired UTI derives from the host's intestinal flora in at least half the patients, supporting the faecal-urethral hypothesis only in these cases, a similar situation to that shown for generic extraintestinal pathogenic *E. coli* in men with febrile UTI.[37] Interestingly, with regard to intestinal colonization by urine strains, there were no differences between men and women and, indeed, the prevalence of intestinal colonization in women with UTI was lower than that has previously been found for generic *E. coli*.[38,39] The investigation of virulence factors would help to better understand the pathogenic behaviour of these isolates.

We found that diabetes mellitus was associated with an increased risk of carriage in UTI patients. We cannot rule out that diabetes mellitus is really a surrogate marker for other unmeasured variables associated with diabetes.[40] However, the relationship

between diabetes mellitus and ESBL requires further study since it has also previously been demonstrated to be a risk factor in community-onset infection due to ESBL- producing *E. coli*.[4,33,34]

Our study has several limitations. First, we have neither studied the molecular relationship of plasmids harbouring ESBLs, which might increase our understanding of the dynamics of the spread of ESBL, nor the presence of virulence factors in the isolates; these investigations are in progress. Secondly, the extent of person-to-person contact among relatives was not investigated in depth, and consequently, we were unable to evaluate whether different types of contact are associated with different risks of transmission; also, we did not collect data about certain household conditions (such as the sharing of toilets and hygiene habits), which could be determinants of faecal carriage. Thirdly, the study may not have enough statistical power to detect some risk factors due to the low number of colonized persons in some subgroups. Finally, not every relative was studied.

In summary, our data suggest that community-acquired UTI caused by ESBL-producing *E. coli* is an endogenous infection in about half the patients; the prevalence of faecal carriage with ESBL-producing *E. coli* is more frequent in relatives of UTI patients than in unrelated persons; both person-to-person transmission and acquisition from a common source, probably related to food or food habits, may contribute to ESBL spreading within families.

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Transparency declarations

None to declare.

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