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BRIEF REPORT

Pandemic Uropathogenic Fluoroquinolone-resistant *Escherichia coli* Have Enhanced Ability to Persist in the Gut and Cause Bacteriuria in Healthy Women

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We report that fluoroquinolone-resistant *Escherichia coli* are found in feces of 8.8% of healthy women, with most bacteria belonging to pandemic multidrug-resistant ST131-*H*30R or ST1193 clonal groups. Moreover, these highly uropathogenic clonal groups demonstrate an especially prolonged gut persistence and high rate of bacteriuria without documented urinary tract infection.

Keywords. gut microflora; urinary tract infections; *Escherichia coli*; fluoroquinolone resistance; bacteriuria.

Urinary tract infections (UTIs) are mostly caused by *Escherichia coli* [1]. Fluoroquinolones (FQs) are among the most often prescribed antibiotics for UTI treatment, despite repeated calls to limit their use and a rampant rise of FQ-resistant (FQR) *E. coli* [2]. Strains from 2 multidrug-resistant clonal groups are dominant among UTI-associated FQR *E. coli*: ST131-*H*30R that emerged in the late 1990s and the more recently discovered ST1193 [3–6]. Both ST131-*H*30R and ST1193 are now pandemic and comprise 60–80% of FQR *E. coli* isolated in the United States from patients with suspected UTIs [6]. While it has been established that patients' gut microflora often serves as a reservoir for UTI-causing *E. coli* strains [7–9], it remains unknown whether the pandemic strains have distinct patterns of gut carriage or bacteriuria in healthy individuals.

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METHODS

This study on the collection and analysis of fecal and urine samples from women without documented UTI was approved by the Kaiser Permanente Washington (KPWA) Research Institute Institutional Review Board and was carried out between July 2015 and December 2016. Details are described in the Supplementary Data.

RESULTS

First, 1031 fecal samples were provided by women with no UTI documented within the last 12 months. *Escherichia coli* was identified in 916 (88.8%) samples and, of those, 91 samples (8.8% of the total and 9.9% of the *E. coli* samples) yielded FQR isolates (Table 1). Non–*E. coli* FQR bacteria were found in 17 (1.6% total) samples. Among FQR *E. coli*, 14 distinct clonal types (clonotypes) were identified, with 86 (94.5%) samples containing a single clonotype. Clonotypes corresponding to *E. coli* ST131-*H*30R and ST1193 were the most predominant, found in about half and one-quarter of the samples, respectively (Table 1). The remaining 12 clonotypes of FQR *E. coli* were found in 1 (1.1%) to 6 (6.6%) samples each (2.9% ± 1.7%, on average).

The average age of participants was 52 ± 17 years (range, 18–89 years). Among all FQR clonotypes only ST131-*H*30R displayed differential age distribution, being more prevalent among participants aged 60 years or older (odds ratio [OR], 2.0; 95% confidence interval [CI], 1.1–3.8; $P = .021$) (Table 1).

A total of 74 fecal FQR *E. coli* carriers (33 carriers of ST131-*H*30R, 17 of ST1193, 23 of other FQR *E. coli*, and 1 of both ST131-*H*30R and other FQR) provided follow-up urine samples, with 26 samples (16 from ST131-*H*30R carriers, 5 ST1193, and 5 other FQR) being positive for *E. coli* growth of 10^5 cfu or more (see Supplementary Table 1). In 24 (92.3%) urine samples positive for the *E. coli* bacteriuria irrespective of the clonal identity, only *E. coli* was isolated in significant numbers (Supplementary Table 1), indicating that fecal contamination of the urine samples was unlikely. Also irrespective of the clonal identity, the *E. coli*–positive and –negative urine samples did not significantly differ in the time elapsed between collection of the fecal and urine samples (on average, 145 ± 14 and 138 ± 8 days, respectively). Out of 26 *E. coli*–positive urine samples from the FQR carriers, in 20 samples (76.9%) FQR bacteria were present, all matching the same clonotype as in the original fecal sample.

