

Absence of CTX-M Enzymes but High Prevalence of Clones, Including Clone ST131, among Fecal *Escherichia coli* Isolates from Healthy Subjects Living in the Area of Paris, France[∇]

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Quinolone-resistant and CTX-M-15-producing *Escherichia coli* isolates belonging to clone ST131 have been reported in the community. This study was designed to identify these *E. coli* isolates in the stools of 332 independent healthy subjects living in the area of Paris, France. Stools were plated on media without antibiotics, in order to obtain the dominant (Dm) fecal *E. coli* strain, and with nalidixic acid (NAL) and cefotaxime. Quinolone susceptibility, phylogenetic groups, and molecular profiles, including multilocus sequence types (ST), were determined for all NAL-resistant (NAL-R) isolates. Groups were also determined for the Dm strains from participants with NAL-R isolates and from a subgroup without NAL-R isolates. All B2 isolates were typed; pulsed-field gel electrophoresis was performed for the ST131 isolates, and the results were compared with those for intercontinental clone ST131. Two participants (0.6%) had extended-spectrum β -lactamase-producing (SHV-2, TEM-52) fecal *E. coli* isolates, and 51 (15%) had NAL-R isolates; 51% of NAL-R isolates belonged to phylogenetic group A, 31% to group D, 16% to group B2, and 2% to group B1. The Dm strain was NAL-R in 3.3% of the 332 subjects. Forty-nine percent of the NAL-R isolates belonged to clones: ST10 and ST606 for group A isolates, ST117 and ST393 for group D isolates. Of all B2 isolates studied from 100 subjects (8 NAL-R strains; 19 NAL-susceptible dominant strains), 52% belonged to three clones: ST131 ($n = 7$), ST95 ($n = 4$), and ST141 ($n = 3$). This is the first study to show the presence of fecal *E. coli* isolates of clone ST131 in 7% of independent healthy subjects not colonized by CTX-M-15-producing isolates.

Escherichia coli, a universal commensal of humans and several animal species, is also one of the most common enterobacterial species to cause extraintestinal infections in their hosts (30). According to several recent publications, *E. coli* isolates producing extended-spectrum β -lactamases (ESBL) of the CTX-M type have emerged in the community in numerous countries (25–28). These isolates resist extended-spectrum cephalosporins because of CTX-M production and are often resistant to other antibiotic families, in particular fluoroquinolones and/or cotrimoxazole (28). This pattern of multidrug resistance is dangerous for the treatment of community-acquired infections, because these drugs are often prescribed by general practitioners, especially for urinary tract infections (UTIs). Moreover, a clonal group producing CTX-M-15 has been identified among fluoroquinolone-resistant and CTX-M-producing *E. coli* isolates (phylogenetic group B2, O25:H4, ST131) in both inpatients and outpatients all over the world, strongly suggesting that this clone can disseminate widely (20).

Because of these recent data and because the main reservoir of extraintestinal pathogenic *E. coli* isolates is the human digestive tract (8), the aim of this study was to

identify CTX-M-15-producing *E. coli* isolates and *E. coli* clone ST131 in the guts of healthy adult subjects living in the area of Paris, France.

MATERIALS AND METHODS

Participants. Fresh stool specimens were provided by 332 healthy adult volunteers who visited the Medical Center IPC (located in Paris) for a checkup between 14 and 27 February 2006. All participants provided informed consent, and the protocol was reviewed and approved by the relevant institutional review board of the medical center.

The physician who examined the participants collected demographic data (age, gender, weight, and address) and clinical data concerning hospital stays and antibiotic intake in the past month, as well as any occupational activity in a health care setting. Information about hospital stays and work in a health care setting was also obtained for people living with the participants, as well as information about any visits made to the IPC center at the same time or during the study period. Stools and participant files were transferred anonymously to the microbiological department of Hôpital Beaujon every day, and stools were stored at -80°C until use.

Sample processing and bacterial strains. Aliquots (50 mg) of each stool specimen were spread onto two Drigalski agar plates (Bio-Rad, Marnes la Coquette, France) supplemented with either 0.5 mg/liter of cefotaxime (CTX) or 20 mg/liter of nalidixic acid (NAL) and were incubated for 48 h at 37°C . Putative *E. coli* colonies were selected arbitrarily from each plate. Biochemical identification was performed by using the API 20E system (bioMérieux, Marcy l'Etoile, France).

