Predominance of the *fimH30* Subclone Among Multidrug-Resistant *Escherichia coli* Strains Belonging to Sequence Type 131 in Italy

To the Editor—We read with interest the article by Johnson et al, which demonstrates that, in extraintestinal pathogenic *Escherichia coli* (ExPEC), fluoroquinolone (FQ) resistance is currently associated with the rapid expansion of a single dominant multidrug-resistant (MDR) strain that emerged within sequence type (ST) 131 [1]. Although the strong predominance of the ST131 clone has been well described in the literature since 2008 [2–5], the peculiarity of Johnson et al's study was to analyze historical and recent ST131 isolates at the sub-ST level, revealing that a specific *fimH*-based subclone (*H30*) is currently responsible for most FQ-resistant ExPEC infections, at least in the United States. This *H30* subclone was demonstrated to possess a unique and conserved *gyrA*/*parC* allele combination conferring FQ resistance, and the authors rightly commented on these data in support of its strict clonality. We are particularly interested in this issue, since ST131 is also currently predominant among MDR ExPEC in Italy and because some ST131 strains we previously analyzed were found to carry the same pattern of *gyrA*/*parC* substitutions described by Johnson et al [1, 6]. No information is available on the *fimH*-based subclones circulating in European countries.

In this study, 172 ExPEC strains isolated from cases of urinary tract infections and sepsis in Italy during April 2012–December 2012 were analyzed. Staff a the 3 enrolled hospitals were asked to collect all consecutive MDR *E. coli* strains and 1 ciprofloxacin-susceptible non-MDR *E. coli* strain for every 3 MDR strains collected. "MDR" was defined as resistance to at least 3 antimicrobial agents of different classes (ampicillin, third-generation cephalosporins, ciprofloxacin, gentamicin, and trimethoprim/sulfamethoxazole). Of 172 strains, 119 were MDR strains that were resistant to ciprofloxacin; 9 were MDR strains that were susceptible to ciprofloxacin; and 44 were non-MDR strains that were susceptible to ciprofloxacin. Antimicrobial susceptibility testing, phylogenetic typing, polymerase chain reaction screening for ST131 followed by confirmation (by *mdh* and *gyrB* gene sequencing), characterization of the extended-spectrum β-lactamase (ESBL) and/or AmpC genes, and *fimH*-based subtyping of the ST131 strains were performed as previously described [1, 7–9]. To investigate which *fimH* allele circulated in Italy before 2012, an additional 91 *E. coli* ST131 strains previously identified were also subtyped [7, 10]. Of these 91 strains, 28 (24 ciprofloxacin-resistant strains and 4 ciprofloxacin-susceptible strains) were isolated in 2006, and 63 (58 ciprofloxacin-resistant strains and 4 ciprofloxacin-susceptible strains) were isolated in 2009.

By phylogenetic typing, overall, the majority of strains isolated in 2012 (110/172 [64%]) fell into phylogenetic group B2, followed by groups D (30/172 [17.4%]),

Table 1. Distribution of Extended-Spectrum β-Lactamase (ESBL)/AmpC types and *fimH* Alleles Among Sequence Type 131 *Escherichia coli* Strains Stratified by Ciprofloxacin (CIP) Susceptibility and Multidrug Resistance (MDR) Status

	MDR Strains, No. (%)		Non-MDR Strains, No. (%)
	CIP Resistant (n = 73)	CIP Susceptible (n = 4)	CIP Susceptible (n = 6)
ESBL type			
CTX-M-15	64 (87.7)	3 (75.0)	. . .
CTX-M-1	. . .	1 (25.0)	. . .
CTX-M-55	1 (1.4)	. . .	. . .
CTX-M-14	1 (1.4)	. . .	1 (16.7)
AmpC type			
CMY-2	. . .	. . .	1 (16.7)
fimH allele			
H30	73 (100)	3 (75.0)	5 (83.3)
H22	. . .	1 (25.0)	1 (16.7)