A total of 45 out of 74 participants who supplied the urine sample also consented to electronic medical records (EMR) examination for the occurrence of UTI within 3 months of the urine collection. The EMR analysis revealed episodes of

Table 1. Comparison of Fecal Fluoroquinolone-resistant *Escherichia coli* Clones *H*30R and ST1193 With Other FQR *E. coli* Clones

Characteristic	All FQR	FQR clones		
		*H*30R	ST1193	Other FQR
Prevalence among all fecal samples (n = 1031)	91 (8.8)	44 (4.3)	20 (1.9)	29 (2.8)[a]
Prevalence among 18–60-year-olds (n = 641)[b]	51 (8.0)	20 (3.1)	12 (1.9)	17 (2.7)
Prevalence among ≥60-year-olds (n = 390)[b]	43 (11.0)*	24(6.2)**	8(2.1)[ns]	12(3.1)[ns]
FQR *E. coli* bacteriuria (consented participants without UTI)[c]	13/42 (30.9)	8/19 (42.1)**	4/9 (44.4)*	1/14 (7.1)[ref]
Fecal persistence[c]	47/78 (60.2)	28/37 (75.7)***	12/17 (70.6)***	7/29 (24.1)[ref]

P < .05 is considered significant for the purpose of this study. *P* values are indicated as **P* < .1, ***P* ≤ .05, ****P* ≤ .01, and [ns]*P* > .1 and [ref] for the clonal group used as reference in statistical analysis. Number of samples with FQR *E. coli* are reported with percent from total number of samples in the group given in parentheses [n (%)].

Abbreviations: FQR, fluoroquinolone-resistant; ns, not significant; UTI, urinary tract infection.

[a]For prevalence among fecal samples, 32 "Other FQR" clones were obtained from 29 samples, with 3 samples yielding 2 distinct nonpandemic clones each and 3 samples yielding 1 nonpandemic and 1 ST131-*H*30 clone each. For bacteriuria, 24 urine samples were obtained from participants whose initial fecal samples yielded 27 nonpandemic FQR *E. coli* clones, with 3 samples yielding 2 different nonpandemic clones each and 1 sample yielding 1 nonpandemic clone and 1 ST131-*H*30R clone. This results in an apparent mismatch between the total of 74 urine samples and the summarized 75 samples (34 fecal samples with ST131-*H*30R, 17 with ST1193, and 24 with other FQR clones).

[b]Difference in prevalence of each FQR clonal group among 2 age categories is compared in multiple logistic regression using the non-FQR samples as a reference.

[c]Difference in bacteriuria and fecal persistence is evaluated in multiple logistic regression with the "Other FQR" clones group as a reference.

documented UTI in 3 (6.7%) of the consented participants (Supplementary Table 1) who were removed from further analysis. Analysis of the FQR bacteriuria rates in 42 participants with no documented UTI revealed that the ST131-*H*30R and ST1193 bacteriuria rates among the corresponding fecal carriers were similar to one another but exceeded by multifold the bacteriuria rate of other FQR bacteria (Table 1).

The combined rate of ST131-*H*30R and ST1193 bacteriuria among the corresponding carriers with no documented UTI (12 of 28, 42.9%) was significantly higher than the combined bacteriuria rate among the fecal carriers of any other *E. coli* clones with no documented UTI. Indeed, among the women having only FQ-susceptible (FQS) *E. coli* in their original fecal sample, a total of 85 urine samples were received, with 62 of the participants consenting to the EMR examination. Among the latter, none had a documented UTI episode, but 12 participants (19.4%) had *E. coli* bacteriuria (all FQS), which was similar to the rate of any bacteriuria (FQR or FQS) of the carriers of nonpandemic FQR *E. coli* with no documented UTI (3 of 14, 21.4% of the urine samples). Thus, the combined bacteriuria rate of the carriers of other *E. coli* (15 of 76, 19.7%) was twice as low than that for the pandemic *E. coli* (OR, 2.9; *P* = .024).

A total of 78 participants who carried FQR bacteria in the original fecal sample supplied a second fecal sample. Almost two-thirds of repeated samples (60.2%) contained FQR bacteria of clonal groups matching those in the original fecal sample (Table 1). In 2 samples, ST131-*H*30R replaced the original FQR strain of a nonpandemic clonal origin. Like bacteriuria, the presence or absence of persistent fecal FQR *E. coli* did not depend on the time elapsed between collections of fecal samples, which was 250 ± 8 and 238 ± 9 days, respectively, indicating that heterogeneity in the sample collection time cannot explain the difference in *E. coli* persistence in the gut. Similar to the urine samples, original ST131-*H*30R and ST1193 clones were found significantly more frequently in the repeated fecal sample compared with other FQR *E. coli* combined (ORs, 9.8 and 7.5; *P* < .001 and .003, respectively; Table 1).