The dominant (Dm) fecal *E. coli* strain was obtained from subjects with fecal *E. coli* isolates resistant (R) to NAL and from a randomly chosen subgroup of subjects without NAL-R isolates, as follows. An aliquot (50 mg) of stools was spread onto a Drigalski agar plate and incubated for 48 h at 37°C . From the terminal streak area of each plate, an isolated colony suspected to be *E. coli* was chosen. If multiple morphologies were noted, all

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TABLE 1. Demographic and clinical data for 329 healthy subjects according to NAL-R *E. coli* digestive carriage status

Parameter	Value for subjects:	
	With NAL-R <i>E. coli</i>	Without NAL-R <i>E. coli</i>
No. of subjects	51	278
Male	33	154
Female	18	124
Mean age (yr)	61	60
Male	62	58
Female	59	61
Mean wt (kg)	71	71
Male	76	79
Female	65	63
No. working in health care settings	1	2
No. with prior hospitalization	0	7
No. with prior antibiotherapy	5	23
No. of households with:		
Work in health care settings	0	5
Prior hospitalization	3	12

unique morphotypes were sampled. Each colony selected was identified using the API 20E system (bioMérieux). Since statistically a strain needs to be fairly prevalent within the fecal flora to be recovered from the terminal streak area, the colony selected was considered to represent the Dm *E. coli* strain (18).

Strain T1, a representative strain of clone ST131 (20), was used as a control strain for the enterobacterial repetitive intergenic consensus 2 (ERIC-2) PCR typing method.

ESBL detection and characterization. *E. coli* isolates from CTX-supplemented plates were tested for ESBL production by using the double-disk synergy test described by Jarlier et al. (13). The type of ESBL produced by *E. coli* isolates with a positive double-disk synergy test was determined by using *bla* gene-specific primers and the PCR conditions described previously (15).

Antibiotic susceptibility testing. *E. coli* isolates from NAL-supplemented plates were tested by disk diffusion for susceptibility to NAL and ciprofloxacin (CIP) by using the method and interpretative criteria recommended by the French AntibioGram Committee (1).

Phylogenetic classification. Phylogenetic groups were determined by a multiplex PCR assay (6).

ERIC-2 PCR typing. The genetic diversity of the NAL-R isolates within each phylogenetic group was determined by the ERIC-2 PCR typing method as previously described (20). This method was also applied to Dm strains when the NAL-R isolate and the Dm strain of a subject belonged to the same phylogenetic group, and it was also applied to all isolates found to belong to group B2 in this study.

MLST. The NAL-R isolates and all group B2 isolates with identical ERIC-2 PCR profiles upon visual comparison were further analyzed by multilocus sequence typing (MLST) as previously described (20).

Serotyping and PFGE typing of isolates identified as *E. coli* ST131 by MLST. O and H antigen determination was performed as previously described (24). XbaI PFGE analysis was performed as previously described (2). Profiles were compared digitally using BioNumerics software (Applied Maths). Cluster analysis of Dice similarity indices based on the unweighted-pair group method using average linkages (UPGMA) was used to generate a dendrogram describing the relationships among PFGE profiles. Isolates were considered to belong to the same PFGE group if their Dice similarity index was $\geq 85\%$ (5). The PFGE profiles of isolates identified as *E. coli* ST131 in the present study were compared with those displayed by the 36 intercontinental ST131 isolates producing CTX-M-15, and the Spanish ST648 strain FV7591, producing CTX-M-1, that we previously studied and published (20).

Statistical analysis. Continuous variables were compared by Fisher's exact test, while the *t* test was used to compare age and weight. A *P* value of ≤ 0.05 was considered statistically significant.

TABLE 2. Phylogenetic groups of the isolates of three *E. coli* populations

Population ^a	Number (%) of isolates in group:				Total no. of participants studied
	A	B1	B2	D	
NAL-R <i>E. coli</i>	26 (51)	1 (2)	8 (16)	16 (31)	51
Dm strains of subjects:					
With NAL-R <i>E. coli</i>	22 (43) ^b	6 (12)	5 (10)	18 (36) ^c	51
Without NAL-R <i>E. coli</i>	23 (47)	4 (8)	14 (28)	8 (16)	49
Total	71	11	27	42	100

^a The NAL-R *E. coli* population and the population of Dm strains of subjects with NAL-R *E. coli* come from the same 51 participants.