A (17/172 [9.9%]), and B1 (13/172 [7.6%]); 2 strains were nontypeable. Most phylogroup B2 strains (83/110 [75.5%]) were ST131. ST131 accounted for 61.3% (73/119), 44.4% (4/9), and 13.6% (6/44) of all MDR ciprofloxacin-resistant, MDR ciprofloxacin-susceptible, and non-MDR ciprofloxacin-susceptible strains, respectively. Of the 83 ST131 strains, 72 (86.7%) showed an ESBL phenotype. Most ESBL-positive strains (71/72 [98.6%]) were CTX-M positive: 67 strains contained $bla_{\text{CTX-M-15}}$, 1 contained $bla_{\text{CTX-M-1}}$, 1 contained $bla_{\text{CTX-M-55}}$, and 2 contained $bla_{\text{CTX-M-14}}$ (Table 1). One strain carried bla_{CMY2}. When the 83 ST131 strains were subtyped by *fimH* sequencing, all but 2 were found to belong to the *H30* subclone, including 3 MDR ciprofloxacin-susceptible strains and 5 non-MDR ciprofloxacin-susceptible strains (Table 1). Both remaining strains were *H22* (one was MDR but ciprofloxacin-susceptible strain, and the other was a ciprofloxacin-susceptible non-MDR strain; Table 1). Finally, the *fimH* subtyping of the 91 ST131 strains collected in previous periods showed that the *H30* subclone has predominated since at least 2006. In fact, 26 of 28 strains and 62 of 63 strains isolated in 2006 and 2009, respectively, belonged to *H30*. The remaining 3 strains (all ciprofloxacin-susceptible non-MDR strains) were *H22*.

Our data confirmed that most FQ-resistant MDR strains carrying CTX-M15 ESBL are also currently associated with the dominant *H30* ST131 subclone in Italy, indicating a worldwide dissemination of this subclone. According to the study by Johnson et al, *H30* expanded abruptly after 2000 together with the emergence of FQ resistance, whereas *H22* predominated before then and was associated with FQ susceptibility in both historical and current isolates [1]. In our study, the *H22* subclone was rare even among FQ-susceptible strains that mostly belonged to *H30*, although the few *H22* strains we found were all FQ susceptible. It is possible that the epidemiology of the ST131 subclones slightly differed in Italy and that *H30* was present before acquiring FQ resistance. Unfortunately, we were not able to analyze ExPEC isolates collected before 2006. Therefore we cannot investigate the emergence of the different ST131 subclones according to FQ susceptibility status.

In conclusion, most MDR ExPEC infections are currently due to the *H30* ST131 subclone in Italy. We emphasize the importance of performing investigations on the clonal structure of the "successful" ST131 clone in different countries and settings.

Notes

Acknowledgments. We thank Tonino Sofia, for editorial assistance; and Alessandra Carattoli (Istituto Superiore di Sanità), for helpful discussion and comments.

Financial support. This work was supported by the Ministry of Health–Centro Controllo Malattie (project title, "Surveillance on Antibiotic Resistance in the Community and in Foodborne and Zoonotic Diseases"; grant 3M24).

Potential conflicts of interest. All authors: No reported conflicts.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

Maria Giufre,[1,a] Marisa Accogli,[1,a] Claudio Farina,[2] Anna Giammanco,[3] Patrizia Pecile,[4] and Marina Cerquetti[1]

[1]Department of Infectious, Parasitic and Immune-mediated Diseases, Istituto Superiore di Sanità, Rome; [2]Microbiology and Virology Institute, Azienda Ospedaliera Papa Giovanni XXIII, Bergamo; [3]Department of Sciences for Health Promotion and Mother and Child Care G. D'Alessandro, University of Palermo, Palermo; and [4]Microbiology Laboratory, Azienda Ospedaliero–Universitaria Careggi, Florence, Italy

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Received 31 July 2013; accepted 17 September 2013.

[a]M. G. and M. A. contributed equally to this work.

Correspondence: Dr. Marina Cerquetti, Department of Infectious, Parasitic and Immune-mediated Diseases, Istituto Superiore di Sanità, Viale Regina Elena, 299, 00161 Rome, Italy (marina.cerquetti@iss.it).

The Journal of Infectious Diseases 2014;209:629–30

DOI: 10.1093/infdis/jit583