Among 89 participants who both provided a second fecal sample and gave consent to review their medical records, 26 were prescribed an antibiotic for any reason within the study period (Supplementary Table 1). In most cases, the drug prescribed was amoxicillin or azithromycin (usually for respiratory infection). Fluoroquinolones were prescribed to 5 participants. There was no significant difference in prescription of any or specific antibiotics between participants with persistent FQR *E. coli* carriage (16 of 29, 55.2%) and those who lost FQR *E. coli* in the second fecal sample (12 of 20, 60.0%; *P* = .73).

A total of 151 carriers of only FQS *E. coli* in the original fecal sample had supplied a repeated sample, with 7 (4.6%) samples yielding newly acquired FQR bacteria, among which 3 were ST131-*H*30R and 2 were ST1193. For the participants with available records, none of those with newly acquired FQR *E. coli* were prescribed any antibiotics during the study period.

Out of 67 participants with FQR bacteria in the original fecal sample who supplied both urine and second fecal samples, there was a strong association between the bacteriuria and fecal persistence (OR, 8.3; 95% CI, 1.5–22.2; *P* = .011) (Table 1).

DISCUSSION

The relative clonal distribution of fecal FQR *E. coli* among healthy carriers and specific age groups strikingly mirrored that in patients with UTI reported within a similar time period in the same patient community (ie, KPWA enrollees [6, 10]). Specifically, in both studies, the pandemic *E. coli* ST131-*H*30R comprised slightly less than half of FQR *E. coli*, with the pandemic ST1193 being about half of ST131-*H*30R and the rest of FQR strains being split among numerous minor clonotypes. Moreover and strikingly similar to UTI, ST131-*H*30R gut carriage was associated with older age. These observations support

previous studies showing that UTI strains of *E. coli* primarily originate from the hosts' own fecal microflora [7–9].

Both pandemic FQR clonotypes of *E. coli* are not only commonly carried in the gut of healthy individuals but their carriage is much more persistent than that of other FQR *E. coli*. Gut carriage of ST131-*H*30R has been reported previously [11, 12]. However, the superior gut persistence of both pandemic strains has not been known and may contribute to the remarkable global spread of ST131-*H*30R and ST1193 and their dominance among FQR *E. coli* of UTI origin. Importantly, antibiotic prescription was not associated with either gut persistence or acquisition of FQR *E. coli*, indicating that the FQ and multidrug-resistant bacteria can be sustained in the gut and spread between healthy individuals even in the absence of antibiotic use.

It is also notable that, relative to other *E. coli* in general (nonpandemic FQR or FQS), ST131-*H*30R and ST1193 were isolated about twice as frequently from the urine of the gut carriers. While we cannot affirm with absolute confidence that the bacteriuria with no documented UTI was truly asymptomatic, the pandemic clones clearly demonstrate a superior capability to invade the urinary tract, which is linked to the superior gut persistence. It is possible that the increased gut and bladder persistence could be due to somewhat similar molecular mechanisms involved. However, it is also intriguing to consider that bacteriuria may contribute to the gut persistence of *E. coli* by, for example, providing a "safe haven" niche protected from the gut bacteriophages or microbiota.

In summary, our study highlights the likely physiological basis of the recent pandemic spread of the FQR *E. coli*, the potential value of identifying the gut carriers to predict resistant infections, and the need to reassess the clinical significance of asymptomatic bacteriuria at the time of this recent pandemic of highly uropathogenic and multidrug-resistant *E. coli*.

Supplementary Data

Supplementary materials are available at *Clinical Infectious Diseases* online. Consisting of data provided by the authors to benefit the reader, the posted materials are not copyedited and are the sole responsibility of the authors, so questions or comments should be addressed to the corresponding author.

Notes

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Potential conflicts of interest. E. V. S. and V. L. T. have patent applications to detect *E. coli* strains, E. V. S. is a major shareholder in ID Genomics, V. L. T. is Scientific Director at ID Genomics, and E. R. is Director of Lab Operations at ID Genomics. All other authors report no potential 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.

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