^b Seven of these isolates were NAL-R.

^c Four of these isolates were NAL-R.

RESULTS

Prevalence of ESBL-producing and quinolone-resistant fecal *E. coli* isolates. Fresh stools were obtained from 332 healthy adult subjects. Demographic data were available for 329. All subjects lived in houses or flats in the Paris area. None of the participants lived in the same house as any other participant in the study. There were 187 men and 142 women, with a mean age of 60 years (range, 24 to 88 years). Globally, 2% of the participants had been hospitalized and 8.5% had been given antibiotics within the month prior to stool sampling.

Two of the 332 subjects (0.6%) had fecal *E. coli* isolates from the CTX-supplemented plates that were positive by the double-disk synergy test. These two isolates harbored genes encoding ESBL SHV-2 and TEM-52, respectively.

Fecal *E. coli* isolates from 51 (15%) of the 332 subjects were selected on NAL-supplemented plates. Forty-nine percent of these NAL-R isolates were classified as resistant to CIP, resulting in a 7% prevalence of CIP-R fecal *E. coli* isolates among the 332 healthy subjects studied.

As shown in Table 1, the 51 subjects with NAL-R isolates did not differ from the subjects without NAL-R isolates with respect to demographic or clinical criteria, particularly hospital stays and antibiotic intake in the month before stool sampling.

Phylogenetic groups and ERIC-2 PCR typing of *E. coli* NAL-R isolates. The phylogenetic groups of the 51 NAL-R isolates were determined in order to know whether *E. coli* isolates of group B2, clone ST131, were present among the quinolone-resistant fecal isolates, even though these isolates did not produce CTX-M-15. Sixteen percent of these isolates belonged to group B2, whereas 51% belonged to group A, 31% to group D, and 2% to group B1 (Table 2). The eight NAL-R group B2 isolates were typed by the ERIC-2 PCR method and compared to strain T1, the representative strain of clone ST131 in this study. Four unique profiles were found in these eight isolates; two profiles (B2.I and B2.II) were each found in a single isolate, and two other profiles, B2a and B2b, were found in four and two isolates, respectively (Table 3). Moreover, profile B2a was absolutely identical to the profile of strain T1 (data not shown).

To ascertain whether clonal strains also existed among isolates from other phylogenetic groups, ERIC-2 PCR profiles were determined for isolates from groups A and D. As shown in Table 3, the 26 NAL-R group A isolates displayed 15 pro-

TABLE 3. Phylogenetic groups and ERIC-2 PCR and ST profiles of 51 NAL-R isolates

Participant code(s)	Phylogenetic group	ERIC-2 PCR profile	ST profile
105 ^a	A	A.I	
114	A	A.II	
115	A	A.III	
136	A	A.IV	
172 ^a	A	A.V	
175	A	A.VI	
228 ^a	A	A.VII	
247 ^b	A	A.VIII	
282	A	A.IX	
286	A	A.X	
289	A	A.XI	
291 ^a	A	A.XII	
312	A	A.XIII	
60, ^a 79, 17, 184, ^a 267	A	Aa	ST10
159, 272, 305, ^a 311	A	Aa	ST167
47	A	Aa	ST709
203	A	Aa	ST744
295, 296	A	Ab	ST606
224	B1	NA ^c	
36	B2	B2.I	
22	B2	B2.II ^d	
2, 183, 39, 187	B2	B2a	ST131
74, 190	B2	B2b	ST95
8	D	D.I	
12 ^a	D	D.II	
24	D	D.III	
34	D	D.IV	
53	D	D.V	
84	D	D.VI	
90	D	D.VII	
151 ^a	D	D.VIII	
156 ^a	D	D.IX	
327	D	D.X	
11, ^a 208, 325	D	Da	ST117
223, 313, 314	D	Db	ST393

^a The NAL-R isolate was the Dm fecal *E. coli* strain in the subject.

^b Subject carrying the SHV-2-producing isolate.

^c NA, not available.

^d This ERIC-2 PCR profile was also found in two Dm susceptible group B2 strains.

files, of which 13 (A.I to A.XIII) were specific for 1 isolate each, 1 (Aa) was specific for 11 isolates, and 1 (Ab) was specific for 2 isolates. The 16 NAL-R group D isolates displayed 12 profiles, of which 2, Da and Db, were found in 3 isolates each and 10 (D.I to D.X) were each specific for 1 isolate (Table 3). Thus, ERIC-2 PCR showed that 25 (49%) of the 51 NAL-R fecal *E. coli* isolates identified in the 332 healthy subjects were clonal strains.

MLST of the NAL-R isolates identified as clones by ERIC-2 PCR typing. MLST was used to confirm the clonal relatedness of the NAL-R isolates with identical ERIC-2 PCR profiles and to characterize the different clones. Identical sequence types (ST) were found for the group B2 isolates: ST131 for the four isolates with the B2a profile and ST95 for the two isolates with the B2b profile (Table 3). The 11 group A isolates with the ERIC-2 PCR profile Aa showed four ST: ST10, ST167, ST709, and ST744 (Table 3). However, since they differed from each other by a single allelic sequence, these four ST belonged to clone complex ST10. The two group A isolates with the Ab profile by ERIC-2 PCR had identical ST (ST606). The three

TABLE 4. Distribution of group B2 isolates into different clones according to their susceptibility to NAL

Strain	No. of isolates in clone:			Total no. of:	
	B2a (ST131)	B2b (ST95)	B2c (ST141)	Clonal isolates	Group B2 isolates
NAL-R	4	2	1	7	8
NAL-S Dm	3	2	2	7	19
Total	7	4	3	14	27

group D isolates with the Da profile by ERIC-2 PCR displayed ST117, whereas those with the Db profile displayed ST393 (Table 3). Overall, the MLST method confirmed the ERIC-2 PCR-based clonal relatedness of 49% of the NAL-R isolates detected in 51 independent healthy subjects and characterized nine clones, including clone ST131 (Table 3).

Rates of NAL-R isolates and NAL-R clonal strains among the Dm strains. The phylogenetic groups of the Dm strains of the 51 participants with NAL-R isolates were determined. When the Dm strain and the NAL-R isolate in a given participant belonged to the same phylogenetic group, their ERIC-2 PCR profiles were determined. Thus, the phylogenetic group distribution of the Dm strains of subjects with NAL-R fecal isolates did not differ significantly from that of the NAL-R isolates (Table 2). In 11 (21.5%) cases, the NAL-R isolate and the Dm strain belonged to the same group and had identical ERIC-2 PCR profiles. These cases included seven group A and four group D isolates, four (36%) of which were clonal strains (two ST10, one ST167, and one ST117 strain). As a result, the NAL-R isolates were more often subdominant ($n = 40$) than Dm ($n = 11$) strains. Five (45%) of the 11 Dm NAL-R strains were CIP-R.

Overall, 11 (3.3%) of the 332 subjects in the study had a NAL-R Dm strain; 4 had (1.2%) a clonal NAL-R Dm strain; 5 (1.5%) had a CIP-R Dm strain; and none had a NAL-R group B2 Dm strain.

Identifying clones in Dm strains of group B2. Table 2 shows that only 10% ($n = 5$) of the Dm strains of the 51 participants with NAL-R isolates belonged to group B2 and that all five of these strains were susceptible (S) to NAL. The Dm strains of a subgroup of 49 randomly chosen participants without NAL-R isolates were studied in order to determine (i) whether the rate of group B2 isolates in participants without NAL-R isolates was different from that in participants with NAL-R isolates and (ii) whether clones existed among the NAL-S group B2 Dm strains. Analysis of the phylogenetic group distribution of the Dm strains of participants without NAL-R isolates (Table 2) showed that it differed, but not significantly, from that of the Dm strains of participants with NAL-R isolates. The 14 NAL-S group B2 Dm strains of participants without NAL-R isolates plus the 5 NAL-S group B2 Dm strains of participants with NAL-R isolates were analyzed by the ERIC-2 PCR and MLST methods and compared to the 8 NAL-R group B2 subdominant isolates (Table 4). Profiles B2a and B2b, identified in four and two subdominant NAL-R isolates (Table 3), respectively, were also found in three and two NAL-S Dm strains, respectively (Table 4). As expected, Dm strains with profile B2a had ST131 and those with profile B2b had ST95. A new clonal B2 strain

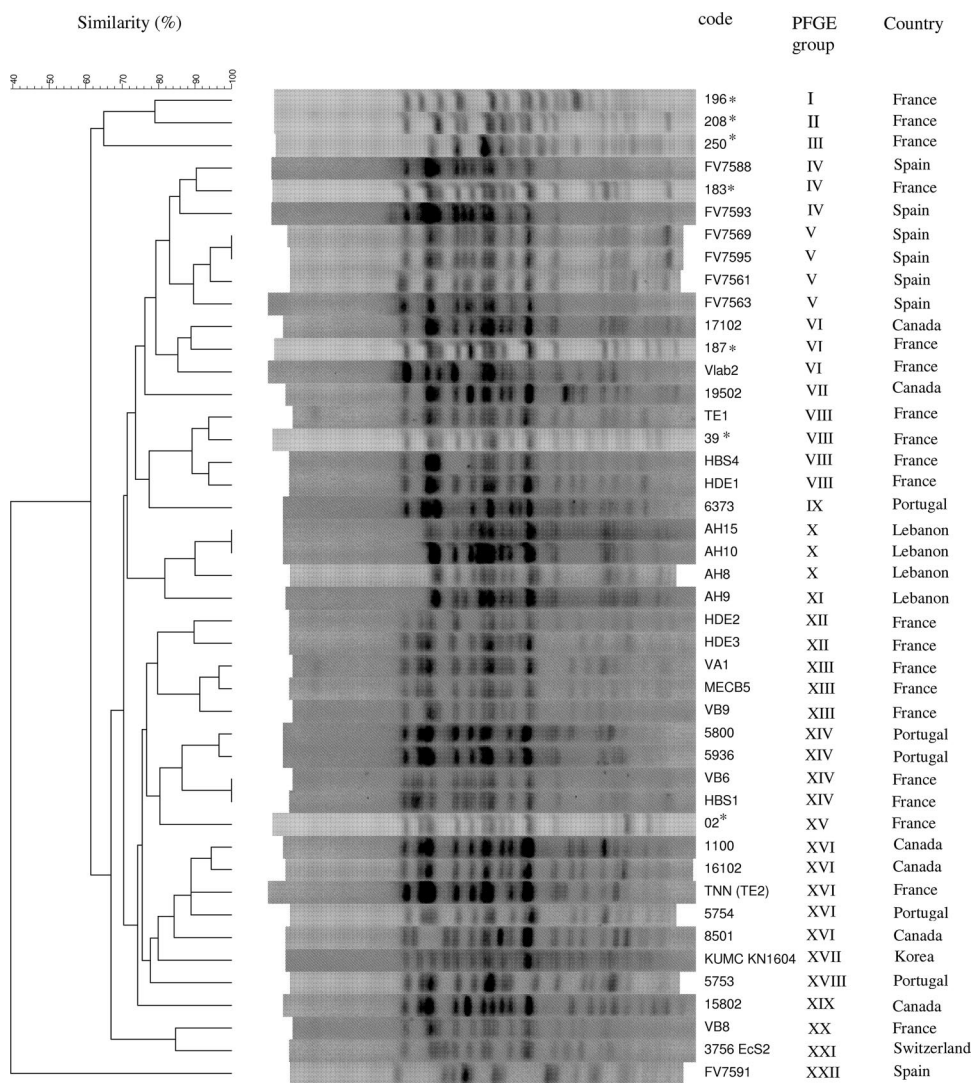


FIG. 1. XbaI PFGE dendrogram of 43 *Escherichia coli* ST131 isolates and a Spanish ST648 strain. The 7 ST131 fecal isolates analyzed in the present study (asterisked) were compared with 36 intercontinental ST131 isolates and the Spanish ST648 strain, which were published previously (24). The dendrogram for the 44 isolates, produced by the UPGMA algorithm based on Dice similarity coefficients, included 22 PFGE groups defined on the basis of $\geq 85\%$ similarity of PFGE profiles.

(B2c) was identified by comparing the ERIC-2 PCR profiles of the 8 NAL-R and 19 NAL-S isolates. The NAL-R isolate of subject 22 (profile B2.II) (Table 3) and two NAL-S Dm strains (Table 4) belong to this clone, whose ST was shown to be ST141.

Overall (Table 4), 7 (37%) of the 19 NAL-S Dm strains belonging to group B2 (from the group of 100 healthy subjects whose Dm strains were studied) were clonal strains. In contrast, clonal strains constituted 87% (7/8) of the group B2 isolates among the NAL-R subdominant isolates detected in 51 of the 100 healthy subjects studied. Thus, 14 (52%) of the 27 group B2 fecal isolates studied were clonal strains. Fifty percent of the B2 clonal strains belonged to clone ST131 (Table 4), identified in 7 of the 100 healthy subjects.

Serotyping and PFGE of seven ST131 isolates. The seven B2 isolates belonging to ST131 (Table 4) were serotyped and shown to belong to serotype O25:H4. Their XbaI PFGE profiles were compared to those of 36 intercontinental CTX-M-15-producing

E. coli clinical isolates previously shown to belong to clone ST131 (20). Figure 1 shows the dendrogram generated from the PFGE profiles of the 43 ST131 isolates. Overall, 21 PFGE groups with a similarity index of $\geq 85\%$ were identified. Four of the seven isolates (Dm strains from subjects 196, 208, and 250 and the NAL-R strain from subject 02) each belonged to a unique PFGE group. In contrast, the three remaining NAL-R isolates belonged to three different PFGE groups, which included other, previously described ST131 isolates (Fig. 1). Thus, the NAL-R isolate of subject 183 was in the same PFGE group as two Spanish strains; that of subject 187 was in the same PFGE group as a Canadian and a French strain; and that of subject 39 was in the same PFGE group as three French strains.

Overall, the 43 ST131 strains constituted one large cluster (defined by a 62% similarity level), which was associated with the “outgroup” ST648 strain FV7591 (PFGE profile XXII) by $< 40\%$ similarity (Fig. 1).

DISCUSSION

Recent studies have shown a significant increase in the number of CTX-M-producing *E. coli* isolates, including CTX-M-15-producing *E. coli* clone ST131, in the community (19, 22, 25, 26, 28). Assessment of this increase has been based on the increase in the number of CTX-M-positive clinical samples obtained from outpatients and inpatients at hospital admission. However, appropriate studies to effectively measure the prevalence of CTX-M-positive *E. coli* isolates in the community—for example, prospective studies evaluating the fecal carriage of ESBL-producing *E. coli* isolates in healthy volunteers without any recent contact with health care settings and without recent exposure to antibiotics—remain scarce. One study performed in 2005 by Palecchi et al. on healthy children in South America reported an ESBL-producing fecal *E. coli* prevalence of 1.7%, with a predominance of CTX-M enzymes (23). A second study, by Valverde et al., in 2003, reported a higher prevalence of ESBL-positive *E. coli* isolates (3.7%) but a lower proportion of CTX-M enzymes (50% CTX-M and 50% SHV-12) in 108 independent healthy Spanish (Madrid) volunteers (32). Despite the apparent similarity of our subjects to those of Valverde et al., our results were quite different: 0.6% of our 332 healthy subjects had *E. coli* fecal isolates producing ESBL that were not CTX-M enzymes. The difference in these results may seem surprising, since both studies were performed several years after the emergence and dissemination of CTX-M enzymes in the respective countries (9, 15, 21, 29), and both were performed on healthy subjects. However, there are no data on the risk factors of fecal carriage of CTX-M enzymes in healthy adult subjects, living in large urban areas of developed countries, who have had no recent contact with health care settings and no recent exposure to antibiotics. Thus, it is difficult to explain the conflicting results between the Valverde study and ours. No information was given on the mean age of the 108 healthy participants in the former study. In that study, the four subjects with ESBL-producing *E. coli* isolates were 23 to 25 years old, so our population seems to have been older (mean age, 60). If age affects the prevalence of CTX-M-positive fecal *E. coli* isolates, the difference between the Spanish and French studies could be related to this factor. Moreover, no details were given about how the healthy Spanish volunteers were chosen and their potential relationships. Our 332 subjects belonged to distinct households, and most probably had no relationship with each other, because their residences extended over an area of 12,000 km² with 11,500,000 inhabitants. If social and geographical links between healthy subjects have an impact on the prevalence of CTX-M-positive fecal *E. coli* isolates, more data about these parameters in the Spanish study would be interesting.

The source of CTX-M-producing fecal isolates in independent healthy subjects without direct or indirect (via households) contact with any health care setting could be food. Although food-producing animals have been shown to be infected or colonized by *E. coli* strains producing CTX-M-1 and -15 in France (11, 17), food-borne CTX-M enzymes do not seem to be a significant health concern, since none of our independent healthy subjects was found to be positive for CTX-M enzymes in fecal *E. coli* isolates.

In contrast, food could be a significant source of human fecal

NAL-R *E. coli* strains, as previously suggested (14), since 15% of our independent healthy subjects harbored such strains in the gut. Once again, comparing our results to previous results is difficult, because the populations studied are often not comparable. Thus, Garau et al. found a 24% prevalence of NAL-R fecal *E. coli* in 104 Spanish (Barcelona) adults resembling our participants except that they were visiting the hospital emergency room for a noninfectious disease when they were included in the study (10). Bruinsma et al. found a prevalence of 1% to 12% depending on the country (Canada, Greece, or The Netherlands) where healthy volunteers in this study lived (4), while Grenet et al. found a prevalence of 8% in the Wayampis Amerindians, an isolated community in French Guyana (12). Globally, our results are similar to those previously published. However, we showed that only 20% of the NAL-R fecal isolates were Dm strains. Distinguishing between Dm and subdominant fecal strains seems to be relevant with regard to *E. coli* extraintestinal pathogenesis. Indeed, Moreno et al. showed that the prevalence of fecal isolates, notably those less virulent, is an important determinant for UTI pathogenesis in women (18). This finding suggests that the risk of having a UTI caused by an antibiotic-resistant *E. coli* isolate could be higher in subjects in whom the Dm strain is resistant to antibiotics than in subjects in whom the antibiotic-resistant isolates are subdominant.

Another important result was the presence of clonal fecal *E. coli* isolates in independent healthy subjects. Almost 50% of the NAL-R isolates were clonal strains, and the clonal structure was independent of the phylogenetic groups. The use of the MLST method made it possible to characterize these clones ($n = 9$) and identify the ST131 clone (group B2), which had been reported, until this study, for isolates that produced the ESBL CTX-M-15 and were resistant to quinolones (20). The discovery of clone ST131 in NAL-R isolates was followed by the discovery of this clone in NAL-S Dm strains. All these results showed that clone ST131 was the dominant clone in the isolates of group B2.

The presence of fecal *E. coli* clones in apparently epidemiologically unrelated subjects has already been suggested by profile comparison methods (16, 24). The present study clearly identifies the clones (ST) that were suggested by ERIC-2 PCR, which we have confirmed to be an accurate method of detecting clone complexes (31). Thus, clone ST69 (also called CgA), which is widely disseminated in North America (3, 16, 31), was shown to be absent in our healthy subjects by MLST, while clone ST131, which is present on three continents in the form of isolates producing CTX-M-15 (20), was present in the form of isolates free of CTX-M enzymes and S or R to quinolones in 7% of healthy subjects.

PGFE typing showed that the seven ST131 isolates of this study formed a PFGE-based cluster (defined at the 62% similarity level) with the 36 previously published international ST131 isolates (20). This strongly suggests recent divergence from a common ancestor. Furthermore, the marked similarity of the PFGE profiles of some of our fecal *E. coli* isolates with those of certain clinical isolates producing CTX-M-15 from either France, Spain, or Canada strongly suggests a recent acquisition of CTX-M-15-mediating-plasmids by clone ST131. Although very few studies on the intercontinental clone ST131 are available, it has been shown to be a virulent clone that

produces biofilms (7, 20). Finding this clone in the guts of independent healthy subjects at a prevalence of 7% could indicate that it has particular, as yet undefined properties.

In conclusion, this study showed that CTX-M-producing fecal isolates were absent, while quinolone-resistant subdominant fecal isolates were highly prevalent, in healthy adults living in the Paris area. We also found that 50% of the NAL-R fecal isolates and 50% of the group B2 fecal isolates were clonal isolates. Finally, this is the first study to show the presence of fecal *E. coli* isolates of clone ST131 that do not produce CTX-M-15 in 7% of independent healthy subjects.